

# Synthetic pyrethroids (Type II) and freshwater fish culture: Perils and mitigations

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**Abstract** As a new class of agricultural insecticides, synthetic pyrethroids are widely used to control insect pests. Synthetic pyrethroids have been shown to enter the aquatic environment from agricultural runoff or drift from aerial and ground-based spraying applications posing threat to fishes which are less tolerant to pesticides through direct exposure. These insecticides interfere with the sodium channel of the nervous system resulting in prolonged sodium tail current. Widespread application of these chemicals has warranted the attention of the ecologist to understand the impact of these chemicals on the aquatic environment. In this perspective, an updated account of toxicological evaluation of three type II synthetic pyrethroids, viz. deltamethrin, cypermethrin and fenvalerate in terms of their physico-chemical, metabolic, hematological, histological, behavioral and reproductive aspects with respect to the fishes has been presented which may be useful for policy makers, academics, environmental scientists and agricultural professionals needing ready access to this information. The aim of the present synoptic literature appraisal was to summarize the main effect of current use, type II

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synthetic pyrethroids (deltamethrin, cypermethrin and fenvalerate) on aquatic environment due to their persistence and accumulation. This article will focus on non-target organisms in inland fresh water environment with special reference to fin fishes and will critically evaluate the toxicity of these pyrethroids in terms of growth inhibition, metabolic disorders, neurotoxicity, reproductive failure, enzymatic dysfunction, haematological alterations, and tissue damages. The rationalized information in this milieu may be useful in ecological risk evaluation and human health management as fish serves as an important bio-indicator for aquatic systems health.

**Keywords** Deltamethrin · Cypermethrin · Fenvalerate · Fishes · Pyrethroids · Toxicological assessment

### Abbreviations

AChE	Acetylcholine esterase
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
ATP	Adenosine triphosphate
CAT	Catalase enzyme
CNS	Central nervous system
DM	Deltamethrin
EPA	Environmental Protection Agency
GABA	Gamma-aminobutyric acid
GOT	Glutamic oxaloacetic acid
GPx	Glutathione peroxidase
GSH	Glutathione
GST	Glutathione s-transferase
Hb	Haemoglobin
HSI	Hepatosomatic index
LDH	Lactate dehydrogenase
LPO	Lipid peroxidase
MCHC	Mean corpuscular hemoglobin concentration
MCH	Mean corpuscular hemoglobin
MCV	Mean corpuscular volume
MDA	Melonaldehyde
NPIC	National Pesticides Information Center
PCV	Packed cell volume
RBC	Red blood cell
SDH	Succinate dehydrogenase
SGR	Specific growth rate
SOD	Superoxide dismutase
TBARS	Thiobarbituric acid reactive substances
WBC	White blood cell
WHO	World Health Organisation

### Introduction

There has been surging increase in the use of agricultural chemicals like pesticides to preserve the standing crops from the attack of pests and to boost up crop production, in order to meet the ever-increasing food demand of the rising human population (Aktar et al. 2009; Chandola et al. 2011; Gupta et al. 2012). Due to injudicious and indiscriminate use of pesticides, the natural water resources such as lakes, reservoirs, wetlands, rivers, ponds, paddy-fields, streams, and other low-lying areas are getting polluted all over the world



(Gilliom et al. 2007; Ngidlo 2013). Pesticides, affect the whole ecosystem, particularly the aquatic ones, leading to unwarranted mortality of aquatic biota, in general and fish in particular as revealed by several workers (Sadhu 1993; Kumari 2005; Gupta et al. 2013). Numerous compounds have been detected in surface water, groundwater, and water supply relating to agricultural activities and human cases of environmental contamination (Sánchez-Bayo 2006). Aquatic pollution has erupted as global problem in recent past. Massive fish kills are recorded rather frequently, and changes in the population of the fauna as a consequence of sublethal effects of aquatic pollutants on ecologically important species have also been described (Koprucu and Aydin 2004; Lazartigues et al. 2013). The pollution of rivers and lakes with chemicals of anthropogenic origin may have adverse consequences. Indiscriminate and wide spread use of pesticides primarily in agricultural sector ultimately leads to the contamination of aquatic environment and becomes hazardous to the non-target aquatic organisms. The widespread application of agricultural pesticides has attracted the attention of ecologists to understand the impacts of these chemicals on aquatic biotic communities (Relyea 2005). In recent years, the manufacture of chemicals such as fertilizers and pesticides, has led to an expansion in the levels of xenobiotic compounds in aquatic ecosystems (Jesus and Carvalho 2008). A new class of agricultural chemicals, the synthetic pyrethroids belonging to major class of insecticides is rapidly replacing other agricultural chemicals in recent years for controlling the insect and pests due to their low mammalian toxicities and potent insecticidal action and accounting for at least 30 % of the global insecticide market (Casida 1980; Shafer et al. 2005). Pyrethroids are synthetic chemicals modeled after the pyrethrin components of pyrethrum, a naturally occurring chemical found in certain chrysanthemum flowers (National Pesticide Information Center 2010). Modern synthetic pyrethroids have been designed to provide enhanced residual activity with greater photostability and high cost effectiveness. Kilgore and Mingyuli (1975) emphasized that concentration of pesticide residues were found to be more in aquatic ecosystem than terrestrial ecosystem. The overall impact is more in aquatic environment, as pesticides and other harmful substances are transported to greater distances in the hydrosphere affecting many more non-target organisms. Synthetic pyrethroid insecticides are photostable analogs of the natural pyrethrins of botanical origin. These halogenated, lipophilic and photostable compounds are active against many target insect and pests. Though these are relatively harmless to birds and mammals but are extremely toxic to many marine and freshwater forms including aquatic invertebrates, insects and fish (DeMicco et al. 2010; Prusty et al. 2011; Vani et al. 2011).

Two distinct types of synthetic pyrethroids have been identified based on different behavioral, neurophysiological, chemical, and biochemical profiles (Coats 1990; Verschoyle and Aldridge 1980), i.e., Type I (noncyano pyrethroids such as permethrin, allethrin, cismethrin, resmethrin, etc.) and Type II ( $\alpha$ -cyano pyrethroids such as deltamethrin, cypermethrin and fenvalerate). Type II ( $\alpha$ -cyano) pyrethroids are more potent neurotoxicants than Type I (non cyano) pyrethroids, with toxicity being solely attributed to  $\alpha$ -cyano substituent (Narahashi 1986; Ecobichon 1991). Diverse characteristic of pesticide toxicity have been extensively reviewed and summarized in the form of review articles, reports and books (Mulla and Mian 1981; Matsumura 1985; Ecobichon 1991; Eisler 1992; Barron and Woodburn 1995; Delorenzo et al. 2001; Akerblom 2004). The potential for widespread use of synthetic pyrethroids have strongly called for thorough examination of their toxic impact on fishes which is a potent indicator for assessing aquatic ecosystem health (Scott and Sloman 2004). Some aspects of fenvalerate toxicity to aquatic organisms have been reviewed (Eisler 1992). Akerblom (2004) has also reviewed toxicity of some agricultural pesticides on aquatic organisms. Given the fact that, insecticides are not selective and impinge on non target species as readily as target organisms, it isn't surprising that a chemical that acts on the insect's different systems will elicit similar effects in higher forms of life (Dogan and Can 2011). Though a lot of advancement has been made in understanding the mode of action and toxic effect of these groups of pesticides on different fishes, concise information of toxic impact of this group of pesticides on various physiochemical, biological and metabolic processes of fishes is lacking. This review attempts to summarize the latest research findings of effects of selected type II synthetic pyrethroids (deltamethrin, cypermethrin and fenvalerate) on freshwater fish fauna.

### Mechanism of pyrethroid action

The synthetic pyrethroids exhibit two different characteristic acidic portions, hrysanthemic or pyrethric acids resulting in type I and type II syndrome (Ecobichon 1991). The differences between type I and II pyrethroids



are expressed in the motor nerve terminals, where type I cause presynaptic repetitive discharges, and type II cause a tonic release of transmitter indicative of membrane depolarization (Salgado et al. 1983). Type II pyrethroids are a more potent toxicant than type I in depolarizing the nerves (Salgado et al. 1983). Type II pyrethroids are associated with faster activation-deactivation kinetics on the Navu sodium channels than type I pyrethroids in vertebrates (Breckenridge et al. 2009). The higher toxicity of type II pyrethroids is mostly attributed to the hyperexcitatory effect on the axons which results from their stronger membrane depolarizing action. Type I pyrethroids modify the sodium channels in the closed state, while Type II pyrethroids modify the open but not inactivated sodium channels (Soderlund 2010). However, this relationship does not always hold true; cis-permethrin and fenvalerate interact with both closed and open sodium channels, but they bind with greater affinity to the open state (Forshaw et al. 2000; Cantalamessa 1993). Type I repetitive discharges have been shown to be suppressed by cypermethrin, indicating that the two pyrethroid types can interact antagonistically. Pyrethrins, pyrethroids, DDT and DDT analogs belong to a group of chemicals that are neurotoxic and share a similar mode of action that is distinctive from other classes of insecticides. There are several ways that pyrethrins and pyrethroids can enter the body of an organism to exert their effects. The first mode is non-stereospecific with rapid penetration through the epidermis, followed by uptake by the blood or hemolymph carrier proteins and subsequent distribution throughout the body. Pyrethroid diffusion along the epidermis cells is the main route of distribution to the CNS after penetration (Naumann 1990). Pyrethroids also can enter the CNS directly via contact with sensory organs of the peripheral nervous system. The sensory structures of both invertebrates and vertebrates are sensitive to pyrethroids (Soderlund and Bloomquist 1989). Pyrethroids can also enter the body through the airway in the vapor phase, but such penetration represents only a small contribution due to the low vapor pressure of pyrethroids. Pyrethroids can also be ingested, and penetration into the blood-hemolymph through the alimentary canal can play an important role in toxicity. Both Type I and II pyrethroid insecticides affect the sodium channels in the nerve membranes, causing repetitive neuronal discharge, the effects being quite similar to those produced by DDT. There appears to be a prolongation of sodium influx with a delay in the closing of the sodium activation gate, resulting in an increased and prolonged sodium tail current (Narahashi 1986; Bradbury and Coats 1989a, b). Type II pyrethroids prolongs the sodium channel open-time much more drastically than Type I (Narahashi 1986). Other sites of action have been noted for the pyrethroid insecticides. Some of them inhibit  $\text{Ca}^{2+}$ ,  $\text{Mg}^{2+}$ -ATPase, thereby interfering with calcium removal from the nerve endings, resulting in increased neurotransmitter release in the postsynaptic gap. In addition, the protein calmodulin, responsible for the intracellular binding of calcium ions to reduce spontaneous neurotransmitter release, can be inhibited. Type II pyrethroids have also been shown to bind to the GABA-receptor chloride channel complex, blocking chloride ion transport into the nerve cell (Ecobichon 1991). Pyrethroids affect the voltage-sensitive calcium channels, GABA-receptors as well as GABA-activated channels and voltage-sensitive chloride channel (Soderlund 2010). Symington et al. (1999) assessed that pyrethroids can modulate the activity of voltage-gated calcium channels. However, these studies reported conflicting results on the inhibitory effects of pyrethroids on voltage gated calcium channels (Meyer et al. 2008). Neal et al. (2010) demonstrated that allethrin significantly altered the voltage dependency of activation and inactivation of L-type voltage gated calcium channels, which suggests that differential modulation of voltage gated calcium channels subtypes could elucidate some of the conflicting observations of other studies. Type II pyrethroids are more potent enhancers of  $\text{Ca}^{2+}$  influx and glutamate release under depolarizing conditions than Type I pyrethroids (Wang et al. 2006; Forshaw et al. 2000). The GABA receptor-chloride ionophore complex is also a target of Type II pyrethroids. GABA is an inhibitory transmitter in the synapse of the CNS of both vertebrates and invertebrates. Pretreatment with diazepam (a benzodiazepine anticonvulsant known to act on the GABA receptors) has been shown to selectively delay the onset of toxic symptoms of Type II, but not Type I, pyrethroids in cockroaches and mice (Soderlund and Bloomquist 1989). Radioligand binding studies have shown that deltamethrin, but not its non-toxic rt-R-cyano epimer, inhibited dihydropicrotoxin binding to the chloride ionophore in the rat brain GABA receptor complex (Soderlund and Bloomquist 1989). Pyrethrins and pyrethroids also inhibit the chloride ion channel function at the GABA receptor-ionophore complex (Casida and Lawrence 1985). An additional target proposed for type II pyrethroids is the membrane chloride ion channel (Hemmen and Brouwer 1995). Generally type II pyrethroids decrease the open channel probability of chloride channels, but the type I pyrethroids do not seem to have an effect on the chlorine channel (Breckenridge et al. 2009; Symington et al. 1999; Burr and Ray 2004). Moreover, Burr and Ray (2004) found that the Type I pyrethroid bioallethrin, and Type II pyrethroids:



cyfluthrin, cypermethrin, deltamethrin and fenprothrin, significantly decreased the probability that the ligand-gated chloride channel would be an open channel. On the contrary, Type I pyrethroids, bifenthrin, bioresmethrin, cis-permethrin and cis-resmethrin, and the Type II pyrethroids, cyfluthrin, lambda-cyhalothrin, esfenvalerate and telluthrin, show the different mode of action. Interestingly, the Type I pyrethroid, bioallethrin, significantly alters the probability of opening the ligand-gated chloride channel, but has generally a weaker response than Type II pyrethroids (Breckenridge et al. 2009). One hypothesis was that bioallethrin may be a mixed-type pyrethroid (Soderlund 2010; Burr and Ray 2004). The blockade of the voltage-sensitive chloride channels is associated with salivation, which is a hallmark of Type II pyrethroid intoxication and could contribute to the enhanced excitability of the CNS (Soderlund 2010). Pyrethroids inhibit the Ca-ATPase, Ca-Mg ATPase neurotransmitters and the peripheral benzodiazepine receptors (Matsumura and Ghiasuddin 1983) but their action on these sites is minor compared with the voltage-gated sodium channels. To summarize, there are many similarities between the mechanism of action of pyrethroid insecticides and organochlorines, consequently there is a risk for additive or even synergistic effects.

### Pyrethrum, pyrethrins and pyrethroids

Pyrethrum is an extract derived from chrysanthemum (*Dendranthema grandiflora*) flowers with insecticidal properties. Pyrethrins are more refined pyrethrum extract, intended to further isolate the insecticidal components of pyrethrum. EPA regulates pyrethrins as one active ingredient; however, the refined extract contains a mixture of six isomers. Pyrethroids are a class of synthetic insecticides that are structurally similar to pyrethrins and act in a similar manner to pyrethrins, but have been modified to increase their environmental stability and their insecticidal properties (Bradberry et al. 2005; EPA 1999; Casida 1980). In general, pyrethrins and pyrethroids are less toxic to mammals and are considered better candidates for replacement of the more toxic organophosphate insecticides. The lowest lethal oral dose of pyrethrum is 750 mg/kg for children and 1000 mg/kg for adults (Occupational Health Services New York 1987). Oral LD50 values of pyrethrins in rats range from 200 mg/kg to greater than 2600 mg/kg (Hayes 1982). Some of this variability is due to the variety of constituents in the formulation. Mice have a pyrethrum oral LD50 of 370 mg/kg (Occupational Health Services New York 1987).

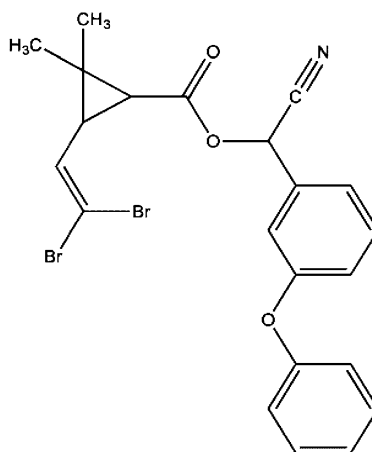
### Deltamethrin

Deltamethrin (DM) is a Type II broad-spectrum pyrethroids, first described and commercialized in 1974 and 1978, respectively (Tomlin 2006). Use of DM extended from agriculture and home formulations to outdoors on lawns, ornamental gardens, golf courses, and indoors as a spot, crack and crevice treatment (Hayes 1982; Elliot et al. 1974). DM is one of the most widely used and known crop protection substances. It is used in many products for outdoor applications in a wide range of crops and in indoor uses against pests like Lepidoptera, Hemiptera, especially Aphids, Coleoptera and Diptera. Deltamethrin is a broad-spectrum insecticide acting as a contact and stomach poison. Deltamethrin is the active ingredient in Butoflin, Butoss, Butox, Cislin, Crackdown, Cresus, Decis, Decis-Prime, K-Othrin and K-Otek. It is the first potent and photostable insecticide belonging to the Type II pyrethroid group (Fig. 1). The mechanism of its toxicity in fish is through blocking of the sodium channels of nerve filaments lengthening the depolarisation phase. Deltamethrin also affects the GABA receptors in the nerve filaments (Eshleman and Murray 1991; Moid et al. 2012). The physico-chemical properties of DM have been represented in Table 1

#### Deltamethrin toxicity to fish

Deltamethrin has the tendency to bind tightly to soil particles. It has a half-life ranging from 5.7 to 209 days that depends on the soil chemistry, texture, microbes in soils (NPIC 2010). Deltamethrin is not likely to evaporate into the air or dissolve easily into water and is moderately to highly toxic to fish under laboratory conditions. Moreover, when products are used according to the label, deltamethrin is less likely to affect fish as it is more likely to be bound to the sediment. Fish and various other aquatic organisms are extremely





**Fig. 1** Chemical structure of Deltamethrin

**Table 1** Physico-chemical properties of deltamethrin

Variable	Information
Appearance	Colorless crystalline powder; white or slightly beige powder
Chemical name	Cyano (3-phenoxy-phenyl) methyl; 2-(2,2dibromoethenyl)—2,2-dimethylcyclopropanecarboxylate (CA); alpha-cyano-m-phenoxybenzyl, (1R,3R)-3-(2,2-dibromovinyl)-2,2-dimethyl-cyclopropan-carboxylate, (S)-alpha-cyano-3-phenoxybenzyl (1R)- <i>cis</i> -3-(2,2-dibromovinyl)-2,2-dimethylcyclopropane-carboxylate
CAS number	52918-63-5
Chemical formula	C <sub>22</sub> H <sub>19</sub> Br <sub>2</sub> NO <sub>3</sub>
Molecular weight	505.24
Water solubility	Less than 0.1 mg/L, Insoluble: <1 ppm at room temperature, 0.002 mg/L at 20 °C, Almost insoluble
Solubility in other solvents	In kerosene and isoalkanes, less than 0.5, isopropanol 0.6, ethanol 1.5, xylene 25, methylene chloride 70 (all in g/100 g at 20 °C). In acetone 500 g/L, benzene 450 g/l, dimethyl sulfoxide 450 g/L, cyclohexanone 750 g/l, dioxane 900 g/l all at room temperature. Toluene 250 g/L
Melting point	98-101 °C
Vapor pressure	2 × 10 <sup>-8</sup> mbar at 25 °C
Partition coefficient	4.6 (25 °C)
Aquatic field test half-life (days)	<2
Terrestrial field test half-life (days)	14–231
Hydrolysis half-life (days)	<33

Hayes (1982), Worthing (1983, 1987), Hayes and Laws (1990a, b), Kidd and James (1991), NPIC (2010), California EPA (2000) and European Commission (2002)

susceptible to pyrethroids as the 96-h Lc50 value determined in laboratory tests generally lies below 10 µg/L (Amin and Hashem 2012). In addition, deltamethrin is based on pyrethroids that have established significantly lower rates of metabolism and removal in fish than those recorded for birds and mammals (Bradbury and Coats 1989a, b). The development of oxidative stress in different fish tissues following deltamethrin exposure has been suggested as the main cause of toxicity (Sayeed et al. 2003; Abdollahi et al. 2004; Diana et al. 2007). Extensive use of deltamethrin may affect the aquatic environment and can kill or at least adversely influence the aquatic life especially fish which are highly sensitive to deltamethrin (De Assis et al. 2009; Pimpao et al. 2008). Rodrigues (2003) explained the high sensitivity of DM absorption due to lipophilic characteristic even at lower concentration by fish gills as compared to mammals. The 96-h LC50 ranges from 0.91 to 3.50 µg/L depending on the fish species (WHO 1990; Tomlin 2006). Acute toxicity data for deltamethrin in fish have



**Table 2** Toxicity studies on the effects of deltamethrin on various fish species

Scientific name	LC 50 value	References
<i>Ctenopharyngodon idella</i>	155.0 µg/L (24 h), 96.0 µg/L (48 h), 91.0 µg/L (96 h)	Rao et al. (1983)
<i>Cyprinodon macularious</i>	0.60 µg/L (24 h) 0.60 µg/L (48 h)	Mulla et al. (1978)
<i>Cyprinus carpio</i>	3.5 µg/L (24 h), 3.5 µg/L (48 h), 3.5 µg/L (96 h)	Lakota et al. (1989)
<i>Cyprinus carpio</i>	4.00 µg/L (48 h) 2.30 µg/L (96 h)	Sun (1987)
<i>Cyprinus carpio</i>	91.0 µg/L (24 h), 89.0 µg/L (48 h), 78.0 µg/L (96 h)	Rao et al. (1983)
<i>Cyprinus carpio</i>	0.058 µg/L (96 h)	Svobodova et al. (2003)
<i>Cyprinus carpio</i>	9.41 µg/L (24 h), 4.47 µg/L (48 h), 2.37 µg/L (72 h), 1.65 µg/L (96 h)	Calta and Ural (2004)
<i>Cyprinus carpio</i>	0.213 µg/L (48 h), 0.074 µg/L (48 h)	Koprucu and Aydin (2004)
<i>Esox Lucious</i>	44.0 µg/L (24 h), 30.0 µg/L (48 h), 23.0 µg/L (96 h)	Rao et al. (1983)
<i>Gambusia affinis</i>	1.00 µg/L (24 h), 1.00 µg/L (48 h)	Mulla et al. (1978)
<i>Oncorhynchus mykiss</i>	0.50 µg/L (24 h), 0.70 µg/L (48 h)	Mulla et al. (1978)
<i>Oncorhynchus mykiss</i>	2.50 µg/L (24 h), 2.30 µg/L (48 h), 2.30 µg/L (96 h)	Lakota et al. (1989)
<i>Oncorhynchus mykiss</i>	8 µg/L (12 h), 10 µg/L (24 h), 12 µg/L (48 h), 25 µg/L (72 h), 50 µg/L (96 h)	Ural and Sanglam (2005)
<i>Tilapia mosambica</i>	0.80 µg/L (24 h), 0.80 µg/L (48 h)	Mulla et al. (1978)
<i>Tilapia nilotica</i>	16.0 µg/L (24 h), 15.0 µg/L (48 h), 14.5 µg/L (96 h)	Golow and Godzi (1994)
<i>Oreochromis niloticus</i>	1.17 µg/L (48 h), 1.70 µg/L (48 h), 1.45 µg/L, 4.85 µg/L	Karasu Benli et al. (2009) Kan et al. (2012) Yildirim et al. (2005)
<i>Poecilia reticulata</i>	24.0 µg/L (24 h), 21.0 µg/L (48 h), 20.0 µg/L (72 h), 19.0 µg/L (96 h), 18.0 µg/L (120 h)	Stalin et al. (2008)



**Table 2** continued

Scientific name	LC 50 value	References
<i>Poecilia reticulata</i>	5.13 µg/L (48 h)	Viran et al. (2003)
<i>Clarias gariepinus</i>	0.01 µg/L (24 h)	Datta and Kaviraj (2003)
<i>H. fossilis</i>	1.86 µg/L (96 h)	Srivastava et al. (1997)
<i>H. fossilis</i>	0.52 mg/L (96 h)	Kumar et al. (1999)
<i>H. fossilis</i>	1.5 mg/L(96 h)	Srivastava et al. (2010)
	0.37 mg/L	
<i>Channa punctatus</i>	0.75 mg/L (96 h)	Jayaprakash and Shettu (2013)
<i>Channa punctatus</i>	0.75 µg/L	Sayeed et al. (2003)
<i>Labeo rohita</i>	1.00 mg/L (96 h)	Rathnamma et al. (2009)
<i>Catla catla</i>	4.83 µg/L (96 h)	Vani et al. (2011)
<i>Xiphophorus helleri</i>	2.87 µg/L (96 h)	Khalili et al. (2012)
<i>Oncorhynchus mykiss</i>	0.3 and 0.6 µg/L	Atamanlp and Erdogan (2010)
<i>Danio rario</i>	0.016, 0.025 and 0.043 µg/L	Sharma and Ansari (2010)
<i>Danio rario</i>	0.5 and 1 µg/L	Koc et al. (2009)
<i>Clarias gariepinus</i>	0.75 µg/L	Amin and Hashem (2012)
<i>Carassius auratus</i>	2 µg/L (48 h)	Diana et al. (2007)

been summarized in a report of the World Health Organization (WHO 1990) and classified as highly toxic to fish. The toxicity may be lethal to fish or can induce stress resulting in alteration at molecular, biochemical or physiological levels. Deltamethrin has been reported to cause significant effects on various physiological and biochemical activities of freshwater fishes. Exposure to deltamethrin interferes with the process of neural transmission, blocking, in open position, the ionic channels and induces changes in enzymatic activity and hormonal disorder in freshwater fishes. Summarized information on studies conducted regarding deltamethrin toxicity in fishes is given in Table 2. Fishes seem to be deficient in the enzyme system that hydrolyzes pyrethroids. The main reaction involved in the metabolism of deltamethrin in fish is largely oxidative (Demoute 1989). After short-term deltamethrin exposure, adult *Heteropneustes fossilis* (freshwater catfish) showed hypocalcemia and the researchers attribute this condition to the possible impairment of either net electrolyte influx at the gill or impairment of renal function (Srivastava et al. 1997). Deltamethrin exposure also reportedly caused hypophosphatemia which could be linked to the possible redistribution of electrolytes between intracellular or extracellular compartments and/or impairment of renal function (Srivastava et al. 1997).

#### Histopathological alterations

The biochemical and histopathological effects on fish, at low and high concentrations of deltamethrin has been studied (Kopru et al. 2006; Velisek et al. 2006a, b, c; Yildirim et al. 2006). Yildirim et al. (2005) observed severe histopathological changes in gills and liver of *Oreochromis niloticus* as a result of acute deltamethrin toxicity. Cengiz (2006) observed histopathological changes due to deltamethrin exposure on the gill (desquamation, necrosis, aneurysm in secondary lamellae, lifting of the lamellar epithelium, edema, epithelial hyperplasia and fusion of the secondary lamellae) and kidney (degeneration in the epithelial cells of renal tubule, pycnotic nuclei in the haematopoietic tissue, dilatation of glomerular capillaries, degeneration of glomeruli, intracytoplasmatic vacuoles in epithelial cells of renal tubules with hypertrophied cells and narrowing of the tubular lumen) of common carp after acute exposure at a concentration of 0.029 and 0.041 mg/L. Histological alteration in the gills such as hyperemia, fusion of secondary lamellae, epithelial layer rupture and chloride cell proliferation were also reported by Diana et al. (2007) in their study on *Carassius auratus* after 48 h of deltamethrin exposure at 2 µg/L concentration. Histological changes in deltamethrin-induced intoxication in liver and kidney of gold fish, *Carassius auratus*, was reported by Staicu et al. (2007). Exposure of zebrafish embryos to pyrethroids caused a dose-dependent increase in mortality and pericardial edema, with Type II compounds being the most potent and as doses approaching the LC50, permethrin and deltamethrin





caused craniofacial abnormalities (DeMicco et al. 2010). The study by Kan et al. (2012) suggested deltamethrin to be the causative agents for creating micronucleus and altering the histopathology of liver and gills of *Oreochromis niloticus*.

### Biochemical and hematological alterations

Pyrethroids are readily absorbed by fish gills. After distribution to bile, liver, kidney and red blood cells, they are metabolized by hydrolysis, hydroxylation and conjugation to glucuronides and sulphates. Chronic effects of pyrethroids include growth retardation, behavioral, blood profile and histopathological changes besides compromised immune system and endocrine disruption (Richterova and Svobodová 2012). The effects of deltamethrin on acetylcholinesterase (AChE) and some biochemical parameters such as glutamic oxaloacetic transaminase (GOT), lactate dehydrogenase (LDH) and glucose in different tissues of adult *Cyprinus carpio* were studied by Balint et al. (1995) who have reported inhibition of AChE with the increase in GOT and LDH activities along with increase in blood glucose level. Inhibitory effect of deltamethrin at concentrations above 2 µg/L on the monooxygenase system of carp liver (*Cyprinus carpio* L.) was reported by Deer et al. (1996). Kumar et al. (1999) studied the deltamethrin-induced hematological changes in *Heteropneustes fossilis* and found a significant increase of erythrocyte, leucocyte, lymphocyte, neutrophil count and a slight reduction in haemoglobin and monocytes. They have also reported an inhibition of LDH activity upon exposure to 1/3rd LC50 of synthetic pyrethroid, deltamethrin. Deltamethrin pollutants exposed for 48 and 96 h resulted in decrease of phosphatidylethanolamine (PE), phosphoglyceride (PG) and phosphatidic acids (PA) in plasma membrane of carp, *Cyprinus carpio* erythrocytes (Kotkat et al. 1999). Sayeed et al. (2003) demonstrated the in vivo inhibition of kidney, liver and gill catalase enzymes of *Channa punctatus* by deltamethrin. Exposure of freshwater catfish, *Channa punctatus*, to deltamethrin increased Catalase (CAT) and Glutathione peroxidase (GPx) activity in the liver and kidney but decreased the same in gill tissue (Sayeed et al. 2003). Changes in biochemical profile of *Cyprinus carpio* upon deltamethrin exposure was also reported by Velisek et al. (2006a, b). Significant decrease in activities of catalase, glutathione peroxidase and glutathione reductase enzymes in *Carassius auratus* was observed upon deltamethrin exposure by Diana et al. (2007). A decrease in GST (Glutathione S transferase) enzyme activity was also observed in the gill of *Ancistrus multispinis* exposed to deltamethrin (Pimpao et al. 2007). In vivo experiments showed that deltamethrin significantly inhibited the glucose-6-phosphate dehydrogenase enzyme activity after 48 h of exposure (Senturk et al. 2009). Depletion in pyruvate level during elevation in lactate level in the gill, muscle and liver tissues of *Labeo rohita* upon sub lethal exposure of deltamethrin has been reported (Rathnamma et al. 2009). Significant inhibition of carbonic anhydrase activity, which plays an important role in the osmotic and acid–base regulation, in the gill tissues of rainbow trout (*O. mykiss*) was also reported (Ceyhun et al. 2010a, b) and it was observed that the activities of the glutathione reductase, glucose 6-phosphate dehydrogenase, 6-phosphogluconate dehydrogenase decreased with the consequent increase in deltamethrin concentrations and exposure time. The pesticide had more inhibitory effects on gill enzymes than those of muscle, liver and kidney. Expression of stress protein, heat shock protein (HSP 70), was also reported by same authors suggesting that deltamethrin induces stress at both protein and mRNA levels (Ceyhun et al. 2010a). In another study, deltamethrin was observed to be significantly inhibiting rainbow trout carbonic anhydrase enzymes invitro and invivo (Ekinci and Beydemir 2010). It was reported that deltamethrin caused gradual decrease on activities of different antioxidant enzymes in the first 3 days (Dinu et al. 2010). Similar inhibition in metabolic enzyme activities in *C. catla* fingerlings were reported (Vani et al. 2011). It is possible that deltamethrin or its lipophilic metabolites may bind to the lipid moiety in vivo which may result in altering the allosteric characteristic of the enzymes thus leading to the inhibition of different enzyme activity (Vani et al. 2011). Deltamethrin inhibited the digestive enzyme, lipase activity in carp, *Cyprinus carpio* (Simon et al. 1999). Similarly inhibition of lipase activity in guppy, *Poecilia reticulata*, was observed by Gunes and Yeril (2011). Deltamethrin-associated inhibition in alkaline phosphatase and acid phosphatase in liver, muscle and ovary tissues of zebra fish, *Danio rerio*, has been observed by Sharma et al. (2012). Alteration in antioxidant enzymes of *Danio rerio* has also been reported by Sharma and Ansari (2010). Ensibi et al. (2013) also observed significant alteration in liver biomarkers such as malondialdehyde (MDA), catalase and glutathione reductase.

Results obtained from various studies conducted on the effects of deltamethrin on hematological parameters are inconsistent. Effects of deltamethrin on nervous, respiratory and hematological systems in fishes are



reported by several authors (Srivastava et al. 1997; Kumar et al. 1999; Csillik et al. 2000; DeMicco et al. 2010; Amin and Hashem 2012; Galeb et al. 2013). In catfish (*Heteropneustes fossilis*) deltamethrin caused a significant increase in RBC, which suggested that erythropoiesis has been accelerated to avoid anemic state that results in increase in number of erythrocytes and decrease in Hb, MCV, MCH and PCV (Kumar et al. 1999). In common carp (*Cyprinus carpio*) acute intoxication of deltamethrin has been reported to cause decrease in RBC, Hb and PCV and no effect on MCV, MCH, MCHC, total leukocyte count and relative as well as absolute counts of lymphocytes, monocytes, neutrophil granulocytes and their developmental forms (Svobodova et al. 2003). Chronic exposure of deltamethrin has been reported to result in decreased lymphocyte and basophile percentages, decreased total leukocyte counts, erythrocyte counts, Hb and PCV simultaneously with serious hypoproteinaemia, hypoalbuminemia, hypercholesterolemia and hyperglycemia in Nile tilapia when exposed to subacute concentration for weeks (El-Sayed et al. 2007; El-Sayed and Saad 2008). It has been suggested that changes in leukocyte number is a sensitive indicator of stress in fish (Barton and Iwama 1991). These changes, however, are mostly related to immunosuppression. Deltamethrin injected intraperitoneally induces leukocytosis and increases the number of erythrocytes and hemoglobin levels (Pimpao et al. 2007). Deltamethrin-induced genotoxic damage to erythrocytes in terms of nuclear abnormalities accompanied by increased lipid peroxidase activity has been observed by Ansari and Waleem 2009). Reduced lipid peroxidase activities in *Clarias gariepinus* have also been observed by Amin and Hashem (2012). One recent Study showed that deltamethrin is moderately toxic for the freshwater fish *H. fossilis* by producing adverse effects on serum calcium and prolactin cells (Srivastava et al. 2010). Vani et al. (2011) have demonstrated that deltamethrin has negative effect on the hematological and biochemical parameters of *C. catla*, which might be due to possible disruption of hematopoiesis and proteosynthesis. Catalase activities in liver, kidney and gill tissues of *Clarias gariepinus* upon 48-h exposure of 0.75 µg/L deltamethrin was reported by Amin and Hashem (2012). The same authors also observed significant increase in serum AST, ALT, urea and creatinine and marked decrease in total protein and serum albumin level.

#### Reproductive physiology of fishes

The effects of deltamethrin on the sensitive early life stages of zebrafish, *Brachydanio rerio*, were examined by George and Nagel (1990). Deltamethrin may disturb the calcium and phosphate homeostasis and may lead to adverse effect on the reproductive state of the fish as these ions are important for vitellogenin synthesis (Srivastava et al. 1997). Koc et al. (2009) studied the effects of different concentrations of deltamethrin—control, 0.5 and 1 µg/L on the ovaries of Zebra fish (*Danio rerio*)—and reported an increase in atretic oocytes which showed significant negative effects of deltamethrin on oogenesis. Koprucu and Aydin (2004) have reported that low level of deltamethrin (0.005 µg/L) in the aquatic environment may have a significant effect on carp populations. The other responses included impairment of ontogenic process and reduction in hatchability of embryos of zebra fish. Similarly reproductive impairment in zebra fish was also reported by Sharma and Ansari (2010). Deltamethrin has also been reported to alter normal ion flux in *Ancistrus multifilis* (De Assis et al. 2009). Further, Gunes and Yeril (2011) noticed that deltamethrin inhibited the lipase activity which has a negative effect on nutrition and physiological condition of fish causing poor reproductive responses in Guppies (*Poecilia reticulata*).

#### Behavioral response of fishes

The main behavioral changes observed as result of deltamethrin exposure are represented by respiratory and neurological manifestations. Rapid gill movement, erratic swimming, swimming at the water surface and gulping for air and prolonged and motionless laying down on the bottom were observed in different fishes; guppy, *Poecilia reticulata* (Viran et al. 2003); common carp, *Cyprinus carpio* (Dziaman et al. 2010); silver catfish (Galeb et al. 2013). Ural and Saglam (2005) observed increase in fish mortality with increase in concentration of deltamethrin which may be attributed to disruption of normal physiological and homeostatic organs. El-Sayed and Saad (2008) described the abnormal behavioral responses and toxic symptom that resulted due to subacute toxicity of deltamethrin in monosex Nile tilapia, (*Oreochromis niloticus* L.). Deltamethrin-associated swimming ability reduction in juvenile rainbow trout (*Oncorhynchus mykiss*) has also been reported by Goulding et al. (2013).



## Amelioration of deltamethrin toxicity in fishes

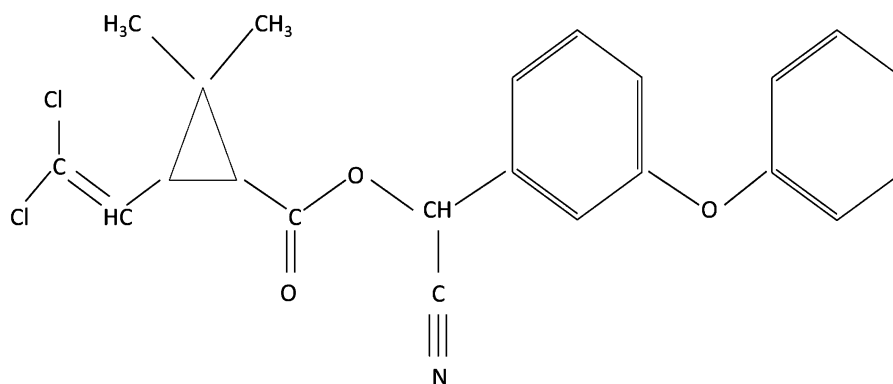
The effect of cadmium pretreatment on deltamethrin-induced oxidative stress and alterations of antioxidants such as activities of glutathione peroxidase, glutathione reductase and glutathione- S-transferase in liver, kidney and gills of freshwater fish *Channa punctata* was reported by Atif et al. (2005). One of the protective mechanisms attributed to cadmium involves induction of metallothioneins, which are reported to be involved in protection against chemically induced oxidative stress and also act as a free radical scavenger (Jeong et al. 2004; Takahashi et al. 2004). DeMicco et al. (2010) observed that treatment with diazepam ameliorated the spasms while treatment with the sodium channel antagonist MS-222 ameliorated both spasms and body curvature which were reminiscent of the classic syndromes observed with pyrethroid toxicity in zebrafish. Vani et al. (2011) have confirmed the ameliorative effect of vitamin C supplementation in diet against harmful effects of deltamethrin exposure in *C. catla* fingerlings. Lycopene, a carotenoid found in tomatoes and tomato products was reported to provide protection against deltamethrin-induced oxidative stress by decreasing lipid peroxidation and altering the antioxidant defense system in blood and tissues in carp, *Cyprinus carpio* (Yonar and Sakin 2011). Furthermore, some of emerging immunostimulant or synthetic compounds may be the alternative choice for developing immunity to specific pesticide or other stress factors in aquatic animals viz. beta glucan, levan, yeast, pyridoxine, etc. (Akhtar et al. 2011, 2012a, b; Meena et al. 2013; Gupta et al. 2012, 2013).

## Cypermethrin

Cypermethrin is an active ingredient in many formula-grade insecticides viz. Colt, Ammo, Avicide, Barricade, CCN 52, Cymbush, Folcord, Imperator, Kafil Super, Polytrin, Ripcord and Stockade. Cypermethrin is relatively stable under sunlight (Fig. 2). Cypermethrin is readily degraded by ester cleavage to give 2,2-dimethyl-3-(2,2-dichlorovinyl) cyclopropane-carboxylic acid (CPA) and 3-phenoxybenzoic acid (PBA) (Camilleri 1984). Cypermethrin is also used as a treatment against parasitic sea louse infestation (*Lepeophtheirus salmonis*) in intensive salmonid aquaculture (Sommerville 1995; Moore and Waring 2001). The Physico-chemical properties of CM have been represented in Table 3.

## Cypermethrin toxicity to fishes

Cypermethrin is the most prevailing and widely employed synthetic insecticide and has been reported to be toxic to various fishes and aquatic invertebrates even at minute concentrations (Collins and Cappello 2006; Wang et al. 2006; Sarkar et al. 2005). Reports show that cypermethrin could affect even early stages of fish more potentially (Polat et al. 2002; Rahmi et al. 2005). It could also result in growth retardation and protein deposition in fish body (Saha and Kaviraj 2013). Aydin et al. (2005) described the acute toxicity of cypermethrin on the common carp (*Cyprinus carpio* L.) embryos and larvae exposed for 1–96 h, which revealed a significant ( $p < 0.05$ ) increase in dead larvae with concomitant increase in concentrations of cypermethrin.



**Fig. 2** Chemical structure of Cypermethrin



**Table 3** Physico-chemical properties of cypermethrin

Variable	Information
Appearance	Pure isomers of cypermethrin form colorless crystals. When mixed isomers are present, cypermethrin is a viscous semi-solid or a viscous, yellow liquid
Chemical name	(R,S)-alpha-cyano-3-phenoxybenzyl(1R)-cis,trans-3-(2,2-dichlorovinyl)-2,2-dimethylcyclopropane-carboxylate
Chemical formula	C <sub>23</sub> H <sub>19</sub> ClF <sub>3</sub> NO <sub>3</sub>
CAS number	52315-07-8
Molecular weight	416.30
Water solubility	0.01 mg/L @ 20 °C; insoluble in water
Solubility in other solvents	methanol vs.; acetone vs.; xylene vs.
Melting point	60-80 °C (pure isomers)
Vapor pressure	5.1 × 10 <sup>-7</sup> nPa at 70 °C
Partition coefficient	6.6020

Ray (1991), Kidd and James (1991) and Wauchope et al. (1992)

The highest concentration 8 µg/L resulted in highest larval mortality. Similarly, Shalvei et al. (2012) revealed the same correlation of cypermethrin concentration and increase in mortality of Caspian Roach (*Rutilus rutilus caspicus*) and Silver Carp (*Hypophthalmichthys molitrix*). Ayoola and Ajani (2008) in their experiment on African catfish (*Clarias gariepinus*) observed that respiratory stress and instant death of fish were observed in exposed fish, which varies with the concentration of the toxicant and showed increased mortality with increase in concentration and concluded that cypermethrin is highly toxic to juvenile fish. Singh et al. (2010) has reported toxicological and biochemical alterations of cypermethrin against *Colisa fasciatus* at different seasons. The results showed a strong piscicidal activity in *Colisa fasciatus* for all the exposure periods (24 or 96 h) in time- as well as dose-dependent manner. In contrast to most of the findings Tiwari et al. (2012) appraised a negative ( $p < 0.05$ ) correlation between effective doses of cypermethrin and exposure periods; that is, LC<sub>50</sub> values decreased from 0.323 µg/L (6 h) to >0.278 µg/L (12 h), >0.240 µg/L (18 h) and >0.205 µg/L (24 h). Exposure to sublethal doses of cypermethrin for 24- and 96-h exposure period caused significant ( $p < 0.05$ ) time- and dose-dependent alterations in most of the hematological and biochemical parameters (Tiwari et al. 2012).

Akinrotimi et al. (2012) assessed the most toxic level of cypermethrin as 0.25 mg/L in their experiment on *Clarias Gariepinus* and suggested that exposure to cypermethrin could cause some level of stress as indicated by changes in the hematological indices of the fish under consideration. Moreover, Bradbury and Coats (1989a) have reviewed the toxicology of pyrethroids in mammals, birds, fish, amphibia and invertebrates and cited 96-h LC50 cypermethrin toxicity as 2.2 µg/L for *Tilapia nilotica*, 0.9–1.1 µg/L for carp (*Cyprinus carpio*), 1.2 µg/L for brown trout (*Salmo trutta*), 0.5 µg/L for rainbow trout (*Salmo gairdneri*) and 0.4 µg/L for *Scardinius erythrophthalmus*. Polat et al. (2002) found the 48-h LC50 value of beta-cypermethrin in male guppies as 21.4 µg/L; Sarikaya (2009) suggested 96-h LC50 values of Alpha-Cypermethrin on *Oreochromis niloticus*, ranges between 0.7 and 350 µg/L ranges (Table 4). Toxicity of cypermethrin to various fishes has always been of immense interest to the researchers in a steady manner and has been documented by several authors (McLeese et al. 1980; Clark et al. 1987; Cakmak and Girgin 2003; Adhikari et al. 2004; Collins and Cappello 2006; Jee et al. 2005; Prashanth and David 2006; Kumar et al. 2007; Korkmaz et al. 2009; Singh et al. 2010; Saha and Kaviraj 2009, 2013).

#### Histopathological alterations

Significant changes such as hyperplasia, disintegration of hepatic mass and focal coagulative necrosis were found in *L. rohita* exposed to cypermethrin (Sarkar et al. 2005). Korkmaz et al. (2009) reported severe histopathological lesions and marked decrease in protein and glycogen levels of different organs of Nile tilapia



**Table 4** Toxicity studies on the effects of cypermethrin on various fish species

Scientific name	LC 50 value	References
<i>Cyprinus carpio</i>	1.1 µg/L (96 h)	Stephenson (1982)
<i>Salmo trutta</i>	1.2 µg/L (96 h)	Stephenson et al. (1982)
<i>Tilapia nilotica</i>	2.2 µg/L (96 h)	Stephenson et al. (1982)
<i>Tilapia mossambica</i>	0.2 µg/L (24 h)	Reddy et al. (1991a, b)
<i>Oncorhynchus mykiss</i>	2.8 µg/L (96 h)	Stephenson (1983)
<i>Oreochromis niloticus</i>	5.99 µg/L (96 h)	Sarikaya (2009)
<i>Labeo rohita</i>	0.139 µg/L ppm (96 h)	Das and Mukherjee (2003)
<i>Labeo rohita</i>	4.0 µg/L (96 h)	Marigoudar et al. (2009)
<i>Cirrhinus mrigala</i>	150 µg/L (96 h)	Vasantharaja et al. (2012)
<i>Poecilia reticulata</i>	21.4 µg/L (48 h)	Polat et al. (2002)
<i>H. fossilis</i>	0.67–1.27 µg/L (72 h)	Saha and Kaviraj (2013)
<i>H. fossilis</i>	3.783 µg/L (96 h)	Bhutia et al. (2013)
<i>H. fossilis</i>	0.67 µg/L (96 h)	Deka and Dutta (2012)
<i>Channa punctatus</i>	0.4 mg/L (96 h)	Kumar et al. (2007)
<i>Rutilus rutilus caspicus</i>	2.314 µg/L (14 h), 1.023 µg/L (48 h), 0.732 µg/L (72 h), 0.627 µg/L (96 h)	Shaluei et al. (2012)
<i>Hypophthalmichthys molitrix</i>	2.962 µg/L (24 h), 1.653 µg/L (48 h), 1.030 µg/L (72 h), 0.917 µg/L (96 h)	Shaluei et al. (2012)
<i>Cyprinus carpio</i>	0.256–5.074 µg/L (48 h)	Aydin et al. (2005)
<i>Clarias gariepinus</i>	0.063 mg/L (96 h)	Ayoola and Ajani (2008)
<i>Colisa fasciata</i>	0.02 mg/L (96 h)	Singh et al. (2010)
<i>Poecilia reticulata</i> ,	9.43 µg/L (96 h)	Yilmaz et al. (2004)
<i>Labeo rohita</i>	0.323 µg/L (6 h), 0.278 µg/L (12 h), 0.240 µg/L (18 h), 0.205 µg/L (24 h)	Tiwari et al. (2012)
<i>Clarias Gariepinus</i>	0.05, 0.10, 0.20 and 0.25 µg/L	Akinrotimi et al. (2012)
<i>Cyprinus carpio</i>	250 µg/L (96 h)	Meenambal et al. (2012)

(*O. niloticus*). Hepatic lesions in the liver tissues were characterized by cloudy swelling of hepatocytes, lipid vacuoles, pycnotic nuclei and focal necrosis. Necrosis in the epithelial cells and pycnosis in the hematopoietic tissue in kidney tissues of *Clarias gariepinus* were observed by Velmurugan et al. (2009). Larval deformities in Japanese medaka (*Oryzas latipes*) due to cypermethrin toxicity were reported by Younghee et al. (2008). Gill hyperemia, fusion of secondary lamellae, epithelial layer rupture and chloride cell proliferation have been observed in deltamethrin exposed gold fish, *Carassius auratus* (Diana et al. 2007). Joshi et al. (2007) observed histopathological changes in the liver cells of *Heteropneustes fossilis*. After 20 days of exposure, the hepatocytes became irregular and lost their polygonal shape. Some cells exhibited cloudy swelling, their contour becoming indistinguishable. Ayoola and Ajani (2008) studied the effects of cypermethrin exposures (96 h) in inducing histopathological changes of gills, liver, kidney and brain tissues. In the gills, filament cell proliferation, cellular infiltration, hemorrhage and epithelial lifting were prominent. In the liver, there was vacuolation of hepatocytes and necrosis. The kidney showed exfoliation and was swollen with pyknotic nuclei. The brain showed neuronal degeneration and spongiosis.

### Biochemical, hematological alterations

Reddy and Philip (1994) reported a significant inhibition of AChE and elevation of acetylcholine content in all tissues viz. gill, brain, liver and muscle of *Cyprinus carpio* with response to exposure of technical grade cypermethrin. Similar results of AChE inhibition in *Channa punctatus* have been reported by Kumar et al. (2009). Amino acids and lactate content increased in the tissue of *Cyprinus carpio* and *L. rohita* when exposed to sub lethal concentration of cypermethrin (Reddy et al. 1991a) causing metabolic diversion in fish to prolong its survivability under severe osmotic imbalance. Decrease in protein content of the fish, *L. rohita* fingerlings exposed to cypermethrin was reported by Das and Mukherjee (2003). Atamanlp et al. (2002a, b) studied the effect of cypermethrin on some biochemical parameters of rainbow trout (*O. mykiss*) and observed marked decline in the calcium and phosphorous of the exposed fishes. Significant increase in levels of free amino acids coupled with marked decline in protein level in *Tilapia mossambica* was reported by Reddy and Yellama (1991a, b). Similar effects with response to cypermethrin in Indain major carp, *C. mrigala*, was observed by Prashanth and David (2006). David et al. (2004) studied the effect of the sub-lethal concentration of cypermethrin on the metabolic profile of *Cyprinus carpio*. Exposure of *C. batrachus* to sublethal concentration of cypermethrin caused alterations in major metabolites and enzymes of protein and carbohydrate metabolism in liver and gill tissues (Begum 2005). Jee et al. (2005) found an increase in levels of serum glutamic-acid oxylacetic- acid-transaminase, glutamic-acid pyruvic-acid-transaminase, glucose, and alkaline phosphatase and a decrease in the concentration of plasma total protein, albumin, cholesterol and lysozyme in Korean rockfish (*Sebastes schlegeli*) exposed to cypermethrin. Velisek et al. (2006b) found an increase in activity of ALP in rainbow trout after exposure to cypermethrin. Cypermethrin was also reported to cause significant alteration in the levels of ammonia and urea in freshwater fish, *C. mrigala* (Prashanth 2007). Marked increase in the activities of transaminases in *C. batrachus* was observed by Begum (2007). Significant increase in asparate and alanine amino transferase along with increased alkaline phosphatase upon cypemethrin exposure has been reported by Loteste et al. (2013) in *Prochilodus lineatus* fish. Significant alterations in enzymes of nitrogen metabolism in fish such as AST, ALT, glutamate dehydrogenase and gutamine synthetase in two freshwater fire breathing fish *Channa striatus* and *C. batrachus* have been reported by Kumar et al. (2011).

Saha and Kaviraj (2009) found a significant increase in glucose level along with marked decrease in glycogen level in *Heteropneustes fossilis* upon cypermethrin exposure. Increase in activities of glycolytic enzyme LDH in gill, kidney, intestine, brain and liver tissues as a result of cypermethrin exposure have been reported by Osman et al. (2013).

Dose- and time-dependent biochemical, hematological alterations have been reported in several fishes: *Colisa fasciatus* (Singh et al. 2010); *C. mrigala* (Vasantharaja et al. 2012); *L. rohita* (Tiwari et al. 2012). Cypermethrin is reported to have paramount effect on different hematological parameters of various fishes (Dorucu and Girgin 2001; Cakmak and Girgin 2003; Adhikari et al. 2004; Jee et al. 2005; Deka and Dutta 2012). Recent work by Ojutiku et al. (2013) reports of significant increase in white blood cell (WBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), monophils and heterophils, while a marked reduction in red blood cells (RBC) and lymphocytes in *Clarias gariepinus* upon cypermethrin exposure.

### Cypermethrin toxicity on reproductive physiology of fishes

Exposure of salmon milt and eggs to a concentration of 0.1 µg/L cypermethrin during fertilization subsequently reduced the number of fertilized eggs, the reason being reduced ability of male parr to respond to the priming effect of the hormone with response to cypermethrin exposure (Moore and Waring 2001).

### Behavioral response of fishes

Responses of fish to cypermethrin toxicity include gill flailing, hyperactivity, loss of buoyancy and inability to remain upright in rainbow trout (Edwards et al. 1986). Montanha et al. (2012) studied the behavior of silver catfish exposed to sub-lethal concentrations of cypermethrin and reported symptoms of poisoning, such as loss of balance, swimming alteration and dyspnea. Breathing and neurological disorders have been reported as a result of cypermethrin exposure in guppies, *Poecilia reticulata* (Polat et al. 2002; Yilmaz et al. 2004), silver catfish, *Rhamdia quelen* (Borges 2007). These changes may be attributed to the neurotoxic effect of



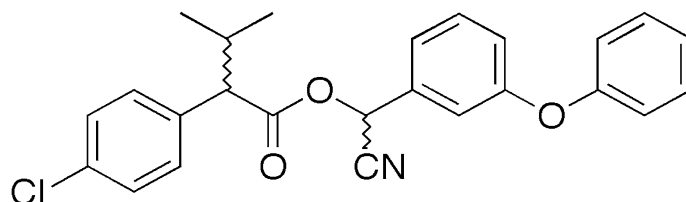
cypermethrin by blocking sodium channels and inhibiting the GABA receptors in the nervous filaments which results in an excessive stimulation of the central nervous system that sometimes can lead to brain hypoxia (El-Sayed et al. 2007). Cypermethrin is reported to act directly on the sodium channels and thereby inhibit the nervous transmissions within the olfactory system (Moore and Waring 2001). Yilmaz et al. (2004) reported behavioral changes of male guppies upon exposure to cypermethrin concentration of 15 µg/L. Significant alterations in different morpho-behavioral patterns of *Channa punctatus* due to cypermethrin exposure were also observed even at sub-lethal doses (Kumar et al. 2007). Ayoola and Anjani (2008) in their experiment on African catfish (*Clarias gariepinus*) observed that respiratory stress and instant death of fish varies with the concentration of the toxicant and showed increased mortality with increase in concentration and concluded cypermethrin to be highly toxic to juvenile fish. *Labeo rohita* in toxic media exhibited erratic and darting movements with imbalanced swimming activity, which might be due to the malfunctioning of neurotransmitters, followed by hyper and hypo opercular activity, loss of equilibrium, and mucus secretion all over the body, upon sublethal exposure of cypermethrin (Marigoudar et al. 2009). *L. rohita* exhibited erratic and darting movements with imbalanced swimming activity on exposure to sublethal dose of cypermethrin, which might be due to the malfunctioning of neurotransmitters, followed by hyper and hypo opercular activity, loss of equilibrium, and mucus secretion all over the body (Marigoudar et al. 2009). Furthermore, Shalwei et al. (2012) reported abnormal behavioral responses such as rapid gill movement, nervous manifestations, erratic swimming, loss of equilibrium and inability to remain upright in Caspian Roach (*Rutilus rutilus caspicus*) and Silver carp (*Hypophthalmichthys molitrix*) due to cypermethrin exposure in a dose- and time-dependent manner. Likewise, Cypermethrin adversely affected behavioral patterns, shifting aerobic pathway of fish respiration towards anaerobic pathway and also inhibiting energy production by suppressing ATP synthesis as reported in *L. rohita* (Tiwari et al. 2012).

#### Amelioration of cypermethrin toxicity in fishes

Plant extract of *Datura*, *Datura stramonium*, might be useful in counteracting some of the negative effects of cypermethrin (Das and Mukherjee 2003). Dietary supplementation of ascorbic acid counters stress exerted by cypermethrin on freshwater catfish, *Heteropneustes fossilis* (Saha and Kaviraj 2009, 2013). Similar recuperating ability of ascorbic acid against cypermethrin-induced toxicity in Nile tilapia, *Oreochromis nilotica*, was observed by Korkmaz et al. (2009). Vasantharaja et al. (2012) have reported recovery of cypermethrin-induced damage in *C. mrigala* by dietary supplement of balloon vine, *Cardiospermum helicacabum* leaf powder. Additionally, Meenambal et al. (2012) demonstrated that inclusion of *Delonix elata* as feed supplement acted as a chelating agent and could reduce the toxic effect of cypermethrin in *Cyprinus carpio*.

#### Fenvalerate

Fenvalerate, first developed by Sumitomo in 1973 and introduced commercially in 1976 as an emulsifiable concentrate, is one of the potent pyrethroid insecticides widely used to control broad-spectrum insect pests. Environmental Protection Agency (EPA) classifies fenvalerate (Fig. 3) products under toxicity class II (*I* most toxic, *IV* least toxic), and includes the word WARNING on all product labels. Fenvalerate persists in the environment and is highly toxic due to its lipophilicity (Bradbury and Coats 1989b). Fenvalerate is the active ingredient in Pudrin, Sumicidin, Sumitox, Sumifly, Phenvalerate, Fenkill, etc. The pesticide is used primarily to control pests of cotton and vegetables (Madan et al. 2000). The physico-chemical properties of Fenvalerate have been represented in Table 5.



**Fig. 3** Chemical structure of Fenvalerate



**Table 5** Physico-chemical properties of fenvalerate

Variable	Information
Appearance	Pure isomers of cypermethrin form colorless crystals. When mixed isomers are present, cypermethrin is a viscous semi-solid or a viscous, yellow liquid
Chemical name	Cyano(3-phenoxyphenyl)methyl 4-chloro- $\alpha$ -(1-methylethyl) benzeneacetate; $\alpha$ -cyano-3-phenoxybenzyl 2-(4-chlorophenyl)-3-methylbutyrate; (RS)- $\alpha$ -cyano-3-phenoxybenzyl(RS)-2-(4-chlorophenyl)-3-methylbutyrate
Chemical formula	C <sub>25</sub> H <sub>22</sub> ClNO <sub>3</sub>
CAS number	51630-58-1
Molecular weight	419.92
Water solubility	0.01 mg/L @ 20 °C; insoluble in water
Solubility in	Methanol > 450 g/L.; acetone > 450 g/L; chloroform > 450 g/L, water 2 µg/L; Other solvents 85 µg/L
Melting point	Relatively stable to heat and moisture
Vapor pressure	$1.1 \times 10^{-8}$ mmHg mercury at 25 °C
Partition coefficient	6.2

Modified from Eisler (1992)

### Fenvalerate toxicity to fish

Fenvalerate belongs to the family of recently developed insecticides classed as synthetic pyrethroids, which exhibit very low avian and mammalian toxicity, but are very toxic to fish (Miyamoto 1976; Casida et al. 1983; WHO 1996). Fenvalerate is absorbed at a high rate through the gills of fishes due to its lipophilicity. However, fish have a poor ability to metabolise and excrete fenvalerate, since they seem to be deficient in the enzyme system that hydrolyzes pyrethroids (Bradbury et al. 1985; Haya 1989), and thus are susceptible to even minute concentration of the pesticide. Tilak et al. (2001a) conducted a study on residual concentration and toxicity of fenvalerate to some selected freshwater fishes and observed that *C. mrigala* is more sensitive to this pesticide followed by *Aplochielus panchax*, *L. rohita*, *C. catla* and *C. idella*. In this study quantitative residues confirmed the toxic action on different fresh water fishes. Datta and Kaviraj (2011) have reported that acute toxicity of fenvalerate remains persistent for a long duration in air-breathing cat fishes viz. *C. batrachus*, *Channa punctatus* and *Heteropneustes fossilis*. Recently, Satyavardhan (2013) reported comparative LC 50 values of fenvalerate for different freshwater fishes at different time intervals as well as for stagnant and continuous flowing water. LC50 values of fenvalerate to different fish species have been summarized in Table 6.

### Histopathological alterations

Histological methods remain the primary tools for the evaluation of pathological changes in tissues in toxicological studies and are gaining considerable attention while conducting sub-lethal exposure of different toxicant in aquatic organisms. Mandal and Kulshrestha (1980) observed liver necrosis, vacuolization and breakdown of cell boundaries in *C. batrachus* exposed to sub-lethal concentrations of Sumithion a fenvalerate-based product. Histopathological damages to gill surface of fishes due to fenvalerate have been attributed by many authors to high accumulations in gills, irritation due to elevated mucus secretion, increased ventilation volume and decreased gill-oxygen uptake efficiency (Bradbury et al. 1985, 1987; Bradbury and Coats 1989a, b). A number of pathological changes have been reported in *C. idella* exposed to synthetic pyrethroid fenvalerate (Tilak et al. 2001a, b; Tilak and Yacob 2002). Fenvalerate has also been reported to cause damage to the early life stages of fishes. Teh et al. (2005) found that exposing 7-day-old larvae of the fish, *Sarcamento splittail* to sublethal concentrations of esfenvalerate for 1 week induced vacuolar degeneration and cell necrosis in the liver. The hepatocytes of fishes exposed to fenvalerate for 10 days showed an obvious reduction in the protein contents and their remnants were mainly located at the peripheries of the hepatic cells which showed sever cytoplasmic vacuolation (Sakr et al. 2005). Similar histopathological alterations in the gill, kidney, liver and intestine tissues of *C. mrigala* were illustrated by Velmurugan et al. (2007). The most



**Table 6** Toxicity studies on the effects of fenvalerate on various fish species

Scientific name	LC 50 value	References
<i>Clarias batrachus</i>	0.6 mg/L (21 days)	Tripathi and Verma (2004)
<i>Clarias batrachus</i>	1.35 µg/L (96 days)	Datta and Kaviraj (2011)
<i>Clarias gariepinus</i>	250 µg/L (48 h)	Sakr et al. (2005)
<i>H. fossilis</i>	0.65 µg/L (96 days)	Datta and Kaviraj (2011)
<i>Ictalurus punctatus</i>	1.8–1.9 µg/L (24 h)	Mayer and Ellersieck (1986)
<i>Channa punctatus</i>	1.0 µg/L (96 days)	Datta and Kaviraj (2011)
<i>Channa punctatus</i>	2.13 µg/L (96 h)	Singh et al. (2007)
<i>Opsanus tau</i> (Gulf Toad Fish)	2.4 µg/L (96 h)	Clark and Brooks (1989)
<i>Cyprinodon variegates</i> (Sheep head minnow)	4.4–5.0 µg/L (96 h)	Mayer (1987) Schimmel et al. (1983) Hansen et al. (1983) Clark et al. (1985)
<i>Pimephales promelas</i> (Fat head minnow)	5.49 µg/L (96 h)	Bradbury and Coats (1989b)
<i>Pimephales promelas</i> (Fat head minnow)	1.69 µg/L (48 h)	Bradbury et al. (1987)
<i>Gambusia affinis</i>	15.0 µg/L (48 h)	Smith and Stratton (1986)
<i>Salmo salara</i>	1.2 µg/L (96 h)	McLeese et al. (1980)
<i>Oncorhynchus mykiss</i>	0.23–2.1 µg/L (96 h)	Bradbury and Coats (1989b) Clark et al. (1987) Mayer and Ellersieck (1986)
<i>Tilapia mosambica</i>	45.0 µg/L (48 h)	Radhah and Reddy (1989)
<i>Mugil cephalus</i>	0.58 µg/L (96 h)	Mayer (1987) Schimmel et al. (1983) Hansen et al. (1983)
<i>Mystus vitatus</i>	6.3 µg/L (96 h)	Verma et al. (1981)
<i>Cyprinus carpio</i>	21–30 µg/L (48 h)	Jagan et al. (1989) Reddy and Bashamohideen (1988)
<i>Catla catla</i>	6.0 µg/L (96 h)	Tandon et al. (2005)
<i>Labeo rohita</i>	5.36 µg/L (96 h)	Prusty et al. (2011)
<i>Cirrhinus mrigala</i>	6.0 µg/L (96 h)	Mushigeri and David (2004)
<i>Carps</i>	<0.1 mg/L (48 h)	WHO (1996)
<i>Oncorhynchus mykiss</i>	0.0036 mg/L (96 h)	WHO (1996)
<i>Cyprinus carpio</i>	2.171 mg/L (S) and 1.775 mg/L (C) (96 h)	Satyavardhan (2013)
<i>Puntius sophore</i>	1.789 mg/L (S) and 1.415 mg/L (C) (96 h)	Satyavardhan (2013)
<i>Ctenopharyngodon idella</i>	2.627 mg/L (S) and 2.121 mg/L (C) (96 h)	Satyavardhan (2013)
<i>Channa punctatus</i>	128.1 mg/L (S) and 110.7 mg/L (C) (96 h)	Satyavardhan (2013)
<i>Anabas testudineus</i>	472.5 mg/L (S) and 376.0 mg/L (C) (96 h)	Satyavardhan, 2013
<i>Cyprinus carpio</i>	3.059 (96 h)	Raja et al. (2010)
<i>Clarias gariepinus</i>	5.83–4.76 µg/L (24 h) and 7 4.24–2.94 µg/L (96 h)	Bhattacharya and Kaviraj (2009)

Here (S) stands for stagnant water, (C) for continuous water flow

common gill changes at all concentrations of fenvalerate were epithelial hyperplasia, epithelial necrosis, desquamation and lamellar fusion where as necrosis of tubular epithelium, pycnotic nuclei in the hematopoietic tissue, hypertrophied epithelial cells of renal tubules, narrowing of the tubular lumen, expansion of space inside the Bowman's capsule and contraction of the glomerulus were observed in kidney tissues of fish. Susan et al. (2012) observed drastic histopathological changes in vital tissue, i.e., gill, liver and kidney of three Indian Major Carps, *L. rohita*, *C. catla* and *C. mrigala* exposed to sub-lethal concentration of fenvalerate for 10 days.



## Biochemical and hematological alterations

Alterations in haemato-biochemical responses have been reported to be an important bioindicator to assess the toxicological changes in various fishes exposed to fenvalerate during the past few decades (Prusty et al. 2011). Sheela et al. (1992) observed that exposure of *Channa striatus* to 0.4–1.0 µg/L fenvalerate decreased the feed intake and nutrient absorption growth of the fish. Chronic exposures to the sub-lethal concentrations of fenvalerate increased mortality and reduced growth, HSI, SGR in *Clarias gariepinus* in a dose-dependent way (Datta and Kaviraj 2006). Bhattacharya and Kaviraj (2009) reported 24-h exposure of fenvalerate caused marked decrease in hepatosomatic index, liver glycogen, alkaline phosphates of liver and significant increase in plasma glucose level and hemoglobin percentage in *Clarias gariepinus*. Significant inhibition in growth and survival of *L. rohita* fingerlings upon short-term exposure of sublethal concentration of fenvalerate was observed in our previous study (Prusty et al. 2011).

Radhaiah et al. (1989) and Radhaiah and Reddy (1989) found fenvalerate-induced changes in the activity and isoenzyme pattern of lactate dehydrogenase (LDH) in the liver, gill, muscle and brain of the freshwater teleost, *Oreochromis mossambicus*. Fenvalerate has been reported to cause a significant reduction in activity of succinate dehydrogenase (SDH) and malate dehydrogenase (MDH), but induction of LDH activity in the liver of *Tilapia mossambica* (Radhaiah and Rao 1990). Shakoori et al. (1996) reported significant decrease in liver LDH activity after 3 weeks of sub lethal concentration of fenvalerate exposure to freshwater fish, *C. idella*, while Tripathi and Verma (2004) reported decrease in LDH activity in *C. batrachus* exposed to fenvalerate. David et al. (2005) reported alterations in lactate and succinate dehydrogenases activity in *L. rohita* upon fenvalerate exposure. Significant inhibition in the activity of antioxidant enzymes like super oxidase dismutase (SOD) and catalase activity in both liver and gill of carp, *L. rohita*, due to fenvalerate exposure was observed in our earlier study suggesting fenvalerate induced oxidative stress (Prusty et al. 2011).

Any change in the transaminase activity can be correlated with the protein and carbohydrate metabolism and thereby help in analyzing the metabolic shifts (Beyer et al. 1996). Fenvalerate exposure resulted in alteration in the enzymes of protein metabolism such as alanine and aspartate amino transaminase enzymes of *L. rohita* fingerlings (Prusty et al. 2011). Glucose-6-phosphate dehydrogenase activity is expected to increase with increased phagocytosis (Das 2002) as it converts Glucose-6-phosphate to 6-phospho-gluconolactone using NADP<sup>+</sup> and release NADPH. Activity of this pathway activity increases when requirement of NADPH increases. The NADPH generated is utilized by NADPH oxidase in producing superoxide anions for destroying phagocytosised material. Tripathi and Verma (2004) reported exposure to fenvalerate gradually decreased the activity of glucose-6-phosphate dehydrogenase in brain, liver and skeletal muscle up to 21 days. Similarly, fenvalerate-induced inhibition in AChE activity has been studied in *C. mrigala* (Mushigeri and David 2005) and *L. rohita* (Prusty et al. 2011).

A reduction of structural and soluble proteins in liver, brain and muscle was found in *C. carpio* following exposure to 10 µg/L of fenvalerate for 6–48 h (Reddy et al. 1991a). Decrease in protein content in tissues of *C. batrachus* exposed to fenvalerate was reported by Tripathi et al. (2002). Harmful biochemical effects of fenvalerate at sublethal concentrations have also been observed in some other swamp-inhabiting Indian fish like *Channa punctatus* (Seth and Saxena 2003) and freshwater pond bottom dwellers like *C. mrigala* (Mushigeri and David 2004). Freshwater fish rohu (*L. rohita*) when exposed to high fenvalerate concentrations, presented the maximum glucose level on the fourth day of exposure, w began to decline over time until depleted (David et al. 2005). Reddy et al. (1991b) assessed the increased amino acids and lactate in the tissue of *C. carpio* and *L. rohila* when exposed to sub lethal concentration of fenvalerate which causing metabolic diversion in fish to prolong its survivability under severe osmotic imbalance. Significant alterations in hematological characteristics and serum biochemical parameters of fenvalerate-exposed *L. rohita* fingerlings indicated fenvalerate-induced impairment of body metabolism and immunity (Prusty et al. 2011).

## Reproductive physiology of fishes

Synthetic pyrethroid, esfenvalerate, is reported to cause reduced fecundity and the failure of eggs to hatch among Australian crimson-spotted rainbow-fish, *Melanotaenia fluviatilis* (Castelnau), (Barry et al. 1995) and reduced fry growth, delayed spawning along with reduced hatching in bluegill, *Lepomis macrochirus* Raf (Tanner and Knuth 1996).

### Behavioral response of fishes

Fenvalerate is one of the pyrethroid insecticides and most widely used in agricultural crops such as cotton, paddy, jowar, maize, soyabean, tomato, lady's finger, cauliflower, tobacco and tea. But the use of this insecticide also tends to affect the biology of non-target species along with pests (Tilak et al. 2003; Tripathi and Verma 2004; Sakr et al. 2005; Velmurugan et al. 2007; Ramaneswari and Rao 2008; Majumdar and Gupta 2009). Anita et al. (2012) reported that early stages of Indian Major Carps are also affected by acute toxicity of Fenvalerate. Signs of fenvalerate poisoning in fish include loss of schooling behavior, swimming near the water surface, hyperactivity, erratic swimming, seizures, loss of buoyancy, increased gill mucus secretions, flaring of the gill arches, head shaking and listlessness before death (Bradbury and Coats 1989a, b). Fenvalerate mainly affects the teleost nervous system, as discussed earlier. It also produces osmoregulatory imbalance, as judged by altered calcium uptake (Symonik et al. 1989), abnormal sodium and potassium excretion rates and elevated urine osmolality (Bradbury et al. 1987; Bradbury and Coats 1989a, b).

### Amelioration of fenvalerate toxicity in fishes

Ascorbic acid is an important intracellular antioxidant and is involved in the self-defense mechanisms of fish. It works as an antitoxic agent against pesticide-induced stress (Guha et al. 1993). Dietary supplementation of ascorbic acid to ameliorate the toxic effects of the fenvalerate in freshwater catfish, *Clarias gariepinus*, has been reported by Datta and Kaviraj (2006). Ameliorative effect of vitamin C and E supplementation at higher doses against fenvalerate-induced stress has been observed by Prusty (2010). Also, Selvam et al. (2013) investigated the potential of the chosen bacterium, *Bacillus cereus* MTCC 1305, in the degradation of various concentrations of fenvalerate (250, 500, 750 and 1000 ppm) and suggested that the utilization of fenvalerate by *P. viridiflava* may be a feasible treatment option for the removal of pesticides from soil environment. Moreover, dietary supplementations of multispecies probiotic bacteria have apparently been helpful in counteracting fenvalerate-induced stress in a tropical freshwater fish, *L. rohita* (Mohapatra et al. 2012; De et al. 2014).

### Habitat alteration and biodiversity loss due to pesticide application

Fish and other aquatic biota may be harmed by pesticide-contaminated water. Pesticide surface runoff into rivers and streams can be highly lethal to aquatic life, sometimes killing all the fish in a particular stream (Toughill 1999). Pesticides can accumulate in bodies of water to levels that kill off zooplankton, the main source of food for young fish (Pesticide Action Network North America 1999). Pesticides can kill off the insects on which some fish feed, causing the fish to travel farther in search of food and exposing them to greater risk from predators. The faster a given pesticide breaks down in the environment, the less threat it poses to aquatic life. Insecticides are more toxic to aquatic life than herbicides and fungicides (Helfrich et al. 1996).

Spraying herbicides can also reduce reproductive success of fish and aquatic animals (Ansari and Waleem 2009). The shallow, weedy nursery areas for many fish species provide abundant food and shelter for young fish. Spraying herbicides near weedy nurseries can reduce the amount of cover and shelter that young fish need to hide from predators and to feed. In a series of different tests pesticide application has been shown to cause vertebral deformities in fish (Koyama 1996). The weed-killers Ronstar and Roundup are also acutely toxic to fish (Folmar et al. 1979; Shafiei and Costa 1996). The toxicity of Roundup is likely due to the high toxicity of one of the inert ingredients of the product (Folmar et al. 1979). In addition to direct acute toxicity, some herbicides may produce sublethal effects on fish that lessen their chances for survival and threaten the population as a whole. Glyphosate or glyphosate-containing products can cause sublethal effects such as erratic swimming and labored breathing, which increase the fish's chances of being eaten (Liong et al. 1988). 2,4-D herbicides caused physiological stress responses in sockeye salmon (McBride et al. 1981) and reduced the food-gathering abilities of rainbow trout (Little 1990). Several cases of pesticide poisoning of dolphins have been reported worldwide. Because of their high trophic level in the food chain and relatively low activities of drug-metabolising enzymes, aquatic mammals such as dolphins accumulate increased



concentrations of persistent organic pollutants (Tanabe et al. 1988) and are thereby vulnerable to toxic effects from contaminant exposures. The continued use of organochlorine pesticides and PCBs in India is of greater concern (Kannan et al. 1992, 1997a, b; Tanabe et al. 1988). The Ganges river basin is densely populated and heavily polluted by fertilizers, pesticides, and industrial and domestic effluents (Mohan 1986; Mutiyar and Mittal 2013). In addition to fish, other marine or freshwater animals are also endangered by pesticide contamination (Aktar et al. 2009). The landowner who sprays a weedy fenceline with herbicides may unintentionally kill the trumpet vine on which hummingbirds feed and the honeysuckle that nourishes deer and quail. Casual use of herbicides for lake or farm pond refinement may reduce fish populations (Masser et al. 2001).

Aquatic plants provide as much as 80 % of the dissolved oxygen necessary for aquatic life in ponds and lakes. Spraying herbicides to kill all aquatic plants can result in severely low oxygen levels and the suffocation of fish. Using herbicides for complete “clean up” of a pond will significantly reduce fish habitat, food supply, dissolved oxygen and fish productivity (Helfrich et al. 1996). Moreover, deltamethrin has a general effect on the ATP concentration that renders dysfunction in normal physiological activities of fish (Kumar 2011). Widenfalk et al. (2004) used microcosm experiments to show that deltamethrin and pirimicarb inhibited the bacterial activity, whereas deltamethrin also affected the microbial biomass negatively.

### Endocrine disruption in fish

Fish and other organisms are especially vulnerable to endocrine disrupting effects during the early stages of development (Jobling and Tyler 2003). Biological effects in wild freshwater fish that have been attributed to the effects of endocrine disruptors include the inappropriate production of the blood protein vitellogenin, altered growth and development (Vos et al. 2000). The pyrethroid insecticides have been shown to affect mechanisms involved in fish reproduction (Barry et al. 1995; Tanner and Knuth 1996; Moore and Waring 2001). Pesticides at low concentrations may act as mimics or blockers of sex hormones, causing abnormal sexual development, feminization of males, abnormal sex ratios and unusual mating behavior. The unique plasticity of sex differentiation in fish suggests that these animals may be very susceptible to disruption of sexual characteristics by pollutants. Pesticides can also interfere with other hormonal processes, such as thyroid functioning and bone development (Ewing 1999). The acute exposure of rainbow trout and common carp to the pyrethroids deltamethrin, cypermethrin, and bifenthrin was associated with alterations in hematological and biochemical indices as well as in tissue enzymes, resulting in stress to the organism. (Velisek et al. 2007, Velisek et al. 2006a, b). These pyrethroids are, therefore, classified as belonging to substances strongly toxic for fish. Cypermethrin reduces the fertilization success in Atlantic salmon (*Salmo salar*) as reported by Richterova and Svobodová (2012). It inhibits ability of male salmon parr to detect and respond to the female salmon priming pheromone PGF<sub>2</sub>α. The increase in expressible milt and the levels of plasma sex hormones are reduced in the presence of the pyrethroid as the result of impaired olfactory detection of the priming pheromone (Moore and Waring 2001). Ansari et al. (2009) suggested that oxidative stress may, in part, be contributing to deltamethrin-induced genotoxic damage to erythrocytes, as observed in fresh water biomarker species *Channa punctatus*.

### Summary

Retrospection of relevant literatures suggest that Type II synthetic pyrethroid insecticides cause alterations in the metabolic processes, hematology, enzymatic activity and reproductive physiology of fishes providing evidence for ecological disturbances in the natural environment due to unintentional spreading of insecticides (Murthy et al. 2013). Given its potential impact, prudence should be used when applying these insecticides to or near water bodies. Extreme care must be taken to avoid or mitigate drift and runoff of these chemicals to surface water bodies. Furthermore, agricultural pesticides are often used in different combinations to protect crops (Aktar et al. 2009), so risk from the joint toxicity of these chemicals to aquatic environment is apparent. There is limited information available regarding synergistic toxicity of these insecticides in fishes. Ecotoxicological information of the transformed or degraded products of the insecticides is also scarce. In order to protect the environment, future research should include not only direct effects of single parent compounds, but



also indirect effects in the presence of other chemicals, including possible pesticide transformation products. Though few studies have highlighted the ameliorative effect of certain vitamins like vitamin C and E, carotenoids like lycopene nutraceuticals like yeast, beta glucan, pyridoxine, microbial levan, probiotic bacteria and certain plant extracts against pesticide-induced stress, additional research is needed in this regard. This will help in formulating regulations with respect to use of hazardous pesticides and preparing appropriate management options for addressing the menace of pesticide contamination of aquatic environment. Based on laboratory data on acute toxicity of pyrethroids against nontargets, the relatively newer photostable and more effective pyrethroids have been ranked, in order of decreasing toxicity, as follows: fenvalerate < cypermethrin < deltamethrin. In general, pyrethroids are more toxic to nontarget aquatic insects and crustaceans than to other phylogenetically distant invertebrates (Mian and Mulla 1992). The impact of these transient effects of pyrethroids on nontarget species will have a short-term bearing on the densities of dependent carnivorous fish species in aquatic ecosystems (Mian and Mulla 1992). Approaches using molecular biology techniques will revolutionize toxicological applications that are cheaper and do not require the use of animals to detect environmental stressors (Murthy et al. 2013). Besides, for safer use of these pesticides, more experimental work should be performed to determine the concentration and time of exposure that do not induce significant sub-lethal effects on fish. Pesticides containing copper appear to behave unpredictably with other compounds in invertebrates and there is evidence to suggest that copper may affect metabolism that could alter the toxicity of other pesticides (Dennis et al. 2012). Therefore, compounds containing copper should be treated with caution when assessing any interactions. The data on environmental-cum-health risk assessment studies may be regarded as an aid towards a better understanding of the problem. Data on the occurrence of pesticide-related illnesses among defined populations in developing countries are scanty (Aktar et al. 2009). Generation of base-line descriptive epidemiological data based on area profiles, development of intervention strategies designed to lower the incidence of acute poisoning and periodic surveillance studies on high-risk groups are needed.

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