International Journal of Zoology and Applied Biosciences **ISSN: 2455-9571**
Volume 8, Issue 6, pp: 51-61, 2023
https://doi.org/10.55126/ijzab.2023.v08.i06.008
http://www.ijzab.com

Review Article

INSIGHTS INTO THE ECOTOXICOLOGICAL EFFECTS ASSOCIATED WITH IMIDACLOPRID: A REVIEW

[a,b]*Stephy Rose K.V, [a]Niladri Sekhar Chatterjee, [a]Suseela Mathew, [a]Anandan Rangasamy

[a]ICAR-Central Institute of Fisheries Technology, Matsyapuri P.O., Cochin - 682029, Kerala, India
[b]Faculty of Marine Sciences, Cochin University of Science and Technology, Cochin - 682016, Kerala, India

Article History: Received 24th October 2023; Accepted 20 November 2023; Published 31st December 2023

ABSTRACT

Imidaclopridis a neonicotinoid insecticide contently used in agricultural fields with excellent systemic and contact activity, used in the largest volume worldwide against sucking pests of Diptera, Coleoptera and Lepidoptera in chilly, cotton, grapes, groundnut, okra, paddy, sugarcane, sunflower and tomato. It functions as an agonist at the acetylcholine receptors of the pest, affecting invertebrate movements, leading to palsy and mortality. It has an approbative toxicity profile, due to its poor penetration of the blood–brain barrier in vertebrates. Moreover, it does not exhibit any mutagenic, carcinogenic, teratogenicor immunotoxic properties. Besides these boons of imidacloprid, several studies reported the high leaching potential and persistence of imidacloprid in the ecosystem, creating threat to non-targeted organisms by altering their biochemical and reproductive processes.

Keywords: Imidacloprid, Properties, Environmental fate, Toxicity.

INTRODUCTION

Imidacloprid is the most renowned and widely used neonicotinoid registered for agricultural and residential usages since past three decades. It is contently used for crop protection, termite control and dermal application on animals. The excellent biological properties, mainly a low application rate, wide spectrum and quick uptake make imidacloprid more promising. Imidacloprid is marketed under trade names Advantage, Admire, Confidor, Hachikusan, Kohinor, Merit, Gaucho, Premise, Prothor, and Winner. The neonicotinoids were brought to light in the 1970s to control pests, by chemists at Shell Chemical Company, while evaluating the insecticidal potential of heterocyclic nitromethylenes (Schroeder and Flattum, 1984).The term "neonicotinoid" was introduced to distinguish from the nicotinoids, as the former is highly efficient as insecticides and less toxic to vertebrates than the later. In 1984, chemists at Nihon Bayer Agrochem discovered a moiety named imidacloprid (CAS 138261-41-3), with greatly increased insecticidal activity, maintaining its photostability, while working with a 3-pyridylmethyl group on the nitromethylene heterocycle structure (Shiokawa *et al*., 1994). It was first enrolled as an insecticide for agricultural application since 1994 in the U.S.

Structure

Neonicotinoid is structurally 6-chloro-3-methylpyridine with a pharmacophore. Neonicotinoids are either nitroguanidines (C = NNO2), nitromethylenes (C = CHNO2), or cyanoamidines (C = NCN) (Compounds with 3-tetrahydrofuranmethyl, 2-chloro-5-thiazolylmethyl and 6-chloro-3-pyridinylmethyl moieties are called tefuryl, chlorothiazolyls or thianicotinyls and chloropyridinyls or chloronicotinyls, respectively. The pharmacophore of the nitroguanidine insecticide imidacloprid is nitroiminoimidazolidine. Imidacloprid, known aschloropyridinyls or chloronicotinyls or 1- (6 –chloro- 3 -pyridylmethyl) N – nitroimidazolidin-2-ylideneamine is anitroguanidine insecticide of neonicotinoid family, with a unique structure (Figure 1). It was acquired by the combining reaction of 2-chloro-5-chloromethyl-pyridine with the 2-nitro-imino-imidazolidine, in acetonitrile with potassium carbonate as the base. The nitrogen atom of the

*Corresponding Author: Stephy Rose K.V, ICAR-Central Institute of Fisheries Technology, Matsyapuri P.O., Cochin -682029, Kerala, India. Email: stephyrose512@gmail.com.

chloropyridine moiety of imidacloprid reacts with the hydrogen donor –nicotinic acetylcholine receptors (nAChR), and the nitrogen atom at the 1-position of the imidazolidine 5-ring reacts with a negatively charged domain (Okazawa *et al*., 2000). Binding of imidacloprid to the nicotinic receptor is irreversible, which results in overstimulation of the neurons.

Several other chemical analogues of imidacloprid such as acetamiprid, nitenpyram, thiacloprid, etc. were also developed for commercial use soon after the discovery of imidacloprid. Later, a "second generation" of neonicotinoidswere introduced by replacing the chloropyridinyl moiety with a chlorothiazolyl group toreduce its activity atmammalian receptors. The major members of the second generation includes Thiamethoxam and Clothianidin. Further research on the compound lead to the discovery of Dinotefuran with a characteristic Acetylcholine moiety without a pyridine ring, which started the era of "third-generation" neonicotinoids. Other members of the third generation neonicotinoids include Sulfoxaflor and Cycloxaprid.

Physical and Chemical Properties

Understanding the physical and chemical properties of imidacloprid (Table 1) is important to know the biochemical interactions of imidacloprid with biotic and abiotic systems, which in turn helps in comparison of toxicological data and interpretation of future research.

Usage of Imidacloprid

Global annual trade of neonicotinoids reaches up to 1000 million dollars, contributing 11%–15% of the entire insecticide merchandise. Neonicotinoids are licensed for use in more than 120 countries, of which imidacloprid holds about 41% of the total neonicotinoid market. Imidacloprid is contently used for the pest in agricultural fields, flea control and termite control. The primary target of imidacloprid includes sucking pests (aphids, whiteflies, and leafhoppers). Excellent crop tolerance of imidacloprid allows its use on crops viz., chilly, cotton, grapes, groundnut, okra, paddy, sugarcane, sunflower and tomato (CSI, 2020). It is now considered a possible replacement for the insecticides under phase revocation due to its quick action on pests even at low doses. However, their large-scale use has raised growing concerns about their potential adverse effects on non target invertebrates. As a result, in December 2013, the European Union banned the use of imidacloprid for seed coating and soil treatment. Nevertheless, recently Environmental Protection Agency (EPA) announced that its use continues to be approved (US EPA, 2020). It used to prevent sucking insects on crops and seeds (Furlan & Tof- fanin, 1998). It used to prevent sucking insects on crops and seeds (Furlan & Tof- fanin, 1998).

Mode of application

Although imidacloprid could be applied by diverse methods viz., sprays, microcosm, glass plate treatment, leaf dip of cotton, etc., the best mode of application is as a foliar spray. Imidacloprid is applied in paddy fields at a concentration of 0.003 ppm at a rate of 0.561 kg /ha. A major part of the imidacloprid applied is emitted to the environment causing toxic effects in non-targeted organisms (Naiel *et al*., 2020). EPA categorizes neonicotinoids as both class II and class III toxicity agents labeled with "Warning" or "Caution"signs.

Neuromodulatory action of imidacloprid

Versatile properties of imidaclopridare increasing its usage than other insecticides. Imidacloprid is well-referred to as "chloronicotinyls" to highlight insecticidal activity of the chlorine atom. It functions as both contact and stomach poison. It binds to the$\alpha4\beta2$ subtype of then AChRs (Figure 2.) in insects interfering with the nerve impulse resulting in twitching, muscle weakness and cramps, leading to paralysis, starvation due to impaired feeding and death (Alexander *et al*., 2007).It was found highly effective on insects and less toxic to vertebrates especially mammals (Tomizawa and Casida, 2005). Imidacloprid is categorized as moderately harmful (Class II WHO; toxicity category II EPA).

Environmental dissemination

Pesticides could be released into the environment (Figure 3), as the ground water moves depending on their physical, chemical, and biological properties. Farm level studies by Tomizawa & Casida (2005) have shown that the application of imidacloprid into the crop field is imposing major threat to the environment as 80 to 98.4% of applied pesticide leaching into the surroundings. The high solubility of imidacloprid in water is directly proportional to its great leaching potential, which leads to incredible persistence in soils and aquatic sediments. Moreover, its half-life of up to one year poses a greater risk of contamination not only to riverine water bodies, but also a greater extent to groundwater system (Diaz *et al*., 2017). The documentary reports of Environmental Protection Agency (1993) have indicated that imidacloprid is labelled as a category I pesticide along with 13 other turf insecticidesdue to its higher leaching potential from the soil environment into the aquatic system. Investigations by Rouchaud *et al*. (1994) had also demonstrated that around 97% of the imidacloprid applied on sugar beet seeds were found to be leached to the soil within 67 days after planting. A comparative study on the leaching potential of pesticides has indicated that imidacloprid is found to be the most leached pesticide into the environment as compared to other different pesticides, which includes chlorpyrifos, diazinon, diuron, etc. Other interest findings by Felsot *et al*. (1998) have shown that imidacloprid applied in a hop field drip irrigation system was detected at a maximum depth of 105 cm within 7 days after application.

Experimental investigations by Bonmatin *et al*. (2005) in different soil types showed that 97% out of 33 soil samples got exposure to imidacloprid coated seeds were found to retain the pesticide residues even after 1 or 2 years, indicating the persistence capability and non-

degradable nature of imidacloprid in the environment. Studies by Kreuger *et al*. (2010) on half-life of imidacloprid demonstrated that the shortest half-life of imidacloprid recorded was 107 days in Georgia in turf-covered soil. Interestingly, it was noticed that the concentration remained the same even after one year after treatment in cornfield soil in Minnesota (Kreuger *et al*., 2010). Furthermore, a field study by Tyor & Harkrishan (2016) reported that 39% of the water samples collected from the greenhouse farming system of the experimental locations was found to be positive for the pesticide residue. Thus application of imidacloprid in the soil is a serious water contaminant concern, which resentfully influence whole aquatic ecosystem.

Factors affecting leaching property of imidacloprid includes its formulations, adjuvants, surfactants, etc. A comparative investigation by Gupta *et al*. (2002) on the leaching property of different formulations of imidacloprid had shown that soluble concentrate has the highest potential to leach through the soil than analytical grade and water-dispersible powder. A field study by Jemec *et al*. (2007) demonstrated that the distribution of imidacloprid in the environment is promoted by high alkalinity of water, low soil sorption, low octanol-water partition coefficient, hydrolysis and stable soil degradation. A recent study by Pang et al. (2020) revealed that the biodegradation (Figure 4.) is one of the most important processes controlling the fate and transformation of imidacloprid. Nevertheless, an earlier report by Tisler *et al*. (2009) had shown that the biodegradation of imidacloprid is relatively slow in aquatic environments.

Although imidacloprid is subjected to active photodegradation, Liu *et al*. (2006) had stated that this occurs only in well-sunlit water surfaces and the photodegradation and hydrolysis of imidacloprid produce imidacloprid–urea as an intermediate compound, which is a potent toxicant. Interestingly, imidacloprid-urea was found to be the predominant metabolite followed by olefine, nitrosimine, 6-chloronicotinic acid, 5-hydroxy and nitroguanidine. The use of imidacloprid in the agricultural fields had been deprived by the European Commission since 2013. According to the EFSA report, imidacloprid causes severe environmental hazards even at low concentrations, regardless of its boons, including its unique structure, target specificity and safety for its user.

Dissipation in the paddy field

Several researchers (Thuyet *et al*., 2011a, 2011b; La *et al*., 2015; Pereira *et al*., 2017) have investigated the degradation of imidacloprid in the paddy fields under different environmental conditions. Dissipation of imidacloprid in paddy field was found to be biphasic first-order kinetics in the water, while single-phase first-order kinetics in soil (Thuyet *et al*., 2011b).The highest recorded concentration of imidacloprid in surface soil and water collected from different paddy fields were 440 µg/kg and 60 µg/l, respectively. (Thuyet *et al*., 2011a, 2011b; La *et al*., 2015; Pereira *et al*., 2017).Study by La *et al*. (2015) has indicated that around 21 - 68 % of the total applied mass of imidacloprid was leached into the surrounding aquatic system. The estimated concentration of imidacloprid was found to be 8.8 µg/Lin the irrigation canal (Pereira *et al*., 2017) and 83 µg /L in afflux of the stream (La *et al*., 2015).

Environmental monitoring of imidacloprid and its metabolites by Akoijam & Singh (2014) has shown that residues of imidacloprid metabolites were found to be present upto 90 days, whereas imidacloprid could be detected only upto 60 days. The estimated DT_{50} of imidaclopridin water and soil of paddy field ranged between 1.3-3.4days and 11.0-229 days, respectively (Thuyet *et al*., 2011 a & b; Pereira *et al*., 2017). Thuyet *et al*.(2011a) reported the biphasic dissipation of imidacloprid in water as 2.0 - 2.4 days during the initial phase (0 - 7 days) and 8.0 - 20.5 days in the later phase (7 - 35 days). Also Thuyet *et al*., (2011b) recorded faster degradation of imidacloprid at a higher pH (pH 10) with a DT_{50} of 44.7 days. Previously, studies by Fossen (2006) had reported the highest DT_{50}value of 229 days for imidaclopridin soil. Experimental field study in rice plot conducted by Daam *et al*. (2013) revealed that application of imidacloprid induced 48 h-EC_{50} immobility ¼84 mg/L for *Daphnia magna* and 6 d-EC_{50} growth inhibition¼0.01–0.015 mg/L for sediment-dwelling ostracod *Heterocypris incongruens*. This observation was in accordance with an earlier reported study (Sánchez-Bayo and Goka, 2006a), which recorded a significantly reduced abundance of both aquatic and terrestrial organisms in paddy fields with presence of imidacloprid residues at a level greater than 1 µg/L.

Table 1. Physical and chemical properties of Imidacloprid.

Chemical formula	**$C_9H_{10}ClN_5O_2$**
Molar mass	255.66
Molecular weight	255.7 g/mol
Appearance	Colourless crystals
Vapour Pressure	$2x10^{-9}$ hPa at 20 ^{0}C
Melting point	136.4 to 143.8 ^{0}C
Density	1.54 g/cm³
Solubility in water	0.514 g/L at 20 ^{0}C
Stability	Stable to hydrolysis at pH 5-11
Log K_{ow}	0.57

K_{oc}	260 mL/g
Leachability	Moderate
Half life in water	0-365 days
Half life in soil	17-6931 days

Kow = octanol water partition coefficient; *Koc* = organic carbon partition coefficient.

Ecotoxicology

Ecotoxicology serves an important role in addressing aquatic ecosystem health challenges in parallel with the rapid advancement in industry and technological gallops. Prolonged exposure to imidacloprid affects non-target organisms in aquatic ecosystems. Toxicity study by Zeid *et al*. (2019) demonstrated that the aquatic organisms are highly perceptive to imidacloprid contaminated ecosystems because of their propensity to accumulate contaminants in their body via their permeable skin, gills and other intrinsic sensitivities. An earlier research by Nyman *et al*. (2013) had also reported the potentiality of imidacloprid to cause lethality even at small concentrations by impairing motility and feeding in aquatic organisms. Metabolism of imidacloprid in organisms (Figure 5) preliminarily involves its oxidative cleavage to imidazolidine and 6-chloronicotinic acid. The former product gets excreted through urine and the later gets excreted as hippuric acid conjugate after degradation. The second route of imidacloprid metabolism includes hydroxylation and formation of an unsaturated metabolite. About 90% of the imidacloprid ingested is eliminated via urine (80%) and feces (20%) within 24 h and the total elimination occurs within 48 h (Sheets *et al*., 2016).

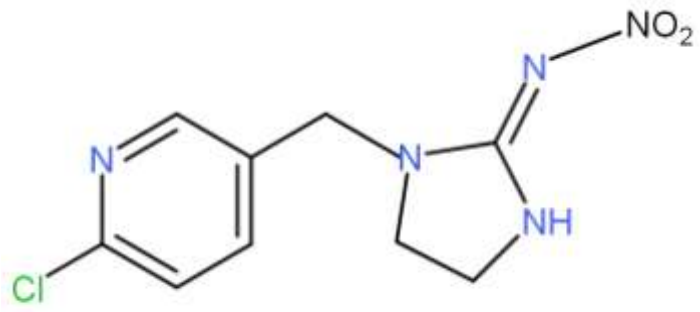

Figure 1. Structure of imidacloprid (Rose *et al*., 2022) K_{ow} = octanol water partition coefficient; K_{oc} = organic carbon partition coefficient.

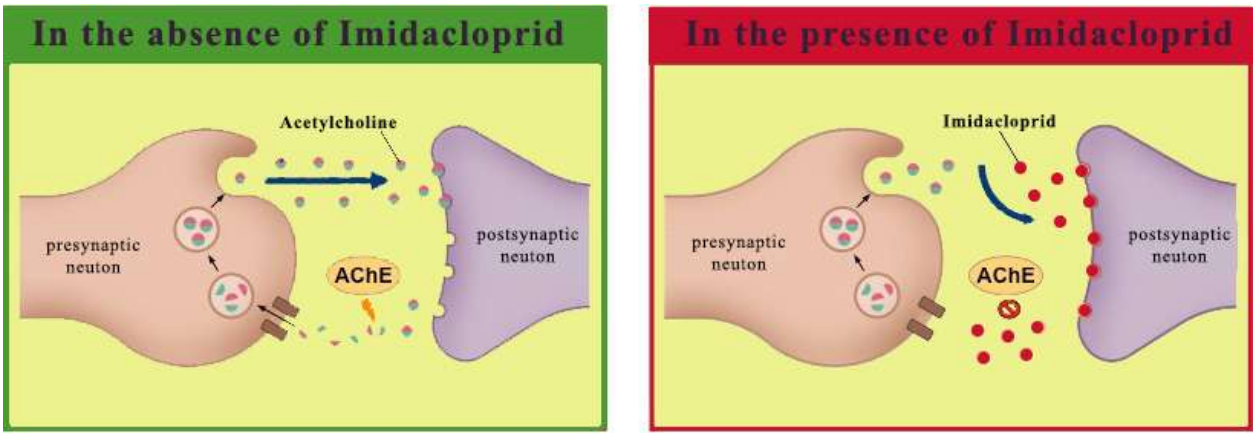

Figure 2. Neuromodulatory action of imidacloprid.

Acute toxicity

Acute toxicity studies are inevitable in evaluating the hazardous nature of environmental contaminants as it throws light on the health status of aquatic organisms in the contaminated ecosystem (Rose *et al*., 2020). Nyman *et al*. (2013) had evaluated the toxicity of imidacloprid inaquatic crustaceans and recorded theLC_{50} values of *Hyalella azteca* and *Mysidopsis bahia* as 55 ppb and 37 ppb, respectively. Another study by Sánchez-Bayo and Goka, 2006b reported48 h LC_{50} value of imidacloprid for cladocerans and ostracods as 65–133 mg/L and 301–715 µg/L, respectively and values of EC_{50} as 2-6 mg/L and 3-16 µg/L, respectively. Chen *et al*., 2010 reported LC_{50} of imidacloprid as 2.1 µg/l in *Ceriodaphnia dubia*. Organisms that survived the exposure showed behavioral alterations such as, lethargy and loss of equilibrium. Records of EPA (1992) had shown that the exposure to imidacloprid at a very low concentration had reduced the growth rate, body size and fecundity in mysid shrimp. Another study reported a reduced number and diversity of invertebrate species in artificial ponds at a concentration of 5 ppb (EPA, 1992).

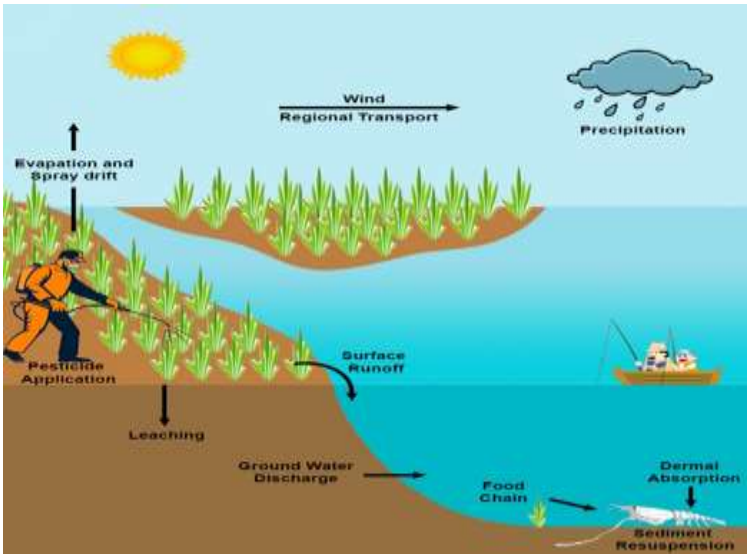

Figure 3. Environmental dissemination of imidacloprid.

The 96 h median lethal concentration of imidacloprid were determined for several fishes viz. *Labeo rohita* by Qadir *et al*. (2014) as 550 mg L^{-1}, Tilapia by Acar *et al*.(2018) as 141.42 mg L^{-1}, *Oncorhynchus mykiss* and *Cyprinus carpio* by Tisler *et al*. (2009) as 211 and 280 mg L^{-1} respectively. Su *et al*. (2007) investigated the toxicity of imidacloprid in *Paralichthys olivaceus* and revealed the median inhibitory concentrations (IC_{50}) of endpoint bioassays viz., neutral red (NR), 3-(4, 5-dimethylthiazol-2-yl)- 2, 5-diphenyltetrazolium bromide (MTT) and total cell protein (TCP) as 41.86, 38.46, and 39.08 µg/ml, respectively. They also found out that the mitochondria are the prime site of action of imidacloprid as the ultra structural observation of the gills cells exposed to 60 µg/ml of imidacloprid for 48 h showed severely damaged mitochondria and normal nuclei and rough endoplasmic reticulum. Furthermore, Sanchez Bayo & Goka (2005) reported Stress syndrome in juvenile *Oryzias latipes*, Xia *et al*. (2016) demonstrated neurobehavioral disorders such as reduced swimming, loss of balance, etc. in *Misgurnus anguillicaudatus* and Crosby *et al*. (2015) studied toxicity of imidacloprid in various life stages of the *Danio rerio* viz., larvae, fingerlings and adult. Desai & Parikh (2013) illustrated that *Oreochromis mossambicus* and *Labeo rohita* exposed to sublethal levels of imidacloprid showed severe degeneration in the liver, biochemical alterations in the gills, muscle and kidney. Besides, Tyor & Harkrishan (2016) reported that imidacloprid is potent to reduce the viability and hatchability of embryos of *C. carpio*, even in low concentrations (10% LC_{50}).

Biochemical alterations

Biochemical changes have prognostic value as it precedes the clinical manifestations of a disease situation. It broadens our understanding of how alterations in the chemical aspects of biological processes are related to physiological alteration in the body of an organism (Figure 6). Iturburu *et al*. (2018) have elaborated on the biochemical aberrations in organisms during stress. They demonstrated that organisms exposed to imidacloprid could induce oxidative damage, hypoglycemia and genetic anomalies. Laboratory study by Shan *et al*. (2020) revealed significant increase inantioxidant enzymes and MDA content in the gills and digestive glands of *Corbicula fluminea*. Sanchez-Bayo and Goka (2005) reported physiological stress in juvenile *O. latipes* treated with imidacloprid. Moreover, Vieira *et al*. (2018) revealed that the exposure to imidacloprid could reduce immunity and result in massive infestation by ectoparasite, *Trichodina* spp. In addition, Bonmatin *et al*. (2005) reported that

imidacloprid and its degradation products could be hazardous even at lower concentrations during chronic intoxication. Jemec *et al*. (2007) explained alterations in biochemical and reproductive parameters in *Daphnia magna* after long-term exposure to Imidacloprid. Another study by Priya *et al*. (2012) in freshwater teleost *Channa punctatus* reported significant elevation in the serum Glucose, Cholesterol, Creatinine and Creatine and decrease in the serum Protein, Albumin and Globulin after 96 h exposure to imidacloprid due to metabolic dysfunction in the fish. Qadir *et al*. (2014) had elaborated the sub-lethal effects of imidacloprid on the biochemical composition of *L. rohita*.

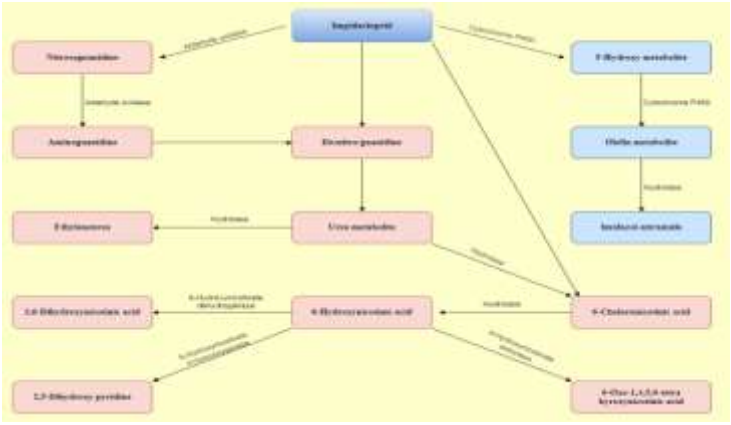

Figure 4. Biodegradation pathway of imidacloprid.

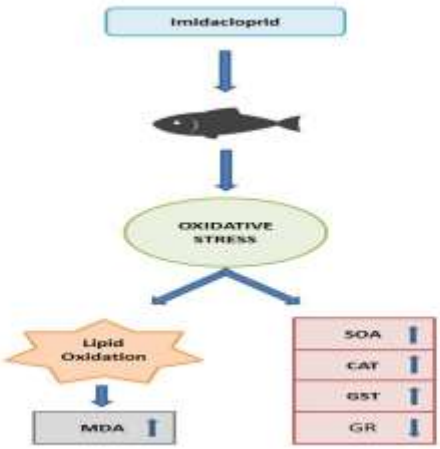

Figure 5.Metabolism of imidaclopridin organisms.

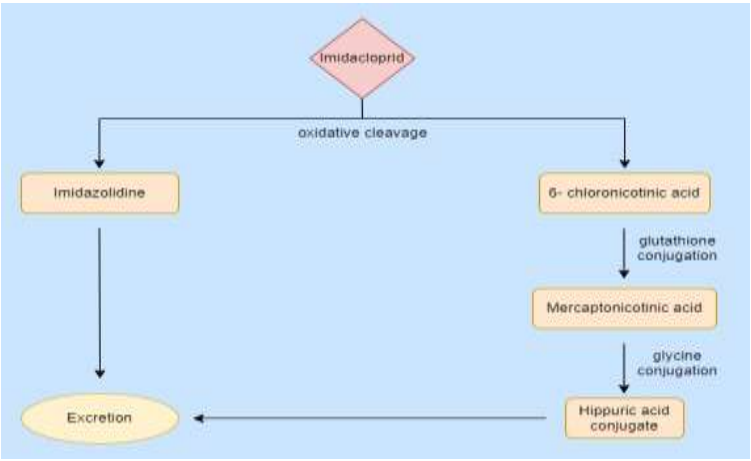

Figure 6. Oxidative stress caused by Imidacloprid.

Tripathi and Singh (2004) demonstrated that anorganism in pesticide contaminated ecosystem require high energy to detoxify the toxicants and to overcome the stress, physiological and histological alterations. Assessment of the biochemical contents, mainly protein could be used as an effective tool to understand the health status of an organism under stress (Prasanth & Arivoli, 2008). Experimental investigations by Xia *et al*., 2016 in *Misgurnus anguillicaudatus* and Desai & Parikh, 2013 in *O. niloticus* demonstrated that the exposure to imidacloprid cause hepatotoxicity. They found out that the alterations in the hepatic cell membrane permeability results in the loss of Alanine and Aspartateamino transaminases (ALT &AST) into the circulatory system, reducing the activation of aminotransferases in the liver. Several studies (Balint *et al*., 1995 and Singh *et al*., 2001) reported the increase in AST & ALT as a sign of tissue damage in the gill, kidney and liver. Hence, assessment of AST & ALT activity is of clinical and toxicological importance as its alterations are indicative of hepatic damage caused by pollutants or in diseased conditions.

Figure 7. Histopathological alterations during imidacloprid toxicity.

Table 2.Bioconcentration factor of imidacloprid in aquatic organisms.

Common name Scientific name	Exposure time	Tissue[a]	Exposure concentration	Bioconcentration factor[b]	References
zebrafish	14 days	NR	0.5 mg/L	1.52 mg/L	Ding *et al*., 2004
Brachydanio rerio	14 days	NR	5 mg/L	0.97 mg/L	Ding *et al*., 2004
Cichlid fish	48 h	Brain	300 mg/L	0.9± 0.3 L/kg	Iturburu *et al*., 2017
Australoheros	48 h	Blood	300 mg/L	0.6± 0.4 L/kg	Iturburu *et al*., 2017
facetus	48 h	Gills	300 mg/L	0.4 ±0.1 L/kg	Iturburu *et al*., 2017
	48 h	Muscle	300 mg/L	0.4 ±0.1 L/kg	Iturburu *et al*., 2017
	48 h	Gut	300 mg/L	1.3 ±0.3 L/kg	Iturburu *et al*., 2017
	48 h	Liver	300 mg/L	1.4 ±0.4 L/kg	Iturburu *et al*., 2017
Freshwater	1 day	NR	0.1 µg/L	66*	Contardo-Jara & Gessner, 2020
oligochaete	5 days	NR	0.1 µg/L	190*	Contardo-Jara & Gessner, 2020
Lumbriculus	1 day	NR	1 µg/L	28*	Contardo-Jara & Gessner, 2020
variegatus	5 days	NR	1 µg/L	119*	Contardo-Jara & Gessner, 2020
	1 day	NR	10 µg/L	26*	Contardo-Jara & Gessner, 2020
	5 days	NR	10 µg/L	75*	Contardo-Jara & Gessner, 2020

[a] Not reported (NR). [b] Unit not mentioned (*).

Researches (Pan & Dutta, 1998 and Huynh *et al*., 2009) proposed acetylcholinesterase activity as a biomarker to assess the pesticide toxicity. Rao *et al*. (2003) reported the inhibition of acetylcholinesterase activity in the brain of fish exposed to pesticides. Van der Wal & Welling, 1988 demonstrated that the inhibition of acetylcholinesterase activity could result in mortality of the organism. A recent study by Guerra *et al*. (2021) reported increased Glutathione S-transferase (GST) activity & carbonyl protein (CP), AChE activity in the brain and decreased locomotory movements in zebrafish after 96 h exposure to imidacloprid. Kappus (1987) reported Cholinergic hyperactivity instigated by the restraint of the AChE initiates the aggregation of reactive oxygen species (ROS), resulting in oxidative stress and lipid peroxidation, which lead to cell injuries. Sies (1986) defines oxidative stress as the disparity between the production and the elimination of ROS by the antioxidant system. McCarthy and Shugart (1990) have prescribed the use of antioxidant biomarkers for environmental monitoring programs. In addition, Cajariville *et al*. (2000) have reviewed the use of antioxidant biomarkers in assessing the environmental contaminants in the field studies. Oakes *et al*. (2004) have also revealed the importance of lipid peroxidation (LPO) as a predictive biomarkerin assessing pollution.

Young-Lai *et al*. (1991) recommended the osmoregulatory capacity of crustaceans as a potential tool that forecasts their physiological status during stress. Na^+/K^+ ATPase plays animportant role in branchial epithelial ion transport. Maintenance of a steady Na^+/K^+ gradient is necessary for the metabolic uptake of glucose & aminoacids, transmembrane movement of Calcium ions during muscle stimulation, regeneration of transmembrane potential and safeguard of osmotic equilibrium in cells via controlled transcellular ion movements. De La Torre *et al*. (1999) reported inhibition of Na^+/K^+ ATPase activity in fishes during short-term exposure to pollutants in the laboratory as well as field conditions. An investigation by Blažič *et al*. (2005) in *Porcellio scaber* observed feeding impairment and altered GST activity after 2 weeks of exposure to imidacloprid. Another experiment conducted in *P. scaber* by Drobne *et al*. (2008) reported alterations in survival rate, feeding rate, weight gain, GST, total protein content, and epithelial thickness of the digestive gland. Lukančič *et al*. (2010) investigated physiological responses of *Asellus aquaticus* and *Gammarus fossarum* to imidacloprid and reported increased respiration and decreased electron transport system activity.

Histopathology

Histopathology is a potent tool to analyze and describe the biological effects (Figure 7) of a toxicant on an organism. Histopathological alterations increase with the concentration of toxicant and exposure period. Shan *et al*. (2020) elaborated the histopathological alteration caused by imidacloprid in gills and digestive glands of *Corbicula fluminea*. Severe hemolymphatic vessel contractions and adhesions, moderate epithelial cellular swelling and damage of cilia were reported in gills. Furthermore, hemolytic infiltration of the connective tissue, severe degeneration of digestive tubules and necrosis of epithelial cells were observed in digestive glands of *C. fluminea*. The findings of Harkrishan *et al*. (2020) depicted several histopathological degenerations in the eyes (ruptured lens tissue and declined retinal pigmentation), gills (ruptured arterial wall, uplifted sclerotic layer and degenerated lamellae) and brain including necrosis, spongiosis, vacuolation, fragmented nuclei, pyknotic nuclei and mononuclear infiltration inhatchlings of *C. carpio*. Similar alterations were also observed by Naiel *et al*. (2020) in *Oreochromis niloticus* and Xia *et al*. (2016) in *Misgurnus anguillicaudatus.* Moreover, Qadir *et al*. (2014) reported moderate-to-severely damaged gills, heart, liver and kidney of *L. rohita*.

Bioconcentration

Bioconcentration refers to the direct transfer of chemicals from the surrounding environment into the organism via gills or other permeable membranes. The Bioconcentration of pesticides in aquatic organisms causes severe ecological problems. Hence bioconcentration is one of the essential tools in the pesticide toxicity assessment in aquatic organisms. Bioconcentration of several pesticides has been studied in different organisms (Sancho *et al*., 1998; Amrani & Pena - Abaurrea, 2012; Zhang *et al*., 2020). Bioconcentration factors give an estimate of the relative uptake of the chemicals from the environment by organisms. Bioconcentration factors for imidacloprid (Table 2.) had been calculated in several organisms including *Brachydanio rerio* (Ding *et al*., 2004), *Australoheros facetus* (Iturburu *et al*., 2017) and *Lumbriculus variegatus* (Contardo-Jara & Gessner, 2020).

CONCLUSION

Review on the ecotoxicological aspects of imidacloprid includes the baseline information regarding the neonicotinoid insecticideimidacloprid, its versatile uses, environmental fate and its toxicological effects on non-targeted organisms. Imidacloprid, being contently used globally at a large scale, with higher leaching potential than other widely used pesticides and persistence of more than a year causes potential risks for non-target organisms, especially aquatic invertebrates. Many researches portrayed the toxicity of imidacloprid on several beneficial species. This paper provides a critical revision of toxic sequel caused by imidaclopridin the biota and recommends to reduce the application of imidaclopridin agricultural fields by developing a better pest management practice that supports the sustainable development of biodiversity,to quench the increasing global food demand.

ACKNOWLEDGEMENT

The authors acknowledge Director, ICAR-Central Institute of Fisheries Technology (ICAR-CIFT), Cochin, Kerala, India for granting permission to publish this review article. The authors would like to express their sincere gratitude to

Cochin University of Science and Technology (CUSAT). The first author is thankful to University Grant Commission - Maulana Azad National Fellowship (UGC-MANF) for financial support. The last author is grateful to ICAR for providing funds under ICAR- National Fellow Scheme.

REFERENCES

Acar Ü, İnanan BE, Zemheri F, Kesbiç OS, Yılmaz S (2018). Acute exposure to boron in Nile tilapia (*Oreochromis niloticus*): Median-lethal concentration (LC50), blood parameters, DNA fragmentation of blood and sperm cells. *Chemosphere,* 213, 345-350.

Alexander, A.C., Culp, J.M., Liber, K., Cessna, A.J., (2007). Effects of insecticideexposure on feeding inhibition in mayflies and oligochaetes. *Environmental Toxicology and Chemistry*. *26*, 1726-1732.

Balint, T., Szegletes, T., Szegletes, Z., Halasy, K., & Nemcsók, J. (1995). Biochemical and subcellular changes in carp exposed to the *Organophosphorus methidathion* and the pyrethroid deltamethrin. *Aquatic Toxicology*, *33*(3-4), 279-295.

Blažič, M., Trebše, P., Drobne, D., (2005). Effect of imidacloprid on growth, feeding rate and activity of AChE and GST enzymes in the terrestrial isopods *Porcellio scaber* (Isopoda, Crustacea). *Zbornik predavanj in referatov. 7. Slovensko posvetovanje o varstvu rastlin, 8.-10 marec, 2005, Zreče, Slovenija*, 106-113. https://www.cabdirect.org/cabdirect/abstract/20153171319.

Bonmatin, J. M., Moineau, I., Charvet, R., Colin, M. E., Fleche, C., & Bengsch, E. R. (2005). Behaviour of imidacloprid in fields. Toxicity for honey bees. In *Environmental Chemistry*, 483-494.

Chen, X. D., Culbert, E., Hebert, V., Stark, J. D., (2010). Mixture effects of the nonylphenyl polyethoxylate, R-11 and the insecticide, imidacloprid on population growth rate and other parameters of the crustacean, Ceriodaphnia dubia. *Ecotoxicology and Environmental Safety, 73*(2), 132-137. https://doi.org/10.1016/j.ecoenv.2009.09.016.

Contardo-Jara, V., & Gessner, M. O. (2020). Uptake and physiological effects of the neonicotinoid imidacloprid and its commercial formulation confidor® in a widespread freshwater oligochaete. *Environmental Pollution*, 264, 114793.

Crosby, E. B., Bailey, J. M., Oliveri, A. N., & Levin, E. D. (2015). Neurobehavioral impairments caused by developmental imidacloprid exposure in zebrafish. *Neurotoxicology and Teratology*, *49*, 81-90.

CSI (Crop Science India). 2020. Crop and target pests of confidor, imidacloprid 200 SL.

Daam, M. A., Pereira, A. C. S., Silva, E., Caetano, L., & Cerejeira, M. J. (2013). Preliminary aquatic risk assessment of imidacloprid after application in an experimental rice plot. *Ecotoxicology and Environmental Safety*, *97*, 78-85.

De la Torre, F. R., Salibian, A., & Ferrari, L. (2000). Biomarkers assessment in juvenile *Cyprinus carpio* exposed to waterborne cadmium. *Environmental Pollution*, *109*(2), 277-282.

Desai, B., & Parikh, P. (2013). Biochemical alterations on exposure of imidacloprid and curzate on fresh water fish Oreochromis mossambicus and Labeo rohita. *Indian Journal of Forensic Medicine & Toxicology*, *7*(2), 87.

Diaz, J. M. C., Martin-Laurent, F., Beguet, J., Nogales, R., & Romero, E. (2017). Fate and effect of imidacloprid on vermicompost-amended soils under dissimilar conditions: risk for soil functions, structure, and bacterial abundance. *Science of the Total Environment*, *579*, 1111-1119.

Ding, Z., Yang, Y., Jin, H., Shan, Z., Yu, H., Feng, J., & Zhou, J. (2004). Acute toxicity and bio-concentration factor of three pesticides on Brachydanio rerio. Ying Yong Sheng tai xue bao. *The Journal of Applied Ecology*, 15(5), 888-890.

Drobne, D., Blažič, M., Van Gestel, C. A., Lešer, V., Zidar, P., Jemec, A., Trebše, P., 2008. Toxicity of imidacloprid to the terrestrial isopod *Porcellio scaber* (Isopoda, Crustacea). *Chemosphere*,71(7),1326-1334. https://doi.org/10.1016/j.chemosphere.2007.11.042.

EPA, (1992). (US EPA) US Environmental Protection Agency, 1992. Framework for Ecological Risk Assessment. EPA/630/R-92/001, Washington, DC.

Fossen M (2006) Environmental fate of imidacloprid. In: Environmental Monitoring, Department of Pesticide Regulation, California, pp 1-16.

Grădilă, M. (2013). Chronic aspects of imidacloprid on the fishes from Cyprinidae family. *Romanian Journal for Plant Protection*, *6*, 11-15.

Guerra, L. J., do Amaral, A. M. B., de Quadros, V. A., da Luz Fiuza, T., Rosemberg, D. B., Prestes, O. D., & Loro, V. L. (2021). Biochemical and Behavioral Responses in Zebrafish Exposed to Imidacloprid Oxidative Damage and Antioxidant Responses. *Archives of Environmental Contamination and Toxicology*, 1-10.

Gupta, S., Gajbhiye, V. T., & Agnihotri, N. P. (2002). Leaching behavior of imidacloprid formulations in soil. *Bulletin of Environmental Contamination and Toxicology*, *68*(4), 502-508.

Huynh, N. N., Harris, E. E., Chin- Dusting, J. F. P., & Andrews, K. L. (2009). The vascular effects of different arginase inhibitors in rat isolated aorta and

mesenteric arteries. *British Journal of Pharmacology*, *156*(1), 84-93.

Iturburu, F. G., Bertrand, L., Mendieta, J. R., Amé, M. V., & Menone, M. L. (2018). An integrated biomarker response study explains more than the sum of the parts: oxidative stress in the fish *Australoheros facetus* exposed to imidacloprid. *Ecological Indicators*, *93*, 351-357.

Iturburu, F. G., Zömisch, M., Panzeri, A. M., Crupkin, A. C., Contardo Jara, V., Pflugmacher, S., & Menone, M. L. (2017). Uptake, distribution in different tissues, and genotoxicity of imidacloprid in the freshwater fish Australoheros facetus. *Environmental Toxicology and Chemistry*, *36*(3), 699-708.

Jemec, A., Tišler, T., Drobne, D., Sepčić, K., Fournier, D., & Trebše, P. (2007). Comparative toxicity of imidacloprid, of its commercial liquid formulation and of diazinon to a non-target arthropod, the microcrustacean Daphnia magna. *Chemosphere*, *68*(8), 1408-1418.

Kappus, H. (1987). Oxidative stress in chemical toxicity. *Archives of Toxicology*, *60*(1-3), 144-149.

Kreuger, J., Graaf, S., Patring, J., & Adielsson, S. (2010). Pesticides in surface water in areas with open ground and greenhouse horticultural crops in Sweden 2008.

La, N., Lamers, M., Bannwarth, M., Van Nguyen, V., & Streck, T. (2015). Imidacloprid concentrations in paddy rice fields in northern Vietnam: measurement and probabilistic modeling. *Paddy and Water Environment*, *13*(2), 191-203.

Liu, W., Zheng, W., Ma, Y., & Liu, K. K. (2006). Sorption and degradation of imidacloprid in soil and water. *Journal of Environmental Science and Health Part, B*, *41*(5), 623-634.

Lukančič, S., Žibrat, U., Mezek, T., Jerebic, A., Simčič, T., Brancelj, A., (2010). Effects of exposing two non-target crustacean species, *Asellus aquaticus* L., and *Gammarus fossarum* Koch., to atrazine and imidacloprid. *Bulletin of Environmental Contamination and Toxicology*, 84(1), 85.https://doi.org/10.1007/s00128-009-9854-x.

McCarthy JF, Shugart LR. Biological markers of environmental contamination(1990). In: McCarthy JF, Shugart LR, editors. Biomarkers of environmental contamination. Boca Raton, FL: Lewis, 1990. p. 3 -14.

Naiel, M. A., Shehata, A. M., Negm, S. S., Abd El Hack, M. E., Amer, M. S., Khafaga, A. F., & Allam, A. A. (2020). The new aspects of using some safe feed additives on alleviated imidacloprid toxicity in farmed fish: A review. *Reviews in Aquaculture*, *12*(4), 2250-2267.

Nyman, A. M., Hintermeister, A., Schirmer, K., & Ashauer, R. (2013). The insecticide imidacloprid causes mortality of the freshwater amphipod Gammarus pulex by interfering with feeding behavior. *PloS one*, *8*(5), e62472.

Oakes, K. D., McMaster, M. E., & Van Der Kraak, G. J. (2004). Oxidative stress responses in longnose sucker (*Catostomus catostomus*) exposed to pulp and paper mill and municipal sewage effluents. *Aquatic Toxicology*, *67*(3), 255-271.

Okazawa A, Nakagawa Y, Akamatsu, M, Ueno T, Nishimura K. (2000). Comparison of binding activities of chloro-nicotinyl insecticides toward the nicotinic acetylcho-line receptors from rats and houseflies. *Journal of Pesticide Science*, 25, 40-43.

Pan, G., & Dutta, H. M. (1998). The Inhibition of Brain Acetylcholinesterase Activity of Juvenile Largemouth Bass Micropterus salmoides by Sublethal Concentrations of Diazinon. *Environmental Research*, *79*(2), 133-137.

Pang, S., Lin, Z., Zhang, Y., Zhang, W., Alansary, N., Mishra, S., & Chen, S. (2020). Insights into the toxicity and degradation mechanisms of imidacloprid via physicochemical and microbial approaches. *Toxics*, 8(3), 65.

Pereira, A. S., Cerejeira, M. J., & Daam, M. A. (2017). Ecological risk assessment of imidacloprid applied to experimental rice fields: Accurateness of the RICEWQ model and effects on ecosystem structure. *Ecotoxicology and Environmental Safety*, *142*, 431-440.

Priya, B.P., V. Rachel, Y.A. Maruthi (2012). Acute toxicity effect of imidacloprid insecticide on serum biochemical parameters of fresh water teleost, *Channa punctatus*. *International Journal of Science & Technoledge'*. 1(2), 18-22.

Qadir, S., Latif, A., Ali, M., Iqbal, F., (2014). Effects of imidacloprid on the hematological and serum biochemical profile of *Labeo rohita*. *Pakistan Journal of Zoology*, 46(4).

Rao, J. V., Pavan, Y. S., & Madhavendra, S. S. (2003). Toxic effects of chlorpyrifos on morphology and acetylcholinesterase activity in the earthworm, *Eisenia foetida*. *Ecotoxicology and Environmental Safety*, *54*(3), 296-301.

Rose, K. V. S., & Joseph, A. (2020). Acute Toxicity of Imidacloprid to Various Life Stages of the Giant Freshwater Prawn *Macrobrachium rosenbergii*, de Man, 1879.

Rose, S.K.V., Mahadevan, R., Anandan, R., Chatterjee, N.S., and Mathew, S. (2022). Evaluating the pressure of agrochemicals on the health and safety of polder based aquaculture in Kuttanad, Southwest coast of India.

Rouchaud, J., Gustin, F. and Wauters, A. (1996). *Bulletin of Environmental Contamination and Toxicology*, 56, 29-36.

Sánchez Bayo, F., & Goka, K. (2006a). Ecological effects of the insecticide imidacloprid and a pollutant from antidandruff shampoo in experimental rice fields. *Environmental Toxicology and Chemistry: An International Journal*, *25*(6), 1677-1687.

Sánchez-Bayo, F., Goka, K., (2005). Unexpected effects of zinc pyrithione and imidacloprid on Japanese medaka fish (*Oryzias latipes*). *Aquatic Toxicology*. 74(4), 285-293. https://doi.org/10.1016/j.aquatox.2005.06.003.

Sánchez-Bayo, F., Goka, K., (2006b). Influence of light in acute toxicity bioassays of imidacloprid and zinc pyrithione to zooplankton crustaceans. *Aquatic Toxicology*, 78(3), 262-271. https://doi.org/10.1016/j.aquatox.2006.03.009.

Sancho, E., Ferrando, M. D., Lleo, C., & Andreu-Moliner, E. (1998). Pesticide toxicokinetics in fish: Accumulation and elimination. *Ecotoxicology and Environmental Safety*, *41*(3), 245-250.

Schroeder, M. E., and R. F. Flattum. 1984. The mode of action and neurotoxic properties of the nitromethylene heterocycle insecticides. *Pesticide Biochemistry and Physiology*. 22, 148 -160.

Shan, Y., Yan, S., Hong, X., Zha, J., & Qin, J. (2020). Effect of imidacloprid on the behavior, antioxidant system, multixenobiotic resistance, and histopathology of Asian freshwater clams (*Corbicula fluminea*). *Aquatic Toxicology*, *218*, 105333.

Sheets, L. P., Li, A. A., Minnema, D. J., Collier, R. H., Creek, M. R., & Peffer, R. C. (2016). A critical review of neonicotinoid insecticides for developmental neurotoxicity. *Critical Reviews in Toxicology*, 46(2), 153-190.

Shiokawa, K., K. Yumoto, Y. Tanaka, T. Oguti, and Y. Kiyama (1994), Low-latitude auroras observed at Moshiri and Rikubetsu (L = 1.6) during magnetic storms on February 26, 27, 29, and May 10, 1992, *Journal of Geomagn. Geoelectr*, 46, 231-252.

Sies, H. (1986). Biochemistry of oxidative stress. *Angewandte Chemie International Edition in English*, *25*(12), 1058-1071.

Singh, S. N., Vats, P., Suri, S., Shyam, R., Kumria, M. M. L., Ranganathan, S., & Sridharan, K. (2001). Effect of an antidiabetic extract of *Catharanthus roseus* on enzymic activities in streptozotocin induced diabetic rats. *Journal of Ethnopharmacology*, *76*(3), 269-277.

Su, F., Zhang, S., Li, H., & Guo, H. (2007). In vitro acute cytotoxicity of neonicotinoid insecticide imidacloprid to gill cell line of flounder Paralichthy olivaceus. *Chinese Journal of Oceanology and Limnology*, *25*(2), 209-214.

Thuyet, D. Q., Watanabe, H., & Motobayashi, T. (2011a). Effect of formulations and treatment methods of nursery boxes applied with insecticide on the behavior of imidacloprid in rice paddy fields. *Journal of Pesticide Science*, *36*(1), 9-15.

Thuyet, D. Q., Watanabe, H., Yamazaki, K., & Takagi, K. (2011b). Photodegradation of imidacloprid and fipronil in rice–paddy water. *Bulletin of Environmental Contamination and Toxicology*, *86*(5), 548-553.

Tomizawa, M., Casida, J. E., (2005). Neonicotinoid insecticide toxicology: mechanisms of selective action. *Annual Review of Pharmacology and Toxicology*. 45, 247-268. https://doi.org/10.1146/annurev.pharmtox.45.120403.095930.

Tripathi, P. K., & Singh, A. (2004). Toxic effects of cypermethrin and alphamethrin on reproduction and oxidative metabolism of the freshwater snail, *Lymnaea acuminata*. *Ecotoxicology and Environmental Safety*, 58(2), 227-235.

Tyor, A. K., & Harkrishan, K. (2016). Effects of imidacloprid on viability and hatchability of embryos of the common carp (*Cyprinus carpio* L.). *International Journal of Fisheries and Aquatic Studies*, *4*, 385-389.

U.S. Environmental Protection Agency (EPA). 2020. Schedule for review of neonicotinoid pesticides. U.S. EPA, Washington, D.C., USA.

Van der Valk, A. G., & Welling, C. H. (1988). The development of zonation in freshwater wetlands: an experimental approach.

Vieira, C. E. D., Pérez, M. R., Acayaba, R. D. A., Raimundo, C. C. M., & dos Reis Martinez, C. B. (2018). DNA damage and oxidative stress induced by imidacloprid exposure in different tissues of the Neotropical fish Prochilodus lineatus. *Chemosphere*, *195*, 125-134.

Vignet, C., Cappello, T., Fu, Q., Lajoie, K., De Marco, G., Clérandeau, C., & Cachot, J. (2019). Imidacloprid induces adverse effects on fish early life stages that are more severe in Japanese medaka (*Oryzias latipes*) than in zebrafish (Danio rerio). *Chemosphere*, *225*, 470-478.

Xia, X., Xia, X., Huo, W., Dong, H., Zhang, L., & Chang, Z. (2016). Toxic effects of imidacloprid on adult loach (*Misgurnus anguillicaudatus). Environmental Toxicology and Pharmacology*, *45*, 132-139.

Young-Lai, W. W., Charmantier-Daures, M., & Charmantier, G. (1991). Effect of ammonia on survival and osmoregulation in different life stages of the lobster *Homarus americanus*. *Marine Biology*, *110*(2), 293-300.

Zeid, E. H. A., Alam, R. T., Ali, S. A., & Hendawi, M. Y. (2019). Dose-related impacts of imidacloprid oral intoxication on brain and liver of rock pigeon (*Columba livia domestica*), residues analysis in different organs. *Ecotoxicology and Environmental Safety*, *167*, 60-68.

Zhang, Y., Whalen, J. K., & Sauvé, S. (2020). Phytotoxicity and bioconcentration of microcystins in agricultural plants: Meta-analysis and risk assessment. *Environmental Pollution*, 115966.