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AGROECOLOGY | MINI REVIEW

Toxicological Effects of Chlorpyrifos Among Different Organisms

Atia Shanjida Shormi[1], Arafat Al Mamun[2], Sheikh Mahatabuddin[3], Khandaker Ashfaqul Muid[1]✉

[1] Department of Zoology, University of Dhaka, Dhaka 1000, Bangladesh
[2] Center for Advanced Research in Science, University of Dhaka, Dhaka 1000, Bangladesh
[3] Bangladesh Atomic Energy Regulatory Authority (BAERA), Agargaon, Dhaka 1207, Bangladesh

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Correspondence
Khandaker Ashfaqul Muid
✉: muid.zoo@du.ac.bd

ABSTRACT

Chlorpyrifos has been used as an insecticide with a long history of toxicity concerns for flora and fauna. However, the sensitivity and severity of toxicity vary greatly among the species. These toxic effects can lead to diseases, including problems with the brain and behavior, development, and reproductive health. For example, in rodents, Chlorpyrifos was found to be responsible for changes in neurotransmitter levels and results in modifications in motor activity, memory, and learning. However, it was reported in birds that this insecticide alters the frequency of egg production, hatching success, and other reproductive problems. Regardless of the adverse effect of Chlorpyrifos, it is still widely used in different areas including the agricultural sectors, particularly for pest control and mosquito control programs. Therefore, it is important to perform research on different aspects to identify the exact toxicological effect and its underlying mechanism of action for the mitigation or alteration of usage of these chemicals. Such studies have the prospect of bringing a permanent chemical modification of this pest-controlling molecule to enhance its activity through considerable reduction in the adverse effects. In this review, we accommodated the molecular action of Chlorpyrifos at synaptic sites and its impacts on anatomy, physiology, cell biology, and development of various organisms, including invertebrates, vertebrates, soil microbes, and plants. Thus, this review aims to highligh potential areas for future research on this significant bioactive compound, paving the way for safer and more effective pest control solutions.

1. Introduction

The extensive usage of pesticides in modern agriculture is a growing source of worry due to environmental contamination and ensuing biodiversity loss. Since 1950, the first line of defense for crop protection has always been pesticides (Jeevanandham, 2020). Despite being used extensively in farming, pesticides leave behind a range of remnants that are harmful to human health when they are consumed in food, water, dairy items, and meat. (R. N. Singh, 2013). Most pesticides now in use are non-selective and have an impact on the biotic component of the ecosystem. The uncontrolled utilization pesticides triggered a variety of issues, including environmental contamination, insect pest resistant, an increase in secondary pests owing to the eradication of its natural enemies, higher manufacturing costs, and risks for both people and animals. (Jeevanandham, 2020). Several of these pesticides inadvertently reach aquatic habitats when they are applied on a wide scale using techniques like crop dusting, orchard and forest spraying, or mosquito control. Regular use of synthetic fertilizers and pesticides also contaminates groundwater, rendering it unsafe for human use. So, before their widespread usage is permitted, understanding of these wide ranging pesticides' impacts on aquatic, terrestrial organisms and their ecosystems is required (Holcombe et al., 1982; Ma et al., 2023).

Chlorpyrifos is a potent organophosphorus pesticide as well as insecticide. Molecular formula is O, O-Diethyl O-3,5,6-trichloro-2-pyridyl phosphorothionate. Alternative names are Lorsban, chlorpyrifos, Dowco 179, ENT2731 (Holcombe et al., 1982). This pesticide is very successful at eradicating a variety of insect vectors that are crucial to human disease, as well as pests that affect stored grains and soil. (Miyazaki & Hodgson, 1972). In underdeveloped nations, organophosphorus (OPs) chemicals continue to be among one of the most popular pesticides since they are widely regarded as the best way to protect crops from insects (Costa, 2018). These substances are exceedingly poisonous, easily entering cells, and significantly altering

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how creatures nervous systems respond (Cao et al., 1999; Saunders et al., 2012). One of the most popular OPs in the world, the active component in the brand-name product Dursban® has numerous applications in pest control for crops in urban and on farms (Lemus & Abdelghani, 2000). This review is aimed at compiling the toxicological aspects of Chlorpyrifos among different organism models.

Chlorpyrifos has a long history of toxicity concerns for the environment. It has severe toxic effects that can cause various diseases including neuro-behavioral effects, developmental effects, and impacts on reproductive health.

2. Mode of Action of Chlorpyrifos

An overabundance of acetylcholine builds up in cholinergic synapses as a result of organophosphates reversible binding to and inactivation of the acetylcholinesterase (AchE) enzyme (Fig.1). This extra acetylcholine overstimulates cholinergic signaling at first, and then paralyzes it. Ach after releasing from the vesicle binds with the Ach Receptor, passes signal then Ach Esterase binds with Ach and stops the signal. Organophosphate inhibit Ach Esterase resulting in continues signal transmission which ultimately can cause to death of the organism.

2.1. The Effect of Chlorpyrifos on Insects

Aquatic insect species are severely affected by chlorpyrifos. The action of this insecticide is influenced by water temperature. At various levels of biological organization, from the organismal up to the ecosystem level, these stressors may promote one another's adverse effects (Galic et al., 2018 ; Holmstrup et al., 2010 and Wang et al., 2019). Thus, it really is essential to incorporate the cumulative influence of such stressors for a more accurate evaluation of ecological risk, particularly in light of projected warming (Noyes & Lema, 2015);Van den Brink et al., 2018).

Chlorpyrifos concentration (0.2 µg L-1) in isopod *Asellus aquaticus* coupled with warmer or variable temperatures, produced harmful effects on development and feed intake rates (Theys et al., 2021) . This results imply that the pesticide became more hazardous in terms of sub-lethal effects under the most difficult temperature conditions (mean of 22 °C with 8 °C Daily Temperature Fluctuation), what is consistent with the idea of CITS-climate-induced toxicant sensitivity. Mean temperatures and DTFs are quite common in the present climate (these are more in cities and lower latitudes), (Verheyen & Stoks, 2019), anticipated to exacerbate even under climate change (Colinet et al., 2015).

The changes in *A. aquaticus'* development and feeding patterns caused by pesticide exposure and global warming may have significant effects on freshwater ecosystems by affecting the mechanism of nutrient recycling (Theys et al., 2021).

Honey bees have been shown to be very toxic to chlorpyrifos (in Dursban 480 EC) when exposed orally, with an LD_{50} of 0.33 g a.i./bee for 48 hours (Giddings et al., 2014). A test involving oral and topical exposure to Dursban (Chlorpyrifos) on *Osmia bicornis* (red mason bee) verified its severe toxicity; it demonstrated extraordinarily high toxicity upon oral exposure to *O. bicornis* over a period of 7 days, with an LC_{50} that was around 250 times lesser than the suggested concentration of field application and after 24 hours, the toxicity went very high (LC_{50} = 0.012 RAC- Recommended field Application Concentration of an active ingredient; 95% CI- Confidence Interval 0.009-0.018), and it continued to rise over the following 96 hours, reaching LC_{50} = 0.004 RAC (95% CI 0.002-0.007) (Mokkapati, Wnęk, et al., 2021). Chlorpyrifos once more demonstrated the maximum toxicity to *O. bicornis* when exposed topically, with an LC_{50} that was around 70% lesser than the concentration advised for field administration of this product and calculated 48-hour LD_{50} for chlorpyrifos was 0.34 g/bee (95% CI 0.28-0.47) (Mokkapati, Wnęk, et al., 2021). The use of *O. bicornis* as a substitute model organism for honey bees in pesticide risk assessment has recently been called into question after Uhl et al., 2019 tested the sensitivities of bees to different commercial pesticide formulations via topical contact exposure and found *O. bicornis* was more resistant than *Apis. Mellifera* (Mokkapati, Bednarska, et al., 2021).

Even at field-realistic concentrations, persistent feeding exposure of *O. bicornis* larvae to Chlorpyrifos significantly reduced the likelihood that the larvae would survive to become adults and also at all investigated doses, the body mass of larvae fed chlorpyrifos-contaminated pollen was decreased and substantially different from control larvae (Bednarska et al., 2022). It has been established that exposure to pesticides can have both lethal and sublethal effects on larval development, including length of time from the second instar to the start of cocoon spinning, body mass, and weight loss during hibernation (Bednarska et al., 2022).

Chlorpyrifos were mild deleterious to the predator green lacewing, *Chrysoperla zastrowi*. It was seen that greater egg mortality was recorded with chlorpyrifos at 2000, 1500, 1000 and 750 ml/ha (65.00%, 62.00%, 61.33% and 56.00% respectively) whereas the low grub mortality was observed in chlorpyrifos 20 EC 750 ml/ha (51.00%) at 48 hours of treatment whereas it was high in chlorpyrifos 20 EC 2000 ml/ha (82.50%) followed by chlorpyrifos 20 EC at 1500 ml/ha (77.50%), chlorpyrifos 20 EC 1000 ml/ha (60.00%) and Dursban 20 EC 1000 ml/ha (62.50%) (Jeevanandham, 2020). The percent mortality after 48 hr due to the concentration of chlorpyrifos 20 EC at 1000, 750, 500 and 250 was 70.00, 66.67, 60.00, 44.00, whereas 63.33 per cent mortality was found with Dursban 20 EC 500 ml/ha (Jeevanandham, 2020).

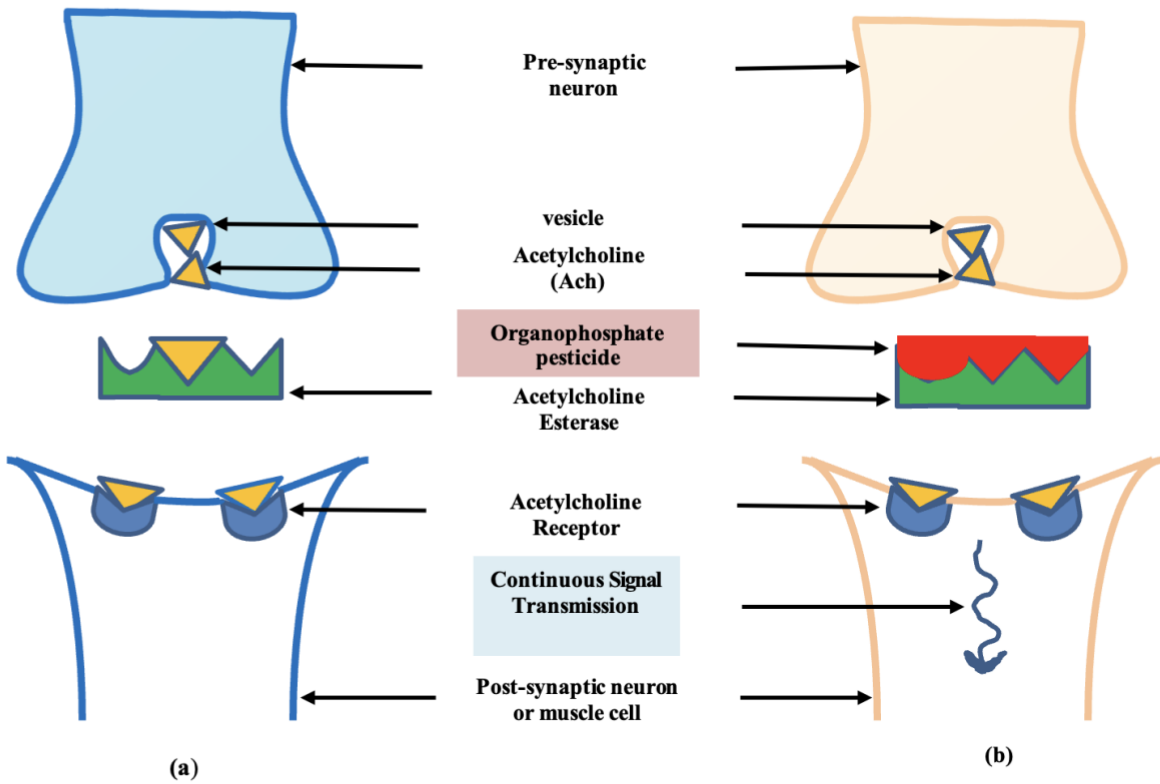

Figure 1. Mode of action of organophosphate in synaptic neuron or muscle cell (a) Ach Esterase stops signal (Ach after releasing from the vesicle binds with the Ach Receptor, passes signal then Ach Esterase binds with Ach and stops the signal) (b) Organophosphate inhibit Ach Esterase (organophosphate inhibits Ach Esterase resulting in continuous signal transmission)

2.2. The effect of chlorpyrifos on fish and amphibian

Fish have proven to be valuable experimental models for analyzing aquatic ecosystems exposed to environmental contamination and related hematological alterations. When determining the structural and functional status of fish exposed to toxins, blood parameters can be employed as good physiological markers of the overall health of the organism. (Seriani et al., 2012).

Chlorpyrifos may have a negative impact on non-target creatures like fish in aquatic ecosystem. For instance, the common carp (*Cyprinus carpio* L) when sub chronically exposed to it for six weeks at a concentration of 36, 53,113 µg /l; leukocytes, erythrocytes, mean corpuscular volume hemoglobin (Hb), packed cell volume (PCV) all decreased in the last two weeks of exposure (Showy & Rabee, 2019). However, there was a considerable rise in the Mean corpuscular hemoglobin and Mean corpuscular hemoglobin concentration (Showy & Rabee, 2019). To show the hazardous nature of insecticide hematological parameters are significant. At 24 hours and thereafter, schooling behavior of fathead minnows *Pimephales promelas* when exposed to 47 µg $litre^{-1}$ chlorpyrifos and above was disrupted; after 48 hours, all concentration exposures appeared to have rigid vertebral columns, and several fish had developed obvious spinal abnormalities at all chlorpyrifos concentrations; the 96-hr LC_{50} s for chlorpyrifos were 8.0 µg $litre^{-1}$ for rainbow trout and 203.0 µg $litre^{-1}$ for fathead minnow (Holcombe et al., 1982).

Adult zebrafish with chronic exposure to CPF have reduced reproductive capacity while long-term exposure to CPF altered the ovary's transcriptome and caused histological damage; it is possible for CPF to be passed straight from exposed adult fish to their unexposed young (Ma et al., 2023).

Tadpoles of the Asian grass frog, *Fejervarya limnocharis* species exhibited LC_{50} of 2.86 g/L for chlorpyrifos and experienced lower survival rates after being exposed at levels greater than 1 g/L; three different morphological changes including an asymmetrical body shape, an aberrant tail shape and the presence of lumps or edema were observed (Ramadani, Marhendra, Wiadnya, et al., 2022).

2.3. The effect of chlorpyrifos on mammals

Spermatogenesis is influenced through oxidative stress, hormonal, or genotoxic pathways (Perry, 2008). The vital semen features of sperm count, motility, and morphology are excellent indicators of male fertility. In mature male Swiss albino mice (*Mus musculus*), the reproductive toxicity of the pesticide chlorpyrifos was investigated of 6 to 8 weeks old, weighing 25 to 28 gm on alternate days for a period of 15 days via intraperitoneal administration of chlorpyrifos (20 mg/kg) (Regeai et al., 2016). Chlorpyrifos was found to show a decrease in all sperm parameter especially sperm motility supporting the hypothesis that exposure to pesticides may be linked to lowered semen quality which leading to infertility (Regeai et al., 2016).

Joshi et al., 2007 examined the toxic effects of the pesticide CPS when orally administered to male rats at doses 7.5, 12.5 and 17.5 mg/kg/day for 30 following days on testicular histology, biochemistry, sperm dynamics, and testosterone levels; their findings showed that chlorpyrifos causes severe testicular damage, lower testosterone levels, and a decrease in sperm count. Similar results were obtained when adult male mice given oral gavage treatments with chlorpyrifos at doses 5, 15, 25 mg/kg/day for 4 weeks; the percentage of morphologically normal spermatozoa decreased in the 15 and 25 mg/kg/day dose groups, but count and sperm motility lowered in all treated groups in comparison to the control (Farag et al., 2010). On the other hand, the testes of chlorpyrifos treated group showed significant harm to the histoarchitecture and organization of seminiferous tubules compared to control group. The histological lesions emphasize the positive correlation between cytogenetic damage and abnormal sperm parameters. The histological lesions of testicular tissue on treated Swiss albino mice include: altered seminiferous tubules, decreased lumen width of seminiferous tubules, decreased spermatozoa numbers in the lumen of the seminiferous tubules, altered and sloughed normal germinal epithelial cells (spermatogonia) lining seminiferous tubules, increased interstitial space and vasodilatation of interstitial blood vessels (Regeai et al., 2016). Within utero exposure throughout gestation, chlorpyrifos cause hyperglycemia, insulin resistance, and dyslipidemia in female nulliparous Wistar rats and their offspring (Ndonwi et al., 2020). The biochemical changes in the offspring last until adulthood, indicating that the chemicals involved either continue to affect the offspring even after exposure ends or modify the epigenome through epigenetic mechanisms (Ndonwi et al., 2020). Therefore, additional study on the epigenome is necessary to learn more about these molecular pathways.

Chlorpyrifos impacted sperm motility and ejaculate volume in Holstein bulls. Up to 6 months after treatment, the pre-freeze discards and the percentage of post-thaw discards were impacted (Everett, 1982). In *Bubalus bubalis* spermatozoa, chlorpyrifos (0.05–0.5 g/ml) alone decreased the sperm straight line velocity, acrosomal integrity, mitochondrial membrane potential, and chromatin decondensation ability (Selvaraju et al., 2014).

Research on lab animals shows that younger animals are more vulnerable than older animals to the acute cholinergic toxicity of chlorpyrifos (Eaton et al., 2008). The early growing neural system may, nevertheless, be comparatively more vulnerable to chlorpyrifos than the more developed neurological system in young kids and infants, according to some in vitro mechanistic evidence; pregnant women should therefore be regarded as a potentially vulnerable demographic (Eaton et al., 2008).

2.4. Impacts of the use of chlorpyrifos on soil microbes

Generally, only relatively a small part of the entire amount of applied pesticides actually reach pests (Pimentel & Burgess, 2012). As a result, pesticides build up in soil and damage soil microorganisms (Wu et al., 2015). This may then have an indirect impact on soil aggregation, soil fertility (Kalia & Gosal, 2011), nutrient cycling, and overall agroecosystem performance (Gattinger et al., 1995 ; Bowles et al., 2014). However, Medo et al., 2015 reported that recommended dose levels of the insecticides cold be considered safe but relatively higher dose can negatively affect the soil microbial community. Chlorpyrifos (100 and 200 mg L^{-1}) was discovered to have a detrimental impact on the production of indole acetic acid and the solubilization of phosphate by bacteria (Akbar & Sultan, 2016).

Groundnut (*Arachis hypogaea* L.) fields when treated with chlorpyrifos showed a relatively brief inhibitory impact on the total bacterial population, although they recovered after seed treatment and soil treatment within 60 and 45 days, respectively; the fungus population rapidly increased (Pandey & Singh, 2004). The results obtained from the study of Supreeth et al., 2016 highlighted that the chlorpyrifos support the *Actinomycete* growth in the soil, but inhibited other microorganisms. *Streptomyces* sp. HP-11 used in this study showed metabolic pathway for detoxification of chlorpyrifos recommending the strain HP-11 would be a promising candidate for the bioremediation of chlorpyrifos contaminated soil and water (Supreeth et al., 2016). By analyzing BIOLOG plates using Average Well-Colour Development (AWCD) values Medo et al., 2015 found increased metabolic potential of microorganisms in the soil treated with high dose of Chlorpyrifos.

From another study found that, on the first day after treatment (DAT), chlorpyrifos dramatically decreased the amounts of bacteria, fungus, and actinomycetes by 44.1%, 61.1%, and 72.8% as compared to those in the controls (Chu et al., 2008). The 53-day chlorpyrifos half-life measured by Karpun et al., 2021 was significantly longer than those found by Chu et al., 2008 and Pandey & Singh, 2004. Environmental variables, like varying soil pH, temperature, moisture, organic carbon content, and pesticide formulation, can be blamed for the significant variance in chlorpyrifos half-life (Singh & Walker, 2006). This difference emphasizes the value of conducting assessments locally rather than projecting the findings of studies conducted in different soils and/or under various situations. It was found that a long half-life of chlorpyrifos associated with a long-term suppression of soil microbial respiration, which was reversed after 180 days of chlorpyrifos application (Karpun et al., 2021). Pandey & Singh, 2004 discovered suppressive effect on the bacterial population, which had fully recovered within 105th day following the application of chlorpyrifos. These research discrepancies can be related to the various environmental factors. Environmental considerations might be a major deciding factor when choosing a pesticide.

From the studies it could be concluded that the application of Chlorpyrifos affects microbial community in the soil and impaired the biodiversity of soil microbes by eliminating or making some microbes predominant. The low biodegradation of chlorpyrifos and the fact that it was harmful to the bacterial community in the soil both contribute to its persistence in the soil. Chlorpyrifos at the dose of 70% ($C_{70}E$) showed a remarkable loss of earthworm weight from day 21 (De Bernardi et al., 2022). For bioremediation of the Chlorpyrifos some predominant community *Achromobacter xylosoxidans, Ochrobactrm* sp. strains (Akbar & Sultan, 2016), *Streptomyces sp.* (Supreeth et al., 2016) HP-11 exhibited the ability to degrade chlorpyrifos in the soil.

2.5. Impacts of the use of chlorpyrifos on plants

The carrot, or *Daucus carota* L., is a significant and healthy winter root vegetable that contains a variety of necessary β-carotene, which in people is converted into vitamin A when bile salts are present in the intestines. Ascorbic acid, minerals, dietary fiber, antioxidants, a little amount of protein, and its nutritional value per 100 g (3.5 oz) are also abundant in this food (Haque et al., 1970). Since carrots are more likely to be infested by pests than other crops, for the higher productivity pesticides are regularly sprayed on carrot fields to increase output. However, indiscriminate pesticide use on vegetables is regarded to represent a significant risk to humans and also lowers carrot yield and mineral content (Reddy et al., 1997). An accumulation of heavy metal and metalloid residues in many agricultural soils as a result of the indiscriminate use of inorganic and organic pesticides has significantly decreased agricultural production.

According to (Naushad Alam, 2016) carrot absorption in the case of Chlorpyrifos treatment ranged from 0.205 - 2.580 ($\mu g\ l^{-1}$) at rates of 0.50 l ha^{-1}, 1.00 ha^{-1}, 2.00 l ha^{-1}, and 4.00 l ha^{-1} during the growth stages; as opposed to this, the residual levels of Chlorpyrifos at various doses ranged from 0.443 - 0.329 ($\mu g\ l^{-1}$). A study was conducted to find and quantify the amount of chlorpyrifos in some selected vegetables sprayed with the recommended dose of chlorpyrifos (3 ml/L of water); found that the amount of chlorpyrifos residue in cauliflower was higher than the Maximum Residue Limit set by European Union (EU-MRLs) up to 9 days after spray (DAS) (0.012 mg/kg), however, the amounts found in brinjal and tomato were higher than EU-MRLs up to 7 DAS (0.029 mg/kg and 0.017 mg/kg residue, respectively) (Ahmed et al., 2021).

Table 1. Effects of Chlorpyrifos on different organisms

Test organism	Species	Effects	References
Isopod	*Asellus aquaticus*	harmful effects on development and feed intake rates	Theys et al., 2021
Red mason bee	*Osmia bicornis*	demonstrated extraordinarily high toxicity upon oral and topical exposure; lethal and sublethal effects on larval development	Mokkapati, Wnęk, et al., 2021; Bednarska et al., 2022
Green lacewing	*Chrysoperla zastrowi*	higher egg mortality	Jeevanandham, 2020
Common carp	*Cyprinus carpio*	RBC, WBC, Hb, MCV PCV decreased, MCH, MCHC	Showy & Rabee, 2019
Fathead minnows	*Pimephales promelas*	spinal deformities	Holcombe et al., 1982
Asian grass frog	*Fejervarya limnocharis*	lower survival rates; morphological changes- asymmetrical body shape, an aberrant tail shape, and the presence of lumps or edema	Ramadani, Marhendra, & Kurniawan, 2022
Earthworm		Weight loss	De Bernardi et al., 2022
Swiss albino mice	*Mus musculus*	Decrease in sperm motility, marked damage in histoarchitecture and organization of seminiferous tubules	Regeai et al., 2016
Male rat		lower testosterone levels and a decrease in sperm count; decrease in sperm motility and count	Joshi et al., 2007 ; Farag et al., 2010
Wistar rats	*Rattus norvegicus*	cause hyperglycemia, insulin resistance, and dyslipidemia in adult female and their offspring	Ndonwi et al., 2020
Holstein bull		impacted sperm motility and ejaculate volume	Everett, 1982
Water buffalo	*Bubalus bubalis*	decreased the sperm straight line velocity, acrosomal integrity, mitochondrial membrane potential, and chromatin decondensation ability	Selvaraju et al., 2014
Soil microbial population	bacteria, fungus, and actinomycetes	decreased the amounts of bacteria, fungus, and actinomycetes by 44.1%, 61.1%, and 72.8%, respectively	Chu et al., 2008
Carrot	*Daucus carota*		Naushad Alam, 2016

In another study, where the samples were collected from different commercial markets of Dhaka it found that; 44% of cauliflower samples had chlorpyrifos residue, while 91% of samples had levels over the maximum residue level (MRL); in 68% of the cabbage samples, the residue was detected, and in 53% of the samples, it exceeded MRL; in the case of eggplant, the residue was detected in 80% of the samples, with 65% of the samples exceeding the MRL (Momtaz, 2024).

3. Cyto and Genotoxicological Properties of Chlorpyrifos

Erythrocytes from *Labeo rohita* treated to sublethal levels of chlorpyrifos for 96 hour displayed significantly higher micronucleus ($P<0.01$) than the control and the hematological criterion such as total red blood cells count, hemoglobin, packed cell volume reduced whereas total white blood cells count rised (Ismail et al., 2018). Another study on erythrocytes of the common carp has showed that although low levels of CPF exposure did not result in raised amounts of micronuclei, they did result in a significantly higher frequency of overall nuclear abnormalities (NAs) which included nicked, nodded and octad shaped nuclei, buds of nucleus, bridges of nucleus and two nucleated cells while abundance of polychromatic red blood cells decreased; DNA damage were identified by single cell gel electrophoresis. (Mitkovska & Chassovnikarova, 2020). Researcher have exposed sublethal and nonlethal concentrations of CPF in freshwater fish *Channa punctatus* (Bloch) for 96 h and found micronucleus induction in the blood was sky-high on 14th day at 203.0 μg/l; the high-rise DNA damage (by comet assay) was found on 5th day, thenceforward a gradational non-linear reduction in the lymphocytes and cells of gill (Ali et al., 2009).

Erythrocytes of Brazilian neotropical tadpoles (*Odontophrynus carvalhoi*) was exposed for 24h, 48h and 96h to various nominal concentrations of this insecticide (10, 100, 200 and 400 μg L^{-1}) showed excessive frequencies of genetic changes (micronuclei) when contrasted with the negative control but the differences in the frequencies of the micronuclei found after 48 hrs of subjection were statistically significant ($p<0.05$) (Silva et al., 2020). CPF when taken orally (1.5 or 3.0 mg/kg) cause significant cholinesterase inhibition but without overt harm can negatively impact the expression levels of crutial genes - nerve growth factor (NGF), reelin, mRNA for the muscarinic acetylcholine receptor (mAChR) M1 subtype, oligodendrocytes (myelin-associated glycoprotein, MAG), significantly decreased whereas, astrocytes (glial fibrillary acidic protein, GFAP) significantly increased which are taking part in the rat's development of the brain throughout the initial postpartum period (Betancourt et al., 2006). Previously it has been exhibited that CPF can make difference in protein levels of NGF (Betancourt & Carr, 2004).

Chlorpyrifos can alter the antioxidant status. Buffalo calves (*Bubalus bubalis*) was sub-chronically exposed to it at a dose rate of 0.05 mg/kg/day for 20 following weeks notably escalated the enzymic activity of glutathione peroxidase (54.8%), glutathione reductase (79.4%), glutathione-S-transferase (34.2%), glucose-6-phosphate dehydrogenase (33.2%), superoxide dismutase (19.3%) and catalase (63.8%) (Kaur & Sandhu, 2008). Using human leukocytes in vitro with a concentration of 35 μg mL^{-1}, a study by Serpa et al. (2019) has shown that CPF has a genotoxic effect by altering the following things: micronuclei, numerical chromosomal abnormalities and apoptotic cells.

4. Conclusion

The data of this study shows that Chlorpyrifos has a wide-ranging and disastrous impact and it has been investigated for their harmful effects on practically all phyla's members. The primary physiological systems of the body, such as the neurological, circulatory, and reproductive systems, exhibit a variety of its effect on different organisms in different ways for example, in the case of insects, it kills; in humans, it affects reproductive health; in fish, it affects blood parameters.. Thus, the frequent release of such poisons into the environment poses a serious risk to both human and animal health, it is necessary to closely monitor and regulate the use of this pesticide. As a result, it is advised to limit the use of chlorpyrifos and concentrate on secure alternatives (such biological management) to address the issues with toxicity and pollution.

List of Abbreviations

CPF-Chlorpyrifos, OPs–organophosphorus, DTF-Daily Temperature Fluctuation, CITS-climate-induced toxicant sensitivity, LC_{50}- Lethal Concentration, LD_{50}-Lethal Dose, RAC- Recommended field Application Concentration, CI-Confidence Interval, EC-Effective Concentration, Hb - hemoglobin, PCV -packed cell volume, DAT -day after treatment, AWCD-Average Well-Colour Development, NA-nuclear abnormalities, NGF-nerve growth factor, mAChR-mRNA for the muscarinic acetylcholine receptor, MAG -myelin-associated glycoprotein, GFAP- glial fibrillary acidic protein.

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Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

References

Ahmed, M. S., Hossain Prodhan, M. D., Begum, A., Afroze, M., & Sarker, D. (2021). 335 Estimation of residue degradation of cypermethrin and chlorpyrifos in brinjal, 336 tomato and cauliflower under supervised field trial. Asian-Australasian Journal of 337 Bioscience and Biotechnology, 6(2), 60–67. 338 https://doi.org/10.3329/aajbb.v6i2.56141

Akbar, S., & Sultan, S. (2016). Soil bacteria showing a potential of chlorpyrifos degradation and plant growth enhancement. *Brazilian Journal of Microbiology*, *47*(3), 563–570. https://doi.org/10.1016/j.bjm.2016.04.009

Ali, D., Nagpure, N. S., Kumar, S., Kumar, R., Kushwaha, B., & Lakra, W. S. (2009). Assessment of genotoxic and mutagenic effects of chlorpyrifos in freshwater fish Channa punctatus (Bloch) using micronucleus assay and alkaline single-cell gel electrophoresis. *Food and Chemical Toxicology*, *47*(3), 650–656. https://doi.org/10.1016/j.fct.2008.12.021

Bednarska, A. J., Mikołajczyk, Ł., Ziółkowska, E., Kocjan, K., Wnęk, A., Mokkapati, J. S., Teper, D., Kaczyński, P., Łozowicka, B., Śliwińska, R., & Laskowski, R. (2022). Effects of agricultural landscape structure, insecticide residues, and pollen diversity on the life-history traits of the red mason bee Osmia bicornis. *Science of the Total Environment*, *809*.

https://doi.org/10.1016/j.scitotenv.2021.151142

Betancourt, A. M., Burgess, S. C., & Carr, R. L. (2006). Effect of developmental exposure to chlorpyrifos on the expression of neurotrophin growth factors and cell-specific markers in neonatal rat brain. *Toxicological Sciences*, *92*(2), 500–506. https://doi.org/10.1093/toxsci/kfl004

Betancourt, A. M., & Carr, R. L. (2004). The effect of chlorpyrifos and chlorpyrifos-oxon on brain, cholinesterase, muscarinic receptor binding, and neurotrophin levels in rats following early postnatal exposure. *Toxicological Sciences*, *77*(1), 63–71. https://doi.org/10.1093/toxsci/kfh003

Bowles, T. M., Acosta-Martínez, V., Calderón, F., & Jackson, L. E. (2014). Soil enzyme activities, microbial communities, and carbon and nitrogen availability in organic agroecosystems across an intensively-managed agricultural landscape. *Soil Biology and Biochemistry*, *68*, 252–262.

https://doi.org/10.1016/j.soilbio.2013.10.004

Chu, X., Fang, H., Pan, X., Wang, X., Shan, M., Feng, B., & Yu, Y. (2008). Degradation of chlorpyrifos alone and in combination with chlorothalonil and their effects on soil microbial populations. *Journal of Environmental Sciences*, *20*(4), 464–469.

https://doi.org/10.1016/S1001-0742(08)62080-X

Colinet, H., Sinclair, B. J., Vernon, P., & Renault, D. (2015). Insects in fluctuating thermal environments. *Annual Review of Entomology*, *60*, 123–140. https://doi.org/10.1146/annurev-ento-010814-021017

Costa, L. G. (2018). Organophosphorus compounds at 80: Some old and new issues. *Toxicological Sciences*, *162*(1), 24–35. https://doi.org/10.1093/toxsci/kfx266

De Bernardi, A., Marini, E., Casucci, C., Tiano, L., Marcheggiani, F., Ciani, M., Comitini, F., Taskin, E., Puglisi, E., & Vischetti, C. (2022). Ecotoxicological effects of a synthetic and a natural insecticide on earthworms and soil bacterial community. *Environmental Advances*, *8*(April), 100225.

https://doi.org/10.1016/j.envadv.2022.100225

Eaton, D. L., Daroff, R. B., Autrup, H., Bridges, J., Buffler, P., Costa, L. G., Coyle, J., McKhann, G., Mobley, W. C., Nadel, L., Neubert, D., Schulte-Hermann, R., & Spencer, P. S. (2008). Review of the toxicology of chlorpyrifos with an emphasis on human exposure and neurodevelopment. *Critical Reviews in Toxicology*, *38*(SUPPL.2), 1–125.

https://doi.org/10.1080/10408440802272158

Everett, R. W. (1982). Effect of Dursban 44 on Semen Output of Holstein Bulls. *Journal of Dairy Science*, *65*(9), 1781–1794. https://doi.org/10.3168/jds.S0022-0302(82)82417-X

Farag, A. T., Radwan, A. H., Sorour, F., El Okazy, A., El-Agamy, E. S., & El-Sebae, A. E. K. (2010). Chlorpyrifos induced reproductive toxicity in male mice. *Reproductive Toxicology*, *29*(1), 80–85. https://doi.org/10.1016/j.reprotox.2009.10.003

Galic, N., Sullivan, L. L., Grimm, V., & Forbes, V. E. (2018). When things don't add up: quantifying impacts of multiple stressors from individual metabolism to ecosystem processing. *Ecology Letters*, *21*(4), 568–577. https://doi.org/10.1111/ele.12923

Gattinger, A., Palojärvi, A., & Schloter, M. (1995). *Chapter 3.4*.

Giddings, J. M., Williams, M. W., Solomon, K. R., & Giesy, J. P. (2014). Reviews of environmental contamination and toxicology: risks to aquatic organisms from use of chlorpyrifos in the United States. In *Reviews of Environmental Contamination and Toxicology* (Vol. 231).

Haque, K., Jahangir, A., Haque, M., Mondal, R., Jahan, M., & Sarker, M. (1970). Yield and Nutritional Quality of Cabbage as Affected by Nitrogen and Phosphorus Fertilization. *Bangladesh Journal of Scientific and Industrial Research*, *41*(1), 41–46. https://doi.org/10.3329/bjsir.v41i1.267

Holcombe, G. W., Phipps, G. L., & Tanner, D. K. (1982). The Acute Toxicity of Kelthane, Dursban, Disulfoton, Pydrin, and Permethrin to Fathead Minnows Pimephales Promelas and Rainbow Trout Salmo Gairdneri. In *Environmental Pollution (Series A)* (Vol. 29).

Holmstrup, M., Bindesbøl, A. M., Oostingh, G. J., Duschl, A., Scheil, V., Köhler, H. R., Loureiro, S., Soares, A. M. V. M., Ferreira, A. L. G., Kienle, C., Gerhardt, A., Laskowski, R., Kramarz, P. E., Bayley, M., Svendsen, C., & Spurgeon, D. J. (2010). Interactions between effects of environmental chemicals and natural stressors: A review. *Science of the Total Environment*, *408*(18), 3746–3762.

https://doi.org/10.1016/j.scitotenv.2009.10.067

Ismail, M., Ali, R., Shahid, M., Khan, M. A., Zubair, M., Ali, T., & Mahmood Khan, Q. (2018). Genotoxic and hematological effects of chlorpyrifos exposure on freshwater fish Labeo rohita. *Drug and Chemical Toxicology*, *41*(1), 22–26. https://doi.org/10.1080/01480545.2017.1280047

Jeevanandham, N. (2020). Effect of chlorpyriphos 20ec on predatory green lacewing Chrysoperla zastrowi Sillemi under laboratory condition. *~ 1162 ~ Journal of Pharmacognosy and Phytochemistry*, *9*(3). www.phytojournal.com

Joshi, S. C., Mathur, R., & Gulati, N. (2007). Testicular toxicity of chlorpyrifos (an organophosphate pesticide) in albino rat. *Toxicology and Industrial Health*, *23*(7), 439–444. https://doi.org/10.1177/0748233707080908

Kalia, A., & Gosal, S. K. (2011). Effect of pesticide application on soil microorganisms. *Archives of Agronomy and Soil Science*, *57*(6), 569–596.

https://doi.org/10.1080/03650341003787582

Karpun, N. N., Yanushevskaya, E. B., Mikhailova, Y. V., Díaz-Torrijo, J., Krutyakov, Y. A., Gusev, A. A., & Neaman, A. (2021). Side effects of traditional pesticides on soil microbial respiration in orchards on the Russian Black Sea coast. *Chemosphere*, *275*, 130040.

https://doi.org/10.1016/j.chemosphere.2021.130040

Kaur, R., & Sandhu, H. S. (2008). In vivo changes in antioxidant system and protective role of selenium in chlorpyrifos-induced subchronic toxicity in bubalus bubalis. *Environmental Toxicology and Pharmacology*, *26*(1), 45–48. https://doi.org/10.1016/j.etap.2008.01.004

Lemus, R., & Abdelghani, A. (2000). Chlorpyrifos: An unwelcome pesticide in our homes. *Reviews on Environmental Health*, *15*(4), 421–433.

https://doi.org/10.1515/REVEH.2000.15.4.421

Ma, J., Zhu, P., Wang, W., Zhang, X., Wang, P., Sultan, Y., Li, Y., Ding, W., & Li, X. (2023). Environmental impacts of chlorpyrifos: Transgenerational toxic effects on aquatic organisms cannot be ignored. *Science of The Total Environment*, *905*, 167311.

https://doi.org/10.1016/j.scitotenv.2023.167311

Medo, J., Maková, J., Kovácsová, S., Majerčíková, K., & Javoreková, S. (2015). Effect of Dursban 480 EC (chlorpyrifos) and Talstar 10 EC (bifenthrin) on the physiological and genetic diversity of microorganisms in soil. *Journal of Environmental Science and Health - Part B Pesticides, Food Contaminants, and Agricultural Wastes*, *50*(12), 871–883.

https://doi.org/10.1080/03601234.2015.1062659

Mitkovska, V., & Chassovnikarova, T. (2020). Chlorpyrifos levels within permitted limits induce nuclear abnormalities and DNA damage in the erythrocytes of the common carp. *Environmental Science and Pollution Research*, *27*(7), 7166–7176. https://doi.org/10.1007/s11356-019-07408-9

Miyazaki, S., & Hodgson~, G. C. (1972). Chronic Toxicity of Dursban and Its Metabolite, 3,5,6-Trichloro-2-pyridinol in Chickens1. In *Toxicology and Applied Pharmacology* (Vol. 23).

Mokkapati, J. S., Bednarska, A. J., & Laskowski, R. (2021). The development of the solitary bee Osmia bicornis is affected by some insecticide agrochemicals at environmentally relevant concentrations. *Science of the Total Environment*, *775*, 145588. https://doi.org/10.1016/j.scitotenv.2021.145588

Mokkapati, J. S., Wnęk, A., Laskowski, R., & Bednarska, A. J. (2021). Acute oral and contact toxicity of three plant protection products to adult solitary bees osmia bicornis. *Polish Journal of Environmental Studies*, *30*(5), 4105–4113.

https://doi.org/10.15244/pjoes/130516

Momtaz, M., & Khan, M. S. (2024). Analysis of Chlorpyrifos pesticide residue in locally grown cauliflower, cabbage, and eggplant using gas Chromatography–Mass spectrometry (GC-MS) technique: A Bangladesh perspective. *Foods*, *13*(11), 1780.

Naushad Alam, M. (2016). Residual Concentrations of Diazinon and Dursban and its Impacts on Soil and Carrot. *Journal of Bioremediation & Biodegradation*, *07*(03).

https://doi.org/10.4172/2155-6199.1000351

Ndonwi, E. N., Atogho-Tiedeu, B., Lontchi-Yimagou, E., Shinkafi, T. S., Nanfa, D., Balti, E. V., Katte, J. C., Mbanya, A., Matsha, T., Mbanya, J. C., Shakir, A., & Sobngwi, E. (2020). Metabolic effects of exposure to pesticides during gestation in female Wistar rats and their offspring: a risk factor for diabetes? *Toxicological Research*, *36*(3), 249–256.

https://doi.org/10.1007/s43188-019-00028-y

Noyes, P. D., & Lema, S. C. (2015). Forecasting the impacts of chemical pollution and climate change interactions on the health of wildlife. *Current Zoology*, *61*(4), 669–689.

https://doi.org/10.1093/czoolo/61.4.669

Pandey, S., & Singh, D. K. (2004). Total bacterial and fungal population after chlorpyrifos and quinalphos treatments in groundnut (Arachis hypogaea L.) soil. *Chemosphere*, *55*(2), 197–205. https://doi.org/10.1016/j.chemosphere.2003.10.014

Perry, M. J. (2008). Effects of environmental and occupational pesticide exposure on human sperm: A systematic review. *Human Reproduction Update*, *14*(3), 233–242. https://doi.org/10.1093/humupd/dmm039

Pimentel, D., & Burgess, M. (2012). Small amounts of pesticides reaching target insects. *Environment, Development and Sustainability*, *14*(1), 1–2. https://doi.org/10.1007/s10668-011-9325-5

Ramadani, S., Marhendra, A. P. W., & Kurniawan, N. (2022). Assessing the Genotoxicity Effect of a Commercial Chlorpyrifos Formulation in Fejervarya limnocharis Tadpoles (Anura: Dicroglossidae) Under Acute and Chronic Exposure. *The Journal of Experimental Life Sciences*, *12*(1), 9–16. https://doi.org/10.21776/ub.jels.2022.012.01.02

Ramadani, S., Marhendra, A. P. W., Wiadnya, D. G. R., & Kurniawan, N. (2022). Effect of Acute Toxicity of Commercial Organophosphate Insecticide Based on Chlorpyrifos on Fejervarya limnocharis Tadpoles (Anura: Dicroglossidae). *Journal of Tropical Life Science*, *12*(2), 231–240. https://doi.org/10.11594/jtls.12.02.09

Reddy, N. S., Dash, S., & Sontakke. (1997). Effect of spraying selected pesticides on the contents of specified minerals in cabbage. *Plant Foods for Human Nutrition*, *51*(4), 357–363. https://doi.org/10.1023/A:1007984813516

Regeai, S., Abusrer, S., Shibani, N., & Jaafari, H. El. (2016). The Effect of Pesticide Chlorpyrifos (Dursban R) on Sperm Parameters and Testicular Tissue in Mice. In *Libyan J. Vet. Med. Sci* (Vol. 2, Issue 2).

Saunders, M., Magnanti, B. L., Correia Carreira, S., Yang, A., Alamo-Hernández, U., Riojas-Rodriguez, H., Calamandrei, G., Koppe, J. G., Krayer Von Krauss, M., Keune, H., & Bartonova, A. (2012). Chlorpyrifos and neurodevelopmental effects: A literature review and expert elicitation on research and policy. *Environmental Health: A Global Access Science Source*, *11*(SUPPL.1), 1–11. https://doi.org/10.1186/1476-069X-11-S1-S5

Selvaraju, S., Jodar, M., & Krawetz, S. A. (2014). *The Influence of Environmental Contaminants and Lifestyle on Testicular Damage and Male Fertility*. 185–203.

https://doi.org/10.1007/7653_2014_13

Seriani, R., Abessa, D. M. de S., Kirschbaum, A. A., Pereira, C. D. S., Romano, P., & Ranzani-Paiva, M. J. T. (2012). Relationship between water toxicity and hematological changes in Oreochromis niloticus. *Brazilian Journal of Aquatic Science and Technology*, *15*(2), 47–53.

Serpa, E. A., Schmitt, E. G., Zuravski, L., Machado, M. M., & de Oliveira, L. F. S. (2019). Chlorpyrifos induces genotoxic effects in human leukocytes in vitro at low concentrations. *Acta Scientiarum - Health Sciences*, *41*(1), 1–6.

https://doi.org/10.4025/actascihealthsci.v41i1.44291

Showy, B. G., & Rabee, A. M. (2019). Hematological alternation in common carp fish (Cyprinus Carpio L. 1758) after exposing to dursban. *Iraqi Journal of Science*, *60*(3), 448–452. https://doi.org/10.24996/ijs.2019.60.3.4

Silva, M. B., Fraga, R. E., Nishiyama, P. B., Costa, N. L. B., Silva, I. S. S., Queiroz, T. S., Rocha, M. A., & Juncá, F. A. (2020). Genotoxic effect of the insecticide Chlorpyrifos on the erythrocytes of Odontophrynus carvalhoi tadpoles (Amphibia: Odontophrynidae). *Ecotoxicology and Environmental Contamination*, *15*, 9–13.

https://doi.org/10.5132/eec.2020.01.02

Singh, B. K., & Walker, A. (2006). Microbial degradation of organophosphorus compounds. *FEMS Microbiology Reviews*, *30*(3), 428–471. https://doi.org/10.1111/j.1574-6976.2006.00018.x

Singh, R. N. (2013). Acute toxicity of an organophosphate, dimethoate to an air breathing fish, Colisa fasciatus (Bl . & Schn .). *Indian J. Sel. Res.*, *4*(1), 97–100.

Supreeth, M., Chandrashekar, M. A., Sachin, N., & Raju, N. S. (2016). Effect of chlorpyrifos on soil microbial diversity and its biotransformation by Streptomyces sp. HP-11. *3 Biotech*, *6*(2), 1–6. https://doi.org/10.1007/s13205-016-0462-2

Theys, C., Verheyen, J., Tüzün, N., & Stoks, R. (2021). Higher mean and fluctuating temperatures jointly determine the impact of the pesticide chlorpyrifos on the growth rate and leaf consumption of a freshwater isopod. *Chemosphere*, *273*, 128528. https://doi.org/10.1016/j.chemosphere.2020.128528

Uhl, P., Awanbor, O., Schulz, R. S., & Brühl, C. A. (2019). Is Osmia bicornis an adequate regulatory surrogate? Comparing its acute contact sensitivity to Apis mellifera. *PLoS ONE*, *14*(8), 1–12. https://doi.org/10.1371/journal.pone.0201081

Van den Brink, P. J., Boxall, A. B. A., Maltby, L., Brooks, B. W., Rudd, M. A., Backhaus, T., Spurgeon, D., Verougstraete, V., Ajao, C., Ankley, G. T., Apitz, S. E., Arnold, K., Brodin, T., Cañedo-Argüelles, M., Chapman, J., Corrales, J., Coutellec, M. A., Fernandes, T. F., Fick, J., … van Wensem, J. (2018). Toward sustainable environmental quality: Priority research questions for Europe. *Environmental Toxicology and Chemistry*, *37*(9), 2281–2295.

https://doi.org/10.1002/etc.4205

Verheyen, J., & Stoks, R. (2019). Temperature variation makes an ectotherm more sensitive to global warming unless thermal evolution occurs. *Journal of Animal Ecology*, *88*(4), 624–636. https://doi.org/10.1111/1365-2656.12946

Wang, Z., Lui, G. C. S., Burton, G. A., & Leung, K. M. Y. (2019). Thermal extremes can intensify chemical toxicity to freshwater organisms and hence exacerbate their impact to the biological community. *Chemosphere*, *224*, 256–264. https://doi.org/10.1016/j.chemosphere.2019.02.103

Wu, X. hu, Xu, J., Liu, Y. zhuo, Dong, F. shou, Liu, X. gang, Zhang, W. wen, & Zheng, Y. quan. (2015). Impact of fluxapyroxad on the microbial community structure and functional diversity in the silty-loam soil. *Journal of Integrative Agriculture*, *14*(1), 114–124. https://doi.org/10.1016/S2095-3119(14)60746-2