



Toxicological Perspectives of Pesticide Use in Mulberry Cultivation: Challenges and Pathways to Sustainable Sericulture

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Authors' contributions

This work was carried out in collaboration among all authors. Author PLD conceptualized the review, conducted the primary literature searches, and wrote the initial draft of the manuscript. Author IN provided expertise on specific topics covered in the review, reviewed and edited the manuscript for important intellectual content, and approved the final version. Authors JS and SS assisted in literature searches and data analysis, contributed to the writing and revision of the manuscript, and provided critical feedback on the draft. All authors read and approved the final manuscript.

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Review Article

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ABSTRACT

Pest management in mulberry cultivation is a critical component of sustainable sericulture, as pest infestations directly influence leaf quality, silkworm health, and cocoon productivity. Conventional insecticides have been widely employed to manage pest outbreaks; however, their indiscriminate application leads to multiple challenges, including pesticide residues in foliage, silkworm toxicity, environmental contamination, and resistance development in pest populations. These issues collectively threaten the economic viability and ecological sustainability of sericulture. This review consolidates current insights on the toxicological impact of insecticides within mulberry–silkworm ecosystems, emphasizing their residual effects on silkworm physiology, cocoon characteristics, and silk quality. It further explores integrated strategies for minimizing pesticide hazards through eco-friendly approaches such as biological control agents, botanical formulations, cultural practices, and selective chemical interventions. The discussion highlights the necessity for region-specific integrated pest management (IPM) modules that align with ecological principles while ensuring effective pest suppression. Additionally, the review underscores the importance of monitoring pesticide residues, adopting safe waiting periods, and implementing farmer-oriented training programs to promote judicious pesticide use. Collectively, these interventions aim to balance pest control efficiency with environmental stewardship, thereby sustaining sericulture under dynamic pest and climate conditions.

Keywords: *Mulberry cultivation; sericulture; pest management; insecticide residues; silkworm toxicity; Integrated Pest Management (IPM); eco-friendly approaches; sustainable sericulture.*

1. INTRODUCTION

Sericulture, the art and science of silk production, plays a vital role in the rural economy and textile industry of many countries, particularly in India, China, and several Southeast Asian nations. India ranks as the second-largest silk producer globally after China, with sericulture serving as an important livelihood source for millions of rural households. Among the critical determinants of successful silk production, mulberry foliage holds paramount importance as it serves as the exclusive food for silkworms, contributing nearly 38.2% to overall productivity and efficiency in sericulture (Vasanth *et al.*, 2025).

Mulberry (*Morus* spp.), being the sole food plant for the silkworm (*Bombyx mori* L.), is highly vulnerable to various biotic stresses, including pests and diseases. The production of superior-quality mulberry leaves is limited by several factors, such as diseases (24%), insect pests (8%), weed infestations (7%), and other constraints accounting for about 51% of total yield loss (Vasanth *et al.*, 2025). Mulberry plants are known to harbor nearly 300 pest species, encompassing both insect and non-insect groups, with over 100 species reported in India (Bandyopadhyay *et al.*, 2005). Among these, sap-sucking insects and defoliators are the most destructive, causing yield losses ranging from 12% to 25%. These losses are largely attributed to reduced leaf nutritional quality and extensive

leaf drop caused by pest attacks (Kumari, 2014). Such infestations impair plant growth, resulting in severe damage and substantial declines in leaf yield, which in turn negatively impacts silkworm rearing efficiency (Yeshika *et al.*, 2019).

In recent years, abrupt climatic fluctuations coupled with continuous harvesting have further exacerbated the susceptibility of mulberry to pests and diseases, leading to marked reductions in both leaf yield and quality. To mitigate these issues, chemical control remains a commonly adopted practice; however, the indiscriminate use of pesticides in mulberry cultivation and pest management deteriorates leaf quality. When such pesticide-contaminated leaves are consumed by silkworms, it adversely affects larval health, rearing performance, cocoon characteristics, and ultimately, silk productivity (Jekinakatti *et al.*, 2024). This situation underscores the growing concern regarding pesticide contamination in the sericulture ecosystem.

Evaluating the effects of insecticide application and its residual toxicity on silkworms has therefore emerged as a critical area of sericulture research. A holistic understanding of these impacts requires consideration of several interconnected factors. The type of pesticide used and its mode of action play a major role in determining its toxic effects (Stuligross & Williams, 2021). Different classes of insecticides,

such as organophosphates, carbamates, pyrethroids, neonicotinoids etc. exhibit variations in toxicity and environmental persistence. Each category operates through distinct mechanisms that can interfere with silkworm physiology and developmental processes (Fajfer & Łochyńska, 2022). Furthermore, the timing, frequency, and method of pesticide application significantly influence the extent of residue deposition on mulberry leaves. Application techniques, including foliar sprays and systemic treatments, alter the pattern and magnitude of contamination in the sericulture ecosystem (Ashoka et al., 2013).

Silkworm larvae (*Bombyx mori*) exhibit extreme sensitivity to pesticides, including insecticides (Lu et al., 2020), fungicides (Li et al., 2019), and insect growth regulators (IGRs) (Santorium et al., 2020). Even at very low concentrations, these chemicals can disrupt the normal development of fifth-instar larvae, causing non-spinning syndrome (NSS) and inducing dauer larval formation. Affected larvae fail to spin cocoons or pupate within the normal period; instead, they remain in an extended larval phase, continue feeding abnormally, and eventually die within one to two weeks (Gürel, 2025). In addition, toxic effects that occur during early instars may manifest later in development, aggravating the risks associated with pesticide exposure. Consequently, pesticide-induced mortality in silkworms poses a significant threat to the sustainability of the silk industry (Nicodemo et al., 2018). Among the different exposure pathways, ingestion of pesticide-contaminated mulberry leaves remains the most common and critical route, as these leaves constitute the sole food source for silkworms (Kalita et al., 2016).

Recent studies have revealed frequent detection of pesticide residues in mulberry leaves, primarily due to drift from neighboring agricultural fields, residual deposits on spray equipment, and direct application of chemicals in mulberry gardens (Sun et al., 2012; Zhang et al., 2008). This multiple-route contamination substantially increases the likelihood of synergistic or additive interactions, which can intensify toxic effects on silkworms (Arakawa et al., 2011; Wang et al., 1999; Yu et al., 2018; Zhang et al., 2008). Despite these concerns, very few studies have examined the combined acute toxicity of pesticide mixtures on silkworms (Yu et al., 2018; Zhang et al., 2008). Most existing assessments have focused on individual pesticide exposures, which likely underestimate the real toxicity risks

encountered under field conditions (Arakawa et al., 2011; Kalita et al., 2017).

This review consolidates current knowledge on the influence of both conventional and emerging insecticides within the mulberry–silkworm ecosystem. It highlights critical aspects such as toxicological responses, residue dynamics, biochemical and physiological alterations, and sublethal effects on silkworm health and productivity. Furthermore, it underscores the significance of adopting sustainable pest management strategies that minimize pesticide risks while ensuring the economic viability of sericulture.

2. CLASSES OF INSECTICIDES, THEIR MODES OF ACTION, AND THEIR IMPACT ON SILKWORM

2.1 Organophosphates

Organophosphates (OPs) represent one of the most widely used groups of synthetic insecticides across agriculture, livestock, and forestry (Yu et al., 2011). Organophosphate insecticides include widely used compounds such as fenitrothion, ethion, dichlorvos, malathion, diazinon, and phoxim, among others (Hazarika et al., 2024). Compounds such as fenitrothion and ethion are broad-spectrum insecticides with strong knockdown potential against chewing and sucking pests (Nath & Kumar, 1999). Fenitrothion is particularly effective against pests of paddy and wheat, while ethion is commonly employed against mites on cattle and citrus, which are often cultivated in proximity to mulberry plantations (Nath & Kumar, 1999). Even in trace concentrations, these insecticides are lethal to silkworms, severely affecting growth, reproduction, and physiological processes, leading to drastic reductions in silk yield (Nath, 1993).

The deleterious effects of OP insecticides on silkworms have been extensively documented (Kuwana et al., 1967; Kashi, 1972; Bhosale et al., 1988; Radhakrishna & Delvi, 1992). Their principal toxic action is the irreversible inhibition of acetylcholinesterase (AChE), resulting in the accumulation of acetylcholine in synaptic clefts, hyperexcitation of neurons, disruption of energy metabolism in nerve cells, and ultimately death (Mileson et al., 1998; Nath & Kumar, 1999). Dichlorvos is specifically recommended for managing sucking pests in mulberry (Dandin et al., 2003), yet even this insecticide poses

significant risks to silkworm rearing. Beyond neurotoxicity, OPs also induce oxidative stress through the excessive production of reactive oxygen species (ROS). During pesticide metabolism, ROS initiate lipid peroxidation, leading to the formation of malondialdehyde (MDA) and trans-4-hydroxy-2-nonenal (4-HNE). Elevated MDA levels are strongly associated with cell and tissue damage in animals, including insects (Suwalsky et al., 2001; Marnett et al., 2003). Thus, OP exposure not only compromises neural integrity but also damages silkworm tissues at the cellular level.

Several OP insecticides, such as dichlorvos and phoxim, are widely used for controlling mushroom flies, aphids, spider mites, caterpillars, thrips, and whiteflies, and are even employed to treat parasitic worm infections in livestock and humans (Zhu et al., 2002; Yarsan & Cakir, 2006). However, their secondary toxic effects on non-target organisms, including silkworms, remain a significant challenge (Vyjayanthi & Subramanyam, 2002a). Given their acute toxicity, wide usage, and persistence in the environment, OPs continue to pose a severe threat to sericulture and highlight the need for careful regulation and integrated pest management (Gupta, 2011).

2.2 Carbamates

Carbamates are a major class of synthetic insecticides that act as reversible acetylcholinesterase (AChE) inhibitors, contrasting with organophosphates, which inhibit the enzyme irreversibly (Colovic et al., 2013). By blocking AChE, carbamates interfere with the breakdown of acetylcholine in synaptic clefts, causing overstimulation of cholinergic receptors, disruption of nerve impulse transmission, and ultimately paralysis and death of insects. Their reversible binding confers comparatively lower persistence of toxicity compared to organophosphates, yet they remain highly effective and broad-spectrum insecticides (Buhroo et al., 2016; Hazarika et al., 2024).

Chemically, carbamate insecticides are characterized by the carbamate ester functional group. Widely known examples include carbaryl (Sevin), carbofuran (Furadan), aldicarb, ethienocarb, and fenobucarb, which are extensively applied against chewing and sucking insect pests across agricultural systems (Buhroo et al., 2016). Carbaryl was the first carbamate marketed as an insecticide and continues to be

one of the most frequently used compounds within this group (Mora-Gutiérrez et al., 2021). Other commonly applied carbamates include aminocarb, carbendazim, carbofuran, mancozeb, and thiodicarb, reflecting the chemical diversity and wide pesticidal utility of this group (Athiappan et al., 2022; Voris et al., 2024; Mdeni et al., 2022).

Beyond their role as insecticides, carbamates have been formulated for use as fungicides and herbicides, thereby expanding their significance in integrated crop protection systems (Athiappan et al., 2022). However, their widespread application poses potential ecological and health risks. In sericulture, residues of carbamates on mulberry leaves can prove hazardous to *Bombyx mori*, given the silkworm's high sensitivity to neurotoxic agents and limited detoxification capacity. Although carbamates are considered somewhat safer compared to organophosphates due to reversible AChE inhibition, repeated or sublethal exposure may still result in growth delays, altered feeding behaviour, and reduced cocoon quality.

2.3 Neonicotinoids

Neonicotinoids represent the most widely used class of systemic insecticides worldwide, increasingly adopted in sericulture regions due to their perceived advantages over earlier generations of pesticides such as organophosphates, carbamates, and pyrethroids (Loknath et al., 2025) (Fig. 1). Together with fipronil, they account for nearly one-third of the global insecticide market (Delso et al., 2014). Their extensive use, combined with herbicides applied in mulberry gardens to compensate for labour shortages, has led to higher levels of residual toxicity on mulberry leaves. This residue accumulation raises the risk of disorders such as non-spinning syndrome and even large-scale mortality of silkworms (El-Ashram et al., 2022). Consequently, both the physiology and behaviour of *Bombyx mori* are adversely affected by neonicotinoid exposure.

Toxicological investigations confirm the complex and multifaceted impacts of neonicotinoids on silkworms. For instance, acetamiprid has been shown to disturb the reproductive system, adversely affect the next generation, and trigger toxic responses mediated by detoxification enzymes (Cheng et al., 2019; Wang et al., 2020). Imidacloprid, one of the most widely studied molecules, demonstrates high acute toxicity in *B.*

mori (Zhang et al., 2021), while dinotefuran exposure damages silkworm cells and disrupts metabolic processes, ultimately impairing normal growth and development (Xu et al., 2022). Similarly, acetamiprid has been documented to negatively affect caterpillars of *B. mori* (Cheng et al., 2019).

Mechanistically, neonicotinoids act on insect nicotinic acetylcholine receptors (nAChRs), located at postsynaptic membranes, causing persistent depolarization and neuronal overstimulation. This leads to paralysis and eventual death (Shakthiet al., 2015). Imidacloprid and clothianidin are commonly used in sericulture and surrounding crop ecosystems (Hazarika et al., 2024). Importantly, these insecticides are systemic: they are absorbed by plants and transported through vascular tissues, rendering them effective against a wide range of piercing and sucking pests (Jeschke et al., 2011; Zhang et al., 2013). However, only about 5% of applied neonicotinoids are absorbed by crops, while the remaining active ingredients disperse into the environment, contributing to soil and water contamination and ecological risks (Sur & Stork, 2003; Goulson, 2014). Off-target drift from neighbouring agricultural fields has been reported to deposit residues on mulberry leaves, reducing silkworm growth and silk production (Gu et al., 2013).

Neonicotinoid compounds include imidacloprid, clothianidin, thiamethoxam, dinotefuran, thiacloprid, acetamiprid, and nitenpyram (Thany et al., 2023; Sun et al., 2023). Nitenpyram, in particular, is promoted in China as a replacement for highly toxic organophosphates due to its systemic action and strong efficacy against piercing-sucking insects (Jeschke et al., 2011; Zhang et al., 2013). Nevertheless, its widespread use still presents risks to non-target organisms such as bees and silkworms (Santorium et al., 2019). Sublethal exposure in silkworms has been shown to prolong larval development, reduce larval weight, and impair cocooning performance. Thiamethoxam and imidacloprid also alter detoxification enzyme activity, notably glutathione-S-transferase and carboxylesterase, indicating metabolic stress responses in *B. mori* (Chen et al., 2023).

2.4 Diamides and Avermectins

Diamides and avermectins are relatively modern classes of insecticides; diamides (e.g., chlorantraniliprole, flubendiamide) act as selective modulators of ryanodine receptors,

causing unregulated calcium release and muscular dysfunction, while avermectins (e.g., abamectin, emamectin) target glutamate- and GABA-gated chloride channels, leading to neural hyperpolarization, paralysis, and eventual insect death.

Chlorantraniliprole, an anthranilic diamide, is widely used in crops such as rice, coffee, sugarcane, apple, and peach due to its high efficacy against Lepidopteran pests (Lahm et al., 2007; Temple et al., 2009). In addition, it has demonstrated activity against other insect orders including Coleoptera, Diptera, Isoptera, and Hemiptera (Hannig et al., 2009; Mao et al., 2019). Once ingested, chlorantraniliprole functions as a selective agonist of ryanodine receptors, a class of intracellular calcium channels critical for excitation-contraction coupling in muscle cells (Wood & Goulson, 2017). Its overstimulation leads to uncontrolled Ca²⁺ release from the sarcoplasmic reticulum, impairing muscle regulation and resulting in sustained contraction and paralysis (Dinter et al., 2010). Characteristic poisoning symptoms include rapid cessation of feeding, lethargy, regurgitation, muscle paralysis, and ultimately death (Cordova et al., 2006; Sattelle et al., 2008; Chen et al., 2010). Although not directly applied to mulberry crops, off-target drift from nearby fields has been associated with cocoon losses in Brazilian sericulture farms (Munhoz et al., 2013).

The domesticated silkworm (*Bombyx mori*) is particularly sensitive to these new molecules. Chlorantraniliprole (Coragen), imidacloprid (Confidor), and fipronil (IXUS), all targeted primarily against Lepidoptera, have been reported to induce larval mortality and reduce cocoon production even with indirect exposure (Vassarmidaki et al., 2000). This underscores the susceptibility of sericulture systems to collateral effects of agricultural insecticides.

Avermectins, including abamectin and emamectin, act through a distinct mechanism by binding to glutamate- and GABA-gated chloride channels, enhancing chloride ion influx and thereby inducing hyperpolarization of nerve and muscle cells. This disrupts neural transmission, causing paralysis and death. Despite being considered safer alternatives than organophosphates, avermectins are not entirely harmless; their residues in mulberry leaves can lead to significant silkworm mortality, delayed larval development, and deterioration of cocoon traits when exposure coincides with rearing cycles.

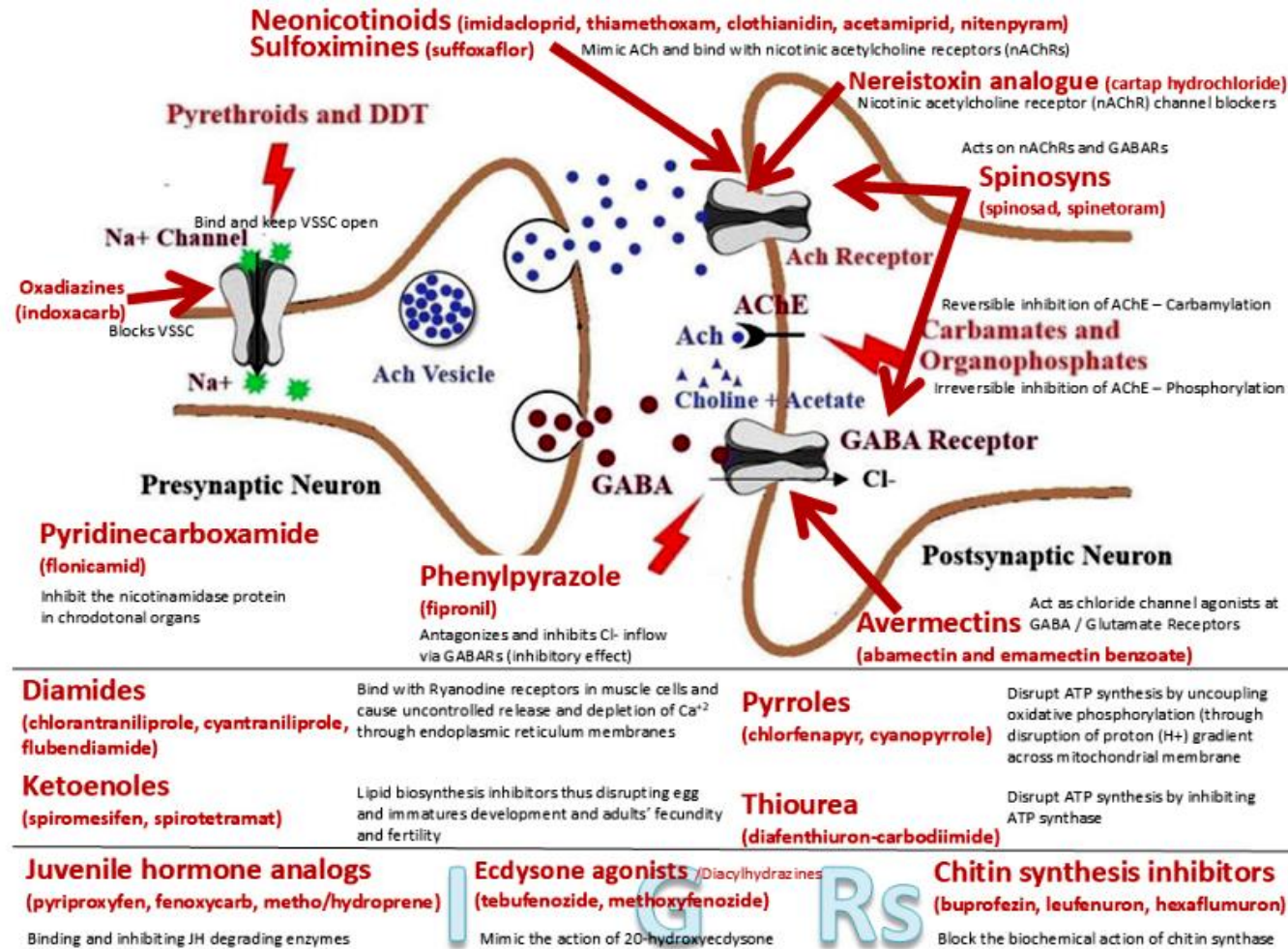


Fig. 1. Diagrammatic representation of the primary modes of action and target sites of major groups of synthetic insecticides in insects, including organophosphates, carbamates, neonicotinoids, diamides, and avermectins

(Source: Majeed et al., 2025; Shamjana & Grace, 2022)

3. RESIDUAL EFFECTS ON SILKWORMS

Residual pesticide contamination on mulberry foliage represents the most critical pathway through which silkworms encounter toxicants. Evidence from both laboratory and field investigations demonstrates that residues from synthetic insecticides exert profound biological effects, even at sub-lethal doses. These impacts range from acute toxicity, which causes considerable larval mortality within short exposure periods, to subtle physiological disruptions that compromise growth and developmental processes. Pesticides often persist on mulberry leaves after application, resulting in extended exposure for silkworms feeding on treated foliage (Kordy, 2014). Alterations in haemolymph ion composition, decreased metabolic stability, and impaired molting cycles further illustrate the biochemical stress induced by such residues. Beyond physiological harm, these disruptions translate into tangible economic losses, including reduced cocoon yield, lower shell ratio, and compromised filament quality, posing a threat to the sustainability and profitability of sericulture.

From a biochemical perspective, understanding the persistence and degradation dynamics of insecticide residues in the silkworm rearing environment is crucial (Hazarika et al., 2024). These residues can accumulate in mulberry foliage and soil, resulting in prolonged exposure and potential bioaccumulation in silkworm tissues (Muthusamy et al., 2016). Continuous application further leads to residue buildup in the soil, which adversely affects soil health, alters microbial diversity, and disrupts nutrient cycling (Soliman & EL Sherif, 2019). Therefore, effective residue monitoring, strict compliance with recommended application protocols, and observance of withdrawal periods are indispensable to mitigate contamination risks and maintain product safety for consumers (Sambanik & Indira 2012).

Field data also emphasize the scale of economic loss due to pesticide contamination. Sik et al., (1976) reported that yield reduction in sericulture exceeded 1.4% due to the indirect effects of pesticide application. Among contamination sources, pesticide use in rice fields accounted for 49.4%, followed by fruit orchards (21.2%) and vegetable cultivation (12.3%). These findings underscore the extreme sensitivity of silkworms, which cannot tolerate even trace levels of toxicants, making selective pest control strategies essential for mulberry ecosystems.

Several studies have demonstrated that organophosphate-based insecticides exert strong residual toxicity on silkworms. Ma et al., (2006) documented the detrimental effects of compounds such as diazinon, dichlorvos, phoxim, and triazophos following mulberry foliage treatment, which disrupted silkworm health. Persistence of residual toxicity was also observed with dimethoate and dichlorvos, reinforcing the potential risk posed by these chemicals in sericulture ecosystems (Yeshika et al., 2019). Similarly, Vasanth et al., (2025) found that residues of chlorantraniliprole (0.02%), imidacloprid (0.05%), and fipronil (0.2%) significantly impaired silkworm development and cocoon quality when leaves were fed 5–30 days after spraying. Imidacloprid exhibited the strongest effect, reducing the third-instar duration from 84.57 h at 5 DAS to 72.45 h at 25 DAS, while chlorantraniliprole caused gradual changes. Fipronil induced shorter instar and moulting durations than the other two. Although cocoon yield and shell weight improved over time, untreated controls maintained superior quality, including the highest shell ratio (54%) and finest silk denier (2.5). These observations stress the need for safe waiting periods and integrated pest management practices to mitigate residue hazards.

In addition to growth-related disruptions, residues also induce acute physiological and behavioral symptoms. Lokanath et al., (2025) reported that pesticide contamination in mulberry ecosystems severely affects *Bombyx mori*, causing vomiting, flaccidity, rectal protrusion, chain-like feces, non-spinning syndrome (NSS), and mortality. Probit analysis revealed that ≥ 0.9 $\mu\text{g/mL}$ caused mortality, 0.025–0.0012 $\mu\text{g/mL}$ induced NSS, and ≤ 0.0006 $\mu\text{g/mL}$ allowed normal cocooning. Flubendiamide exhibited the highest toxicity, causing 100% mortality even at 0.006 $\mu\text{g/mL}$, while imidacloprid and chlorantraniliprole significantly reduced spinning efficiency (Lokanath et al., 2025; Munhoz et al., 2013). Neonicotinoids, even at trace levels, triggered rapid symptoms such as food refusal and body spasms (Avramova et al., 2012), corroborating reports of prolonged larval growth and poor cocoon traits under sub-lethal exposure (Chen et al., 2023). A field survey identified pesticide residues in 35 out of 62 samples, primarily due to pesticide air-drift and indiscriminate application practices (Jyothi et al., 2019; Lokanath et al., 2025). At the biochemical level, imidacloprid strongly inhibited acetylcholinesterase (AChE), validating its potential as a biomarker for

pesticide exposure (Lokanath *et al.*, 2025). Moreover, sub-lethal pesticide exposure increased vulnerability to viral infections, further amplifying crop losses (Gu *et al.*, 2017).

Commercial parameters and reproductive traits are equally compromised by residual contamination. Sandhya *et al.*, (2024) reported that chlorfenapyr (10EC) and novaluron (10EC) caused the highest larval mortality (13.93% and 12.35%), whereas carbofuran (3G), dimethoate (30EC), and azadirachtin (0.03EC) were comparatively safer (>90% survival). Residual toxicity decreased fifth-instar larval weight (2.13 g) and cocoon shell ratio (14.92%) under novaluron treatment compared to controls (3.17 g and 18.86%). Cocoon and shell weights were significantly lower in chlorfenapyr and novaluron treatments, and reproductive performance suffered, with moths laying fewer eggs (326.65–382.70) and exhibiting higher deformity rates than controls, which showed higher fecundity (510.35 eggs). These findings emphasize the necessity of strict adherence to safe harvest intervals to prevent residue-induced losses in sericulture.

The cumulative evidence illustrates that pesticide residues, irrespective of chemical class, exert long-lasting adverse effects on silkworm physiology, cocoon economics, and reproduction. These impacts not only compromise the quality of silk production but also pose significant ecological and economic challenges for the sericulture sector. Ensuring residue-free mulberry foliage through selective pesticide application, robust monitoring protocols, and integrated pest management remains critical to sustaining a healthy silkworm–mulberry ecosystem.

4. SUBLETHAL AND CHRONIC EFFECTS

The impact of insecticides on *Bombyx mori* extends far beyond acute toxicity. Many compounds induce chronic and sublethal effects, which, though less obvious, can profoundly impair larval physiology, growth, reproduction, silk production, and overall sericultural productivity. Understanding these chronic responses is essential to evaluate long-term pesticide risks to silkworm health and sericulture sustainability.

4.1 Chronic Effects

Several insecticides display cumulative toxicity even when acute lethality is not observed.

Chlorfenapyr, for example, was not acutely toxic to silkworm larvae, yet continuous feeding on contaminated leaves produced concentration-dependent chronic mortality (Stanley and Preetha, 2016). Similarly, chronic mortality occurred at chlorpyrifos concentrations below 1 mL/L (Lin *et al.*, 2009). Triazophos exposure, beginning from the second instar, also caused chronic mortality. The LC₅₀ at 48 hours after treatment (HAT) was progressively reduced across instars—1.54 mg/L at the third, 0.74 mg/L at the fourth, and 0.53 mg/L at the fifth instar indicating increasing susceptibility with larval development (Zhu *et al.*, 2006). These observations confirm that continuous exposure to low pesticide concentrations significantly compromises silkworm survival.

4.2 Sublethal Toxicity

4.2.1 Effect on food intake, growth and development

Sublethal exposure to pesticides profoundly alters feeding behavior, growth, and developmental physiology of the silkworm *Bombyx mori*. Multiple studies have demonstrated that pesticides interfere with nutrient ingestion, assimilation, and utilization, thereby impairing silkworm productivity and silk yield.

Organophosphates such as ethion and fenitrothion significantly reduce food consumption and frass output in fifth instar larvae, while sublethal doses (1/5 LD₅₀) paradoxically stimulate slight increases in feeding activity (Nath & Kumar, 1999; Nath, 2002). Dichlorvos and phoxim reduce feeding and induce vomiting, whereas pyrethroids including permethrin, tetramethrin, bifenthrin, and ethofenprox cause abnormal “S-” or “C-” shaped body contractions indicative of neurotoxicity (Zhang *et al.*, 2008) (Fig. 2). Similarly, fenvalerate reduces ingestion, assimilation, and conversion efficiency, thereby disrupting nutrient metabolism (Vyjayanthi & Subramanyam, 2002a).

Insect growth regulators (IGRs) also compromise food utilization. At doses exceeding 100 pg/larva, IGRs markedly reduce ingestion and food conversion efficiency (Leonardi *et al.*, 1996; Monconduit & Mauchamp, 1998; Kamimura & Kiuchi, 1998). Fenoxycarb specifically decreases ingestion efficiency and frass output in a dose-dependent manner (Assal, 1994; Leonardi *et*

al.,2001), while buprofezin-treated mulberry leaves significantly reduce larval weight (Vassarmidaki et al., 2000). Avermectins such as abamectin further decrease food intake, body mass, and conversion efficiency, although they increase approximate digestibility, suggesting metabolic stress (Zhang et al., 2006; Zhu et al., 2008). Interestingly, methoprene exhibits a biphasic effect: at optimal doses it enhances larval growth and cocoon weights, but beyond threshold levels, growth and silk yield decline sharply (Miranda et al., 2002).

Pesticides also prolong development and disrupt normal metamorphosis. Pyrethroids at 10 ng/L delay larval growth (Sun et al., 2002), while juvenile hormone analogues and methyl parathion extend larval and pupal durations, thereby reducing reproductive output and silk productivity (Akai & Kobayashi, 1971; Kumutha et al., 2009; Begum et al., 2011). Hexachlorocyclohexane exposure lowers pupal and shell weights and reduces adult emergence (Bhagyalakshmi et al., 1995). Among newer insecticides, chlorantraniliprole at 0.1–0.2 ppm



Fig. 2. Morphological and physiological abnormalities observed in fifth-instar silkworm larvae (*Bombyx mori*) following exposure to different pesticide treatments [Bifenthrin 10% EC (BIF), Flubendiamide 39.35% SC (FLU), Buprofezin 23.1% + Fipronil 3.85% SC (BUF), Emamectin benzoate 5% SG (EMB), Chlorantraniliprole 18.5% SC (CHL), and Imidacloprid 17.8% EC (IMI)]. Symptoms include: a) larval mortality, b) flaccid body, c) rectal protrusion, d) chain faeces, e) body shrinkage, f) partial spinning, g) vomiting, h) S-shaped contraction, and i) hook-shaped posture

(Source: Loknath et al., 2025)

has been shown to cause high mortality in silkworms, with symptoms including vomiting, flaccid bodies, rectal protrusion, chain feces, body shrinkage, partial or complete failure in spinning, and eventual death (Lionetto *et al.*, 2013; Loknath *et al.*, 2025). Dead larvae frequently adopt characteristic S-shaped or hook-shaped postures, which appear to be pesticide-specific. For instance, mortality associated with bifenthrin (BIF), flubendiamide (FLU), and imidacloprid (IMI) treatments is often accompanied by these distinct postural changes (Loknath *et al.*, 2025).

Neonicotinoids such as imidacloprid, even at trace levels, significantly impact *B. mori*, impairing growth and silk productivity (Avramova & Grekov, 2013). Similarly, chlorantraniliprole exposure leads to rapid cessation of feeding, lethargy, regurgitation, muscle paralysis, and eventual mortality in silkworms (Munhoz *et al.*, 2013). These findings demonstrate that pesticide-induced disruptions in feeding behavior and metabolism directly translate into delayed development, reduced cocoon production, and compromised sericulture sustainability.

4.2.2 Effect on cocooning and spinning

The cocooning process is highly sensitive to sublethal pesticide effects (Table 1). IGRs at low doses (50–100 pg/larva) caused non-spinning syndromes and dauer larval arrest even in late instars (Leonardi *et al.*, 1996; Monconduit & Mauchamp, 1998; Kamimura & Kiuchi, 1998). Fenoxycarb, when continuously administered, prolonged the larval period and suppressed

cocooning (Stanley and Preetha, 2016). Topical application of 60 pg/larva caused 50% dauer larvae (Leonardi *et al.*, 1996). Pyriproxyfen extended instar duration and reduced cocoon formation ability (Sun *et al.*, 2008). In contrast, the antijuvenile compound K-22 accelerated cocoon spinning in a dose-dependent manner (Asano *et al.*, 1984). Other insecticides including dimehypo, chlorfenapyr, and herbicides such as clodinafop-propargyl reduced cocooning rates, pupation, and adult emergence (Wang *et al.*, 1999; Stanley and Preetha, 2016; Yin *et al.*, 2008).

4.2.3 Effect on silk production and fiber quality

Sublethal doses of pesticides may impair both the quantity and quality of silk. Dimehypo exposure reduced silk production, damaged posterior silk gland ultrastructure, and disrupted fibroin biosynthesis (Wang *et al.*, 1999). Chlorantraniliprole contamination caused feeding cessation and thin-shelled cocoons (Munhoz *et al.*, 2013). Juvenile hormone analogues influenced cocoon and shell weights: methoprene increased cocoon parameters at low doses but negatively affected fibroin synthesis at higher concentrations (Miranda *et al.*, 2002; Begum *et al.*, 2011). Diflubenzuron altered silk gland growth and produced opaque glands (Kim & Sohn, 2001), while hexachlorocyclohexane impaired fibroin content and reduced silk thread quality (Bhagyalakshmi *et al.*, 1995). Conversely, juvenile hormone treatments (SJ-42-F, JH injections) have been reported to enhance fibroin biosynthesis and silk output under controlled conditions (Chowdhary *et al.*, 1986; Akai *et al.*, 1971).

Table 1. Effect of various pesticide concentrations on silkworm leading to various symptoms

Particulars	CHL	BIF	FLU	BUF	EMB	IMI
Range of Conc. (µg/ml)	500–0.00001	1000–0.00003	250–0.00006	1500–0.00006	250–0.00006	1500–0.00006
100% Mortality	≥ 0.025	≥ 0.2	≥ 0.006	≥ 0.04	≥ 0.012	≥ 0.35
LC ₅₀	0.002	0.022	0.003	0.009	0.001	0.013
Non-spinning	0.012–0.06	0.1–0.012	0.003–0.0003	0.02–0.001	0.006–0.003	0.17–0.04
Non-spinning (%)	56	100–66	100	60–53	60–53	66–43
Spinning	≤ 0.003	≤ 0.006	≤ 0.00001	≤ 0.001	≤ 0.0015	≤ 0.02
Spinning (%)	83–50	93–63	90–83	90–40	90–73	70–63
Flimsy Cocoons	0.003–0.015	0.006	0.00001–0.0006	Not Observed	0.0015–0.0007	0.02–0.00006

Bifenthrin 10% EC (BIF), Ryanoid – Flubendiamide 39.35% SC (FLU), Buprofezin 23.1% + Fipronil 3.85% SC (BUF), Avermectin – Emamectin benzoate 5% SG (EMB), Anthranilic diamide – Chlorantraniliprole 18.5% SC (CHL), and Neonicotinoid – Imidacloprid 17.8% EC (IMI). Among these, FLU at the lowest concentration led to 100% mortality. Non-spinning was observed in all treated larvae, while cocoon spinning ranged from 40% to 100%
(Source: Loknath *et al.*, 2025)

4.2.4 Effect on reproduction and fecundity

Larval exposure to pesticides adversely affects adult reproductive performance. Organophosphorus insecticides such as parathion, fenthion, isoxathion, diazinon, and disulfoton significantly disrupted mating behavior and reduced fecundity (Kuribayashi, 1981a). Similarly, chlordimeform, trifluralin, and metepa reduced egg production (Kuribayashi, 1981a). Hexachlorocyclohexane decreased egg numbers and hatching rates (Bhagyalakshmi *et al.*, 1995). Dichlorvos exposure reduced egg output by 11.3%, fertilization rates, and hatching percentages compared with controls (Kumutha *et al.*, 2013). Moreover, parathion and disulfoton acted as ovicides, with embryos dying shortly before or after hatching due to pesticide transfer through the eggs (Kuribayashi, 1981b).

4.2.5 Effect on hemolymph

Sublethal pesticide exposure disturbs hemolymph composition and enzyme activity. Ethion and fenitrothion reduced protein content and altered carbohydrate metabolism (Nath *et al.*, 1997; Nath, 2000). Fenoxycarb and pyriproxyfen inhibited protein synthesis (Monconduit & Mauchamp, 1998). Pyriproxyfen-treated larvae showed lower glucose and uric acid levels, while methoprene treatment increased trehalose and glycogen reserves (Etebari *et al.*, 2007; Begum *et al.*, 2011). Ethion and fenitrothion shifted metabolism from aerobic to anaerobic, reducing pyruvate and increasing lactate in hemolymph and fat body (Nath, 2000). Enzymatic activities, including alanine aminotransferase, aspartate aminotransferase, esterases, and phosphatases, were significantly reduced after exposure to various insecticides (Kordy, 2014; Kim, 2002; Zhan *et al.*, 2006; Etebari *et al.*, 2007).

4.2.6 Effect on fat bodies and cellular toxicity

Fat bodies exhibited marked metabolic disruptions. Ethion and fenitrothion depleted glycogen and proteins while enhancing glycogenolysis (Nath *et al.*, 1997; Nath, 2002). Methoprene increased glycogen and trehalose stores (Begum *et al.*, 2011). Phoxim exposure induced oxidative stress markers such as malondialdehyde, increased GST and cytochrome P450 activities, and caused apoptosis-like changes, including mitochondrial swelling (Yu *et al.*, 2011; Wang *et al.*, 2013; Gu *et al.*, 2014).

4.2.7 Effect on enzymes and hormones

Digestive and metabolic enzymes were significantly impaired. Fenvalerate exposure reduced amylase, sucrase, and protease activities while elevating trehalase (Vyjayanthi & Subramanyam, 2002b). Abamectin reduced amylase and sucrase activities (Zhu *et al.*, 2008). Phoxim decreased dehydrogenase activities in carbohydrate metabolism (Li *et al.*, 2012). Rac-metolachlor suppressed lactate dehydrogenase and alkaline phosphatase activity, influencing silk quality (Zhan *et al.*, 2006; Miao, 2002). Methoprene enhanced protease and mitochondrial enzyme activities, indicating boosted oxidative metabolism (Mamatha *et al.*, 2008). Diflubenzuron, however, blocked JH degradation, disrupting silk gland growth and protein synthesis (Riddiford, 1993; Okuda *et al.*, 1985; Garel, 1983; Susumuet *et al.*, 1984; Tomino, 1985). Fenoxycarb further inhibited responsiveness of prothoracic glands to PTTH, disturbing endocrine regulation (Monconduit & Mauchamp, 1998; Dedos & Fugo, 1999).

4.2.8 Genotoxicity of pesticides

Pesticides can cause DNA and gene-level alterations even without visible physiological symptoms. Clodinafop-propargyl exposure induced significant DNA damage in comet assays (Yin *et al.*, 2008). Avermectin exposure led to concentration-dependent DNA fragmentation and altered mRNA expression (Shen *et al.*, 2011). Phoxim exposure altered expression of 254–266 genes, including detoxification-related cytochrome P450s, esterases, and GSTs (Gu *et al.*, 2013; 2014). Detoxification-related genes such as CYP6ae22, CYP9a21, GSTo1, and Bmcce were strongly upregulated in midgut and fat body following phoxim treatment (Wang *et al.*, 2013). Acetylcholinesterase genes (Bm-AChE-1 and Bm-AChE-2) showed tissue-specific regulation after phoxim exposure, while BmGSTe8 was associated with phoxim resistance (Peng *et al.*, 2011; Yu *et al.*, 2011).

5. ENVIRONMENTAL AND ECOLOGICAL IMPLICATIONS

Silkworms are highly vulnerable to environmental pesticide contamination, as exposure occurs not only through direct application on mulberry but also via drift from adjacent rice, cotton, and tea fields, as well as horticultural crops where pesticide use is intensive. Aerial spraying,

surface runoff, and irrigation water act as important contamination routes, leading to the deposition of residues on mulberry leaves, which is the sole food source of *Bombyx mori* (Etebari et al., 2007; Zhang et al., 2008). For instance, sericulture farms in Assam, located adjacent to paddy and tea plantations, are frequently exposed to pesticide drift, while in southern Brazil, chlorantraniliprole application in sugarcane fields has been directly linked to silkworm crop losses and reduced cocoon yield (Bora et al., 2012; Munhoz et al., 2013).

Residues from insecticides not only accumulate on mulberry foliage but also persist in the soil, creating prolonged exposure risks for both the silkworm and the broader mulberry ecosystem. Studies indicate that pesticide residues can persist through several rearing cycles, leading to chronic bioaccumulation and compounding toxic effects in silkworm populations (Muthusamy & Rajakumar, 2016). Persistent residues also compromise sericulture sustainability, reduce farmer income, and may alter soil health by disrupting microbial communities and nutrient cycling processes (Soliman & Sherif, 2019).

From a biochemical standpoint, knowledge of residue persistence and degradation dynamics is essential to assess long-term risks. Monitoring and regulating residue levels, combined with adherence to safe withdrawal periods before leaf harvest, are vital to minimizing risks to silkworm health and ensuring consumer safety in silk-based products (Sambanai& Indira 2012).

The ecological implications of indiscriminate pesticide use extend beyond sericulture. Non-target and beneficial insects such as pollinators (bees, butterflies) and natural enemies (ladybirds, spiders) are highly susceptible to pesticide drift and residues, thereby destabilizing natural pest regulation mechanisms and reducing biodiversity (Stanley et al., 2016). Runoff into aquatic ecosystems results in accumulation of residues in water bodies, threatening fish, amphibians, and aquatic invertebrates. Continuous exposure contributes to pest resistance, necessitating higher doses or more potent chemicals, which in turn exacerbates environmental pollution (Sato & Sugihaya, 1984) (Fig. 3).

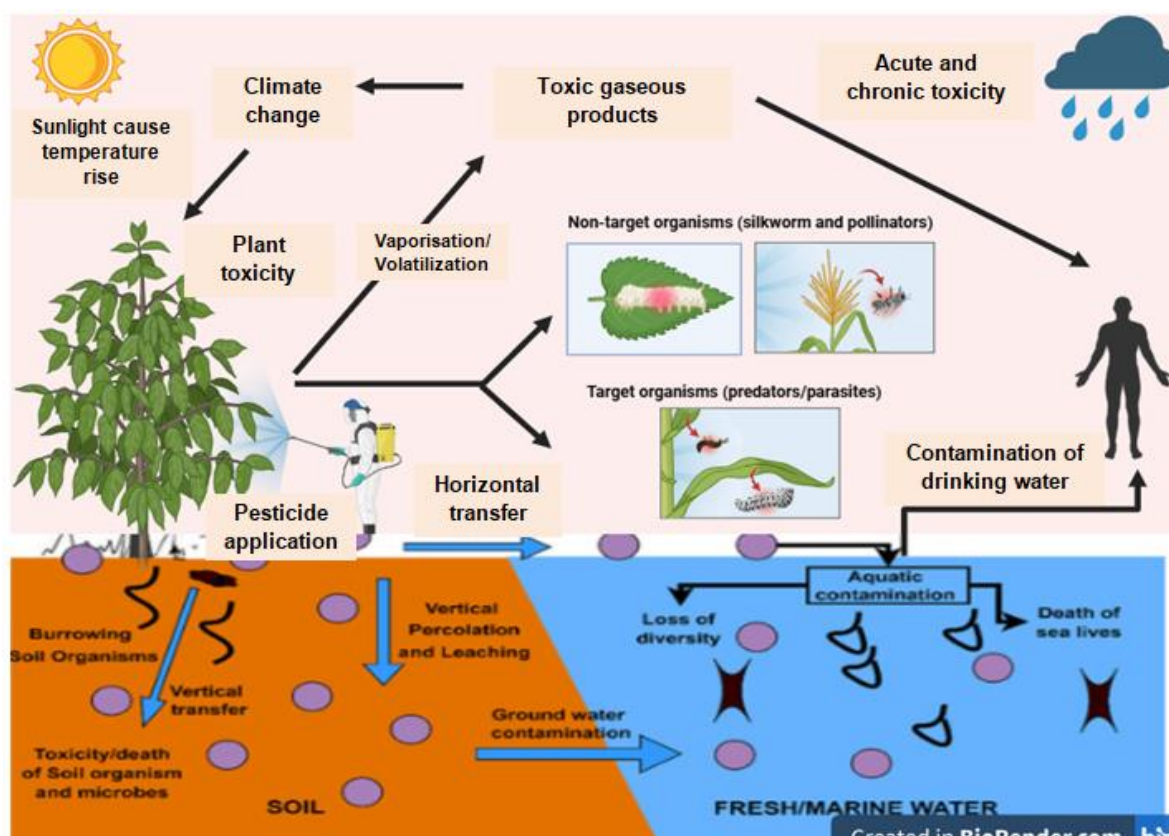


Fig. 3. Environmental and ecological implications of pesticide application in sericulture ecosystems

At a global scale, pesticide pollution has become a major ecological and human health concern. Pesticides are biologically active compounds that interfere with essential processes in living organisms, and their classification spans insecticides, fungicides, herbicides, nematicides, molluscicides, and rodenticides, among others (Fascendini *et al.*, 2002; Carvalho *et al.*, 2006). However, it is estimated that less than 0.1% of applied pesticides actually reach target pests, leaving over 99% as unintended contaminants in soil, air, water, or vegetation (Buhroo *et al.*, 2016). This inefficiency greatly magnifies ecological risks by contributing to soil and water contamination and the transfer of residues across the food chain.

Among modern pesticides, neonicotinoids are particularly concerning due to their systemic nature. Once absorbed, they accumulate in plant vascular tissues and leaves, protecting crops from herbivorous insects. Yet, studies show that only ~5% of applied neonicotinoids are actually taken up by plants, with the remainder dispersed into the wider environment where they persist and accumulate (Sur & Stork, 2003; Goulson, 2014). This persistence increases the likelihood of residues in soil and water systems. Furthermore, degradation products of compounds such as nitenpyram have been identified in water treatment facilities, raising concerns about contamination of drinking water sources (Noestheden *et al.*, 2016).

Taken together, these findings underscore that pesticide pollution in sericulture landscapes is not an isolated concern but part of a wider environmental crisis that threatens biodiversity, ecosystem services, and human health. The

sensitivity of silkworms to even trace residues positions them as effective sentinel organisms for ecotoxicological monitoring (Terçariol & Godinho, 2011). A balanced approach to pest management—integrating ecological sustainability with economic viability—is therefore indispensable to secure the long-term resilience of both sericulture and surrounding ecosystems (Kuribayashi, 1988).

6. SUSTAINABLE PEST MANAGEMENT IN MULBERRY ECOSYSTEMS

The sustainability of mulberry-based sericulture relies heavily on effective pest management strategies that minimize environmental risks while ensuring silkworm safety and crop health. While conventional chemical insecticides are effective in controlling pest outbreaks, their indiscriminate use often results in residual toxicity, pest resistance, and ecological imbalances. These challenges have driven the need for safer, eco-friendly alternatives that combine biological control agents, botanical formulations, cultural practices, and need-based chemical applications within a structured integrated pest management (IPM) framework. Prioritizing selective and judicious pesticide use, adherence to safe harvest intervals, and the inclusion of bio-rational inputs is essential to reduce the chemical footprint in mulberry ecosystems. Such strategies not only safeguard silkworm health and improve silk quality but also support soil fertility, biodiversity conservation, and the long-term economic sustainability of sericulture. The major strategies for achieving sustainable pest management in mulberry ecosystems are summarized in Table 2 below.

Table 2. Sustainable pest management strategies for mulberry ecosystems

Strategy	Description	References
Integrated Pest Management (IPM)	Integrated Pest Management (IPM) focuses on a combination of strategies, such as encouraging natural enemies, implementing cultural measures like crop rotation, and reserving chemical control as the last line of defense. This holistic approach significantly lowers dependency on insecticides and mitigates adverse environmental effects.	Mukhopadhyay <i>et al.</i> , (2016)
Alternative Pest Control	The application of plant-derived extracts or microbial agents as substitutes for synthetic insecticides offers a strategy to minimize chemical inputs in pest management.	Hazarika <i>et al.</i> , 2024
Selective and Low-Persistence Insecticides	Apply insecticides with minimal impact on non-target species and reduced environmental persistence, coupled with regular monitoring and dosage adjustments to limit ecological consequences.	Hazarika <i>et al.</i> , 2024

Strategy	Description	References
Protective Buffer Zones with Management Protocols	Create buffer zones surrounding sericulture farms to prevent pesticide runoff into nearby water bodies, and adopt best management practices by optimizing application timing to minimize non-target exposure and reduce spray drift.	Stanley & Preetha, (2016)
Systematic Surveillance and Threshold Determination	Consistent surveillance of pest populations combined with the establishment of action thresholds for insecticide application can effectively minimize unnecessary chemical interventions.	Hazarika et al., 2024
Biological Control	Promoting the activity of natural predators and parasitoids offers an effective means to suppress pest populations, thereby reducing the dependence on chemical insecticides.	Singh & Maheshwari, (2002).
Cultural Management Approaches	Adopting appropriate pruning techniques, implementing crop rotation, and synchronizing pesticide applications can enhance pest control efficiency while reducing environmental contamination.	Hazarika et al., 2024
Capacity Building and Skill Development	Raising awareness among farmers, agricultural workers, and stakeholders regarding the ecological consequences of insecticide use, while encouraging the adoption of practices that mitigate these impacts, is essential for sustainable pest management.	Hazarika et al., 2024

Implementing these strategies requires a paradigm shift from reliance on chemical interventions to an integrated, knowledge-based approach. Strengthening research on bio-rational alternatives, improving farmer training programs, and developing region-specific pest management modules will be crucial in ensuring that mulberry ecosystems remain productive, resilient, and environmentally sustainable in the face of evolving pest dynamics and climate challenges.

7. FUTURE DIRECTIONS

The mitigation of pesticide-induced risks in mulberry–silkworm ecosystems necessitates a multidimensional research and management framework. Future investigations should focus on the quantitative modeling of pesticide residue dynamics in mulberry foliage, soil, and silkworm biological matrices to accurately predict persistence, degradation kinetics, and potential bioaccumulation. Establishing robust residue surveillance protocols supported by chromatographic and spectrometric techniques will enable precise risk assessment and enforceable residue threshold regulations for sericulture.

Advancement of Integrated Pest Management (IPM) remains a critical research priority. Emphasis should be placed on designing region-specific IPM modules that integrate biological control agents, botanical insecticides, cultural

operations, and selective low-persistence chemicals. Development of decision-support systems based on pest population thresholds and real-time field monitoring will further optimize chemical interventions, thereby reducing unnecessary applications.

The exploration of next-generation bio-rational insecticides with enhanced target specificity and reduced off-target toxicity is imperative. Innovations in nano-formulated pesticides, controlled-release delivery systems, and biodegradable carriers should be accompanied by comprehensive ecotoxicological evaluations to ascertain safety in sericulture ecosystems.

At the molecular level, future research should leverage functional genomics, transcriptomics, and proteomics to elucidate detoxification pathways, stress-response mechanisms, and endocrine disruptions induced by sublethal pesticide exposure. These insights may inform selective breeding programs or genetic interventions aimed at improving silkworm resilience without compromising cocoon and silk quality.

In addition, predictive modeling under climate change scenarios is essential to anticipate shifts in pest dynamics, pesticide efficacy, and residue behavior, ensuring adaptive management strategies. Finally, strengthening capacity-building initiatives, farmer training, and policy-

driven incentives for the adoption of eco-friendly pest control technologies will be fundamental to operationalizing sustainable sericulture.

Collectively, these future directions underscore the necessity of transitioning from chemically intensive practices toward knowledge-based, ecologically harmonized pest management systems that ensure silk productivity, environmental integrity, and economic sustainability.

8. CONCLUSION

The findings underscore the critical significance of adopting sustainable pest management practices in the mulberry–silkworm production system to minimize pesticide-induced risks. Conventional chemical pesticides, while effective in controlling pest outbreaks, often pose severe challenges such as residue accumulation, silkworm toxicity, and ecological imbalance. These impacts not only compromise cocoon quality and yield but also threaten the long-term sustainability of sericulture.

The review highlights that effective pest management must move beyond reliance on chemical inputs toward a holistic, integrated approach. Strategies such as Integrated Pest Management (IPM), which incorporates biological control agents, botanical formulations, cultural operations, and need-based chemical applications, offer a promising pathway to reduce dependency on hazardous pesticides. Moreover, adopting safer chemicals with low persistence, combined with rigorous adherence to recommended waiting periods, can substantially lower residual toxicity in mulberry foliage and silkworm tissues. Another crucial aspect is the generation and dissemination of awareness among stakeholders, including farmers and extension personnel, regarding pesticide hazards and best practices for residue mitigation. Continuous monitoring of pesticide residues and strengthening of regulatory frameworks are imperative to maintain product safety and environmental integrity.

In essence, the future of sustainable sericulture hinges on the convergence of scientific innovation, eco-friendly pest management, and farmer-centric capacity-building initiatives. By prioritizing environmentally compatible solutions and rational pesticide use, the industry can achieve the dual objective of maintaining high-

quality silk production while safeguarding ecological health.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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COMPETING INTERESTS

Authors have declared that they have no known competing financial interests or non-financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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