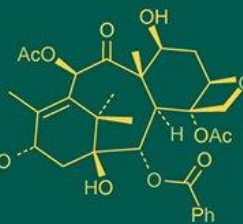
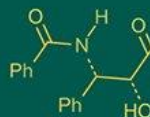


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Insecticide resistance and management in diamondback moth (*Plutella xylostella* L.): A Review

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Abstract

Diamondback moth (*Plutella xylostella* L.) is one of the most destructive pests of cruciferous crops worldwide and has developed resistance to over 100 insecticidal compounds due to prolonged and indiscriminate use of insecticides. This review summarizes the major resistance mechanisms in DBM (diamondback moth) broadly categorized into behavioral and physiological adaptations. Behavioral resistance includes avoidance of insecticide-treated surfaces, whereas physiological mechanisms involve reduced cuticular penetration, enhanced metabolic detoxification via cytochrome P450s, glutathione S-transferases (GSTs) and esterases and target-site insensitivity arising from mutations in acetylcholinesterase (AChE), ryanodine receptors (RyRs) and GABA receptors. This review also outlines key resistance detection methods, including toxicological bioassays for LC₅₀ estimation and biochemical assays for detoxification enzyme activity. To mitigate escalating resistance, various Insecticide Resistance Management (IRM) strategies are highlighted, such as window rotation of insecticides with different modes of action, use of synergists like piperonyl butoxide (PBO) and botanical oils and application of insecticide mixtures for improved efficacy. Emerging molecular approaches including RNA interference (RNAi) and CRISPR/Cas9 genome editing offer promising avenues for restoring susceptibility and reducing selection pressure in field populations. Collectively, these strategies provide a sustainable and integrated framework for managing insecticide resistance in *P. xylostella*.

Keywords: Diamondback moth, insecticide resistance, resistance mechanisms, integrated pest management (IPM)

Introduction

Excessive and continuous insecticide use has resulted in three major issues, known as the “3R’s”: resurgence, residue and resistance. Resurgence refers to the rapid re-increase of pest populations following repeated insecticide applications, while residues represent the persistent traces of insecticides in the environment and food systems. Resistance is defined as a heritable reduction in pest sensitivity evident when an insecticide repeatedly fails to provide effective control despite being used according to label recommendations (Anonymous, 2025a) ^[1]. Multiple factors contribute to resistance development in the diamondback moth (DBM), including improper insecticide use, high application frequency, enhanced detoxification mechanisms and rapid reproductive capacity. Globally, the APRD (Arthropod Pesticide Resistance Database) reports that up to 2024, 632 arthropod species have developed resistance to 364 chemical compounds, accounting for 18,871 resistance cases (Payumo *et al.*, 2024) ^[40]. DBM ranks first among resistant species, exhibiting resistance to over 104 insecticidal compounds (Eslamlou and Ito, 2025) ^[10]. Within the genus *Plutella*, six species are recognized: *P. xylostella*, *P. antiphona*, *P. australiana*, *P. armoraciae*, *P. hass* and *P. sera*, of which *P. xylostella* is the most widely distributed worldwide (Kfir, 1998) ^[22].

The diamondback moth (*Plutella xylostella* L.), believed to have originated in the Mediterranean region, was first reported in India by Fletcher in 1914. It is now recognized as one of the most destructive pests of cruciferous crops across major states, including Punjab, Uttar Pradesh, Bihar, Maharashtra, Tamil Nadu, Karnataka and Gujarat. DBM is highly adaptable to diverse climatic conditions, exhibits rapid dispersal ability and has a short life cycle. Being oligophagous and multivoltine, it can complete 4 to 20 generations per year depending on temperature.

The gregarious larvae, equipped with chewing mouthparts, feed continuously on leaf tissues, causing characteristic windowing holes and severe defoliation. Damage may extend from seedlings to head formation stages, occurring year-round except during the rainy season (Mubashir and Seram, 2022) ^[35]. In the world diamondback moth first time

developed resistance against DDT in 1953 at Lembang, Indonesia (Johnson, 1953) ^[20]. Later, in 1989 it also developed resistance to *Bacillus thuringiensis* var. *kurstaki*. In India, they first time they develop resistance against DDT and Parathion in 1968, while to from *B. thuringiensis* *kurstaki* in the year 1995 (Rabindra *et al.*, 1995) ^[42].

Table 1: Resistance reported against *P. xylostella* from the world

Year	Insecticides / Bt Toxin	Country	Citation
1953	DDT	Indonesia	Johnson (1953) ^[20]
1967	Carbaryl	Vietnam	FAO (1967) ^[12]
1968	DDT, parathion	India	Varma and Sandhu (1968) ^[58]
1989	<i>Bacillus thuringiensis</i> var. <i>kurstaki</i>	USA (Hawaii)	Tabashnik <i>et al.</i> (1989) ^[54]
1999	Chlorpyrifos	Australia	Baker and Kovaliski (1999) ^[4]
1995	<i>B. thuringiensis</i> var. <i>kurstaki</i>	India	Rabindra <i>et al.</i> (1995) ^[42]
2000	<i>B. thuringiensis</i> var. <i>kurstaki</i> Biobit®	India	Mohan and Gujar (2000) ^[34]
2010	Abamectin, Emamectin benzoate	China	Pu <i>et al.</i> (2010)
2011	Chlorfenapyr, Spinosad, Spinetoram, Deltamethri	Brazil	Oliveira <i>et al.</i> (2011) ^[36]
	Indoxacarb, Abamectin, Lufenuron	Brazil	Santos <i>et al.</i> (2011) ^[45]
2014	Chlorantraniliprole	China	Gong <i>et al.</i> (2014) ^[13]
2016	Beta-cypermethrin, Chlorfluazuron, Chlorfenapyr, Indoxacarb, Abamectin, Spinosad	China	Zhang <i>et al.</i> (2016) ^[62]
2017	Indoxacarb	China	Zhang <i>et al.</i> (2017) ^[61]
	Chlorantraniliprole	Brazil	Ribeiro <i>et al.</i> (2017) ^[44]
	Chlorfenapyr	Brazil	Lima and Siqueira (2017) ^[26]
2018	Dichlorvos, Fenvalerate	India	Meghana <i>et al.</i> (2018) ^[33]
2019	Spinosad	Malaysia	Knight and Elias (2019) ^[23]
2020	Chlorantraniliprole, Cyantraniliprole, Flubendiamide	India	Chaudhari (2020) ^[9]
	Cypermethrin, Cartap hydrochloride, Acephate, Cry2Ab	India	Sunitha <i>et al.</i> (2020) ^[53]
2021	Spintoram	India	Tamilselvan <i>et al.</i> (2021) ^[55]
2022	Chlorpyrifos, Indoxacarb, Chlorantraniliprole	China	Wang <i>et al.</i> (2022) ^[59]
2024	Lepimectin, Lufenuron, Fipronil and Dieldrin	Japan	Oplopoiou <i>et al.</i> (2024) ^[37]

Types of resistance found in *P. xylostella* (IRAC, 2011)

In diamondback moth, majorly three types of resistance are found like.

- **Cross resistance:** When insect develop resistance against one insecticide they also develop resistance against another insecticide belong to same class of insecticide without exposure of second one (Ishadi and Mazlan *et al.*, 2015) ^[18].
- **Example:** DBM develop resistance against chlorantraniliprole and also they develop resistance against flubendiamide because both insecticide belong to same class diamide (IRAC, Group 28)
- **Multiple resistance:** When insects develop resistance against more than one insecticide or more than one class of insecticide, it is called as multiple resistance.
Example: DBM develop resistance against many class of insecticide like, organochlorines, organophosphates, carbamates, synthetic pyrethroids and diamide.
- **Polygenic resistance:** When insects develop resistance involving more than one gene, it is called as polygenic resistance.

Example: DBM has developed resistance to theCry1Ac and Cry2Ab genes, both genes are derived from *B. thuringiensis*.

Mechanisms of development of resistance

Insects develop resistance mainly ways reduced target site sensitivity increased detoxification by detoxifying enzymes (cytochrome P450s, esterases or glutathione S-transferases) and behavioural mechanism (Hubbard and Murillo, 2024) ^[15]. Target sites are the parts of the insect where insecticides act. When these sites change due to mutations, insecticides can not bind properly, so they cause less harm (Charaabi *et al.* 2018) ^[8]. Detoxification occurs when enzymes quickly break down insecticides into metabolites. Insects may also resist insecticides by reducing penetration through the cuticle or by changing their behaviour to avoid exposure to insecticide (Karaagac, 2012) ^[21]. Many mechanisms are responsible for the development of resistance in insects but, two major mechanisms are commonly observed in insects: behavioural mechanisms and physiological mechanisms (Sparks *et al.*, 1989) ^[52].

Behavioural mechanisms

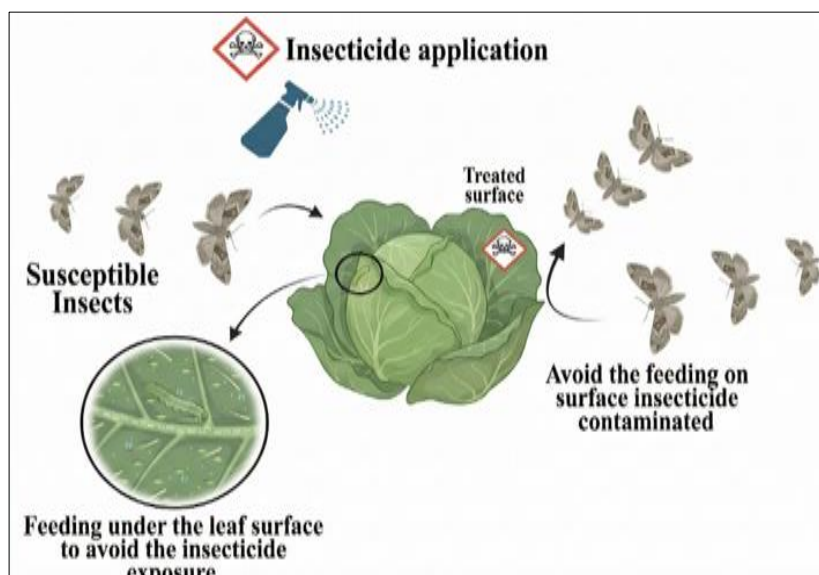


Fig 1: Systemic diagram of behavioural resistance, where insects avoid treated areas and feed on non-treated parts of the host plant

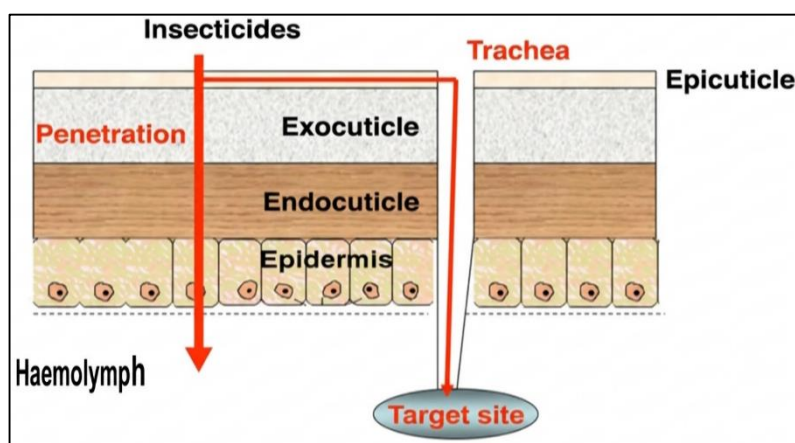
Sparks *et al.* (1989) [52] described behavioural resistance as adaptive changes in insect behaviour that reduce or prevent exposure to insecticides, enabling survival under otherwise lethal conditions. Such behaviours evolve under selective pressure and allow insects to detect, avoid or minimize contact with toxicants, for example, by ceasing feeding, moving to untreated leaf surfaces, shifting deeper into the crop canopy or dispersing from sprayed areas. In *P. xylostella* adults exhibit selective oviposition on safer sites while larvae actively migrate away from treated foliage, thereby limiting exposure to foliar insecticides. These avoidance behaviours contribute significantly to the diamondback moth's resistance to multiple insecticide

classes, including organochlorines, organophosphates, carbamates and synthetic pyrethroids (Shehzad *et al.*, 2023) [49].

Physiological mechanism

Refers to any internal change like reduce penetration, enhance detoxification and change target site shape by mutation is called physiological mechanism. It include three mechanisms, viz., penetration resistance, metabolic detoxification and altered target-site mutation (Siddiqui *et al.*, 2023) [50].

Penetration resistance



Source: https://encrypted-tbn0.gstatic.com/images?q=tbn:ANd9GcSa5fPKXYkkBnxIigW-g_l0TlVjMjeI_iJ_1Z-ZZK_HGwHddg

Fig 2: Systemic diagram of penetration resistance mechanism

Penetration resistance refers to the ability of insect cuticles to prevent insecticides from penetrating, thereby reducing their effectiveness (Lin *et al.*, 2012) [27]. For an insecticide to reach its target site, it must penetrate multiple cuticular layers, the epicuticle, exocuticle, endocuticle and epidermis before entering the haemolymph. In resistant insects, alterations in cuticle composition, increased cuticle thickness or reduced permeability slow insecticide

penetration resulting in lower concentrations reaching the target site (Balabanidou *et al.*, 2018) [5]. This reduced uptake allows detoxification enzymes to inactivate the insecticide efficiently, enabling the insect to survive and reproduce without toxic effects (Siddiqui *et al.*, 2023) [50]. Although cuticular resistance in *P. xylostella* is less studied, recent findings indicate distinct differences in cuticle-associated microRNAs between resistant and susceptible populations.

MicroRNAs (miRNAs), small non-coding RNAs that regulate gene expression, can influence cuticle formation and modification, thereby contributing to the development of insecticide resistance (Shabbir *et al.*, 2021) [48].

Metabolic detoxification

Metabolic resistance is a major defence mechanism in insects, driven by enzymatic detoxification systems that neutralize insecticide molecules (Panini *et al.*, 2016; Hilliou *et al.*, 2021) [39, 14]. Three key enzyme families are primarily responsible for resistance development: Cytochrome P450 monooxygenases (P450s), Glutathione S-transferases (GSTs) and Carboxyl/cholinesterases (CCEs) (Panicker, 2025) [38]. Metabolic detoxification occurs in two phases.

Phase I involves oxidation or hydrolysis of insecticides mainly mediated by P450s and esterases, producing reactive intermediates that become more water soluble (Zhou *et al.*, 2019) [63]. Phase II consists of conjugation reactions catalyzed by GSTs, converting Phase I metabolites into highly water soluble, less toxic compounds that are readily excreted (Mak and Denisov, 2018) [31]. Through this mechanism, diamondback moth develops resistance to several insecticide groups, including carbamates, organophosphates and pyrethroids. Overall, metabolic detoxification relies on a diverse suite of enzymes capable of detoxifying multiple insecticide classes (Shehzad *et al.*, 2023) [49].

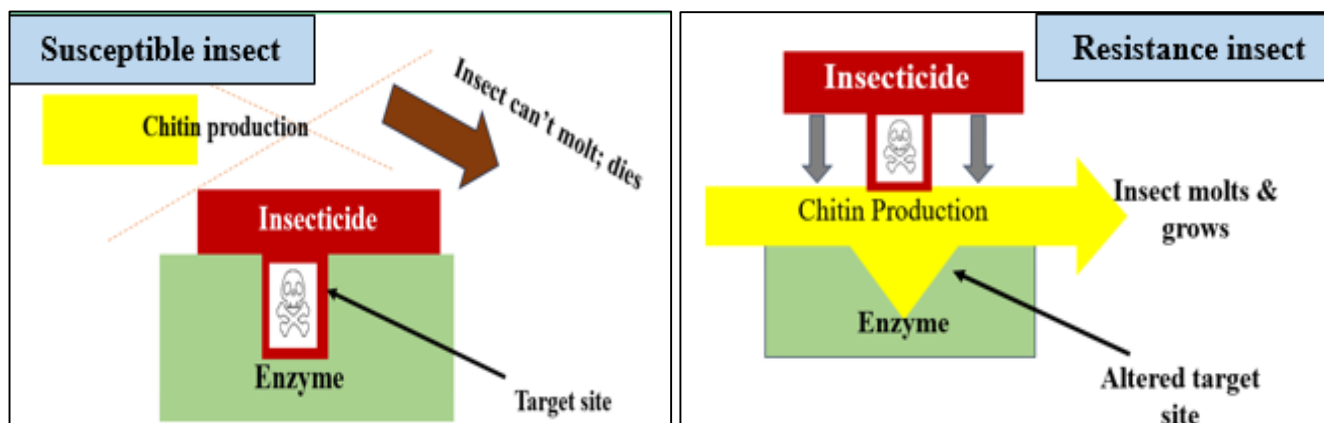
Table 1: Major detoxification enzymes and the insecticide classes against which they confer metabolic resistance in insects.

Detoxify enzymes	Classes
Carboxylesterases	Carbamates, organophosphates, synthetic pyrethroids
Glutathione-S-transferase	Diamides, organophosphates, synthetic pyrethroids
Cytochrome P450	Carbamates, organophosphates, synthetic pyrethroids

Altered target-site mutation

Target-site insensitivity is one of the most important and second most common mechanism (primary mechanism) of resistance (Liang *et al.*, 2025; Karaagac, 2012) [25, 21]. In this mechanism, the site where the toxin normally binds becomes modified, reducing the effectiveness of the insecticide (Charaabi *et al.*, 2018; Anonymous, 2025b) [8, 2]. Such modifications usually occur due to small genetic changes in insects, for example, a single nucleotide mutation

in the DNA. These mutations alter the amino acid sequence of the target protein, changing its binding structure. As a result, the insecticide cannot attach properly to its target site, allowing the insect to survive the treatment. Several classes of insecticides, including diamides, organophosphates, organochlorines and synthetic pyrethroids, have developed resistance in *P. xylostella* through this target-site mechanism (Siddiqui *et al.*, 2023) [50].



Source: <https://pesticidestewardship.org/resistance/insecticide-resistance/insecticide-resistance-mechanisms/>

Fig 3: Systemic diagram of altered target-site mutation mechanisms in susceptible and resistance insect

In susceptible insects, insecticides effectively bind to their target sites, inhibiting chitin synthesis and disrupting the molting process. This leads to paralysis and eventual death, as the target site remains fully sensitive to the insecticide. This interaction follows a classic lock and key model where the insecticide (key) fits precisely into the target protein (lock). In resistant insects, mutations in the target site alter its structure, reducing or preventing insecticide binding. As a result, chitin synthesis continues normally, allowing the insect to grow, molt and reproduce despite insecticide exposure. The lock and key mechanism is compromised because the “lock” is modified and the insecticide no longer fits effectively. Such target-site alterations represent a major mechanism of resistance and can confer cross-resistance to multiple classes of insecticides.

Table 2: Major target-site alterations in resistant insects and the insecticide classes affected by these resistance mechanisms.

Target sites	Resistance to
Altered Acetylcholinesterase (AChE)	Organophosphates and carbamates
Ryanodine receptor	Diamides
Reduced GABA receptor sensitivity	Organochlorines

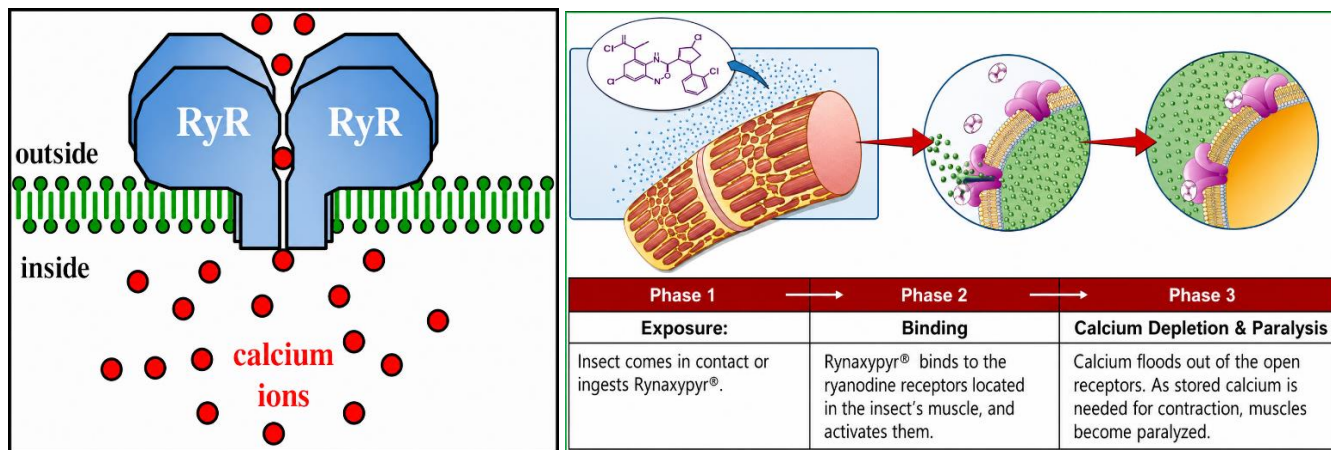
Altered Acetylcholinesterase (AChE)

Acetylcholinesterase (AChE) is essential for normal nerve impulse transmission in insects. Under susceptible conditions, AChE rapidly hydrolyses acetylcholine (ACh) into choline and acetate at the synaptic cleft. This timely breakdown prevents continuous stimulation of the postsynaptic neuron and ensures smooth transmission of

nerve impulses from one neuron to the next. When an AChE-inhibiting insecticide reaches the synapse in susceptible insects, it binds to the AChE enzyme and blocks the breakdown of ACh. The accumulated acetylcholine overstimulates postsynaptic receptors, leading to continuous nerve firing, paralysis and eventually death. In resistant insects mutations in the AChE gene alter the shape of the enzyme active site. Because of this structural modification, the insecticide cannot bind effectively to the target site.

AChE continues to function normally and breaks down acetylcholine into choline and acetate allowing proper nerve impulse transmission. As the insecticide fails to inhibit AChE, the insect exhibits no toxic effects and can survive grow and reproduce even under insecticide exposure (Ranganathan *et al.*, 2022) [43].

Ryanodine receptor



Source: <https://irac-online.org/documents/diamide-moa-rotation/>

Fig 4: Mechanism of Diamide insecticide action and RyR-mediated resistance development

Diamide insecticides (IRAC Group 28) act specifically on ryanodine receptors (RyRs) which are large calcium release channels located on the endoplasmic and sarcoplasmic reticulum of insect muscle cells and play an important role in muscle contraction (Truong and Pessah, 2019) [57]. In susceptible insects, diamides bind tightly to RyRs, blocking the channels in an abnormally open state. This triggers continuous calcium efflux into the cytoplasm, causing sustained muscle contraction, depletion of calcium stores, paralysis and eventually death. In resistant insects, point

mutations in the RyR gene modify the insecticide binding site, reducing or preventing diamide binding. Consequently, the RyR channels do not remain abnormally open, calcium homeostasis is maintained and muscle function remains normal. Because the toxic mode of action is effectively neutralized, resistant insects continue to feed, develop and reproduce without adverse effects from diamide exposure (Jeanguenat, 2013) [19].

Reduced GABA receptor sensitivity

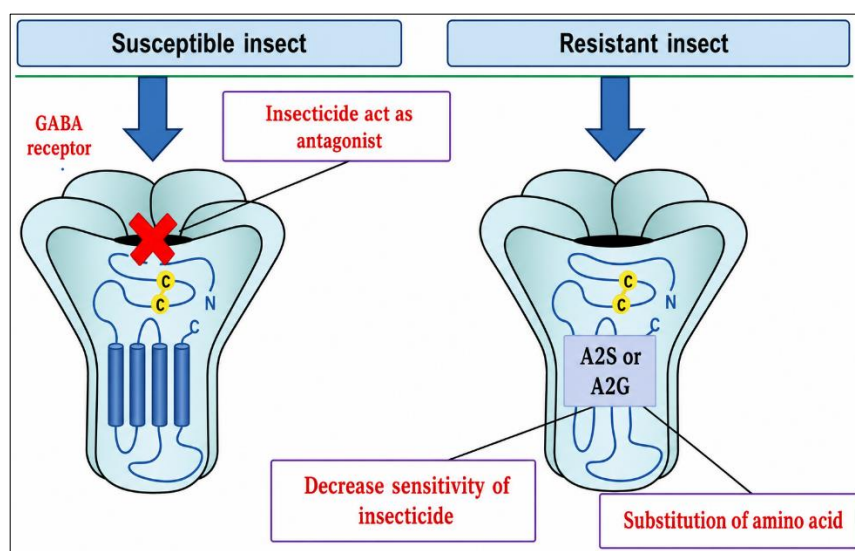


Fig 5: Target site resistance in GABA receptors due to amino acid substitution

Reduced GABA receptor sensitivity arises from mutations or structural modifications in the GABA (γ -aminobutyric acid) receptor, a chloride ion (Cl^-) channel located on insect nerve membranes that normally mediates hyperpolarization and prevents excessive nerve firing. In susceptible insects,

GABA-targeting insecticides bind to the receptor as antagonists, blocking chloride influx. This prevents hyperpolarization and triggers uncontrolled nerve excitation leading to convulsions, paralysis and death. In resistant insects point mutations in the GABA receptor gene most

notably the A2S (Alanine → Serine) and A2G (Alanine → Glycine) substitutions alter the insecticide-binding site, reducing binding affinity. Consequently, chloride ion flow through the GABA channel remains unaffected, normal hyperpolarization is maintained and nerve signaling proceeds normally. These structural changes enable resistant insects to survive insecticide exposure.

Methods for detection of insecticidal resistance

Resistance is determined by using different detection methods like conventional standard bioassay and biochemical, immunological and molecular methods. Among them, conventional standard bioassay and biochemical detection methods are widely used.

Conventional method

Conventional method published by International Resistance Action Committee (IRAC), in standard detection method of resistance insect collect from the field and rear them up to next generation. Larvae or adults are then exposed to different doses of an insecticide and their mortality is recorded. The LD₅₀ or LC₅₀ values for both susceptible and field populations are calculated using probit analysis (Karaagac, 2012) ^[21]. Total five steps are involved in conventional method.

- **Step 1:** The process begins with the collection of insects from the field and ensuring all collected populations are homogeneous in nature.
- **Step 2:** Then collected population are reared in controlled condition with their favourable condition with good quality of food resources up to 3-5 generations and ensures all reared population on same stage of development and healthy.
- **Step 3:** Once the next generation is ready, both resistant and the susceptible insect populations are exposed to various concentrations of the insecticide. This is done in a controlled condition and observes how the insects react to different doses. This step helps establish a dose-response curve for each population.
- **Step 4:** After the exposure of insecticide, calculate the lethal dose (LD₅₀) or lethal concentration (LC₅₀) for each population by the use of probit analysis.
- **Step 5:** After calculation of the LD₅₀/LC₅₀ values, then calculate resistance Ratio (RR)
(RR= LC₅₀ value of resistance population /LC₅₀ value of susceptible) and compare each other. A high RR indicates that the insects have developed resistance. For example, if the LD₅₀ of the resistant population is 100 times higher than the susceptible population, the RR is 100, which confirms a high level of resistance. This comparison helps determine the level of resistance and is the final step in the process.

Wang *et al.* (2012) concluded that diamondback moth populations from Northern China showed low to moderate resistance (RR=1.7 to 5.4) among them population of Wuhan, Hubei show highest resistance ration is 5.4 fold. In contrast, population of southern china showed very high resistance, especially population of Zengcheng, Guangdong showed over 2000 fold resistance ratio. Meghana *et al.* (2018) ^[33] reported that Dichlorvos 76 EC showed the highest resistance ratio against *P. xylostella* with a resistance level of 4.99 fold and LC₅₀ value is 15.63 mg/L.

Biochemical method

Biochemical assays can be used to measure the activity of detoxification enzymes (Esterase, Glutathione-S-transferase (GST) and CYP450) enzymes that help to break down insecticides, and also identify which mechanism is involved in insecticide resistance. Once a population is well characterized, these assays can also be applied to monitor changes in the frequency of resistance related genes in field populations exposed to different levels of selection pressure (Karaagac, 2012) ^[21].

- **Step 1:** The biochemical assay begins with the collection of insects from the field, ensuring that collected population is homogeneous in nature. The collected insects are then individually or collectively homogenized in a phosphate or potassium phosphate buffer which facilitates the extraction of detoxification enzymes required for biochemical detection of insecticide resistance.
- **Step 2:** After enzyme extraction, the enzyme solution is mixed with specific reactants depending on the type of enzyme being tested. Each enzyme requires a different substrate for example, alpha naphthol (α -N-OH), for esterases, 1-chloro-2, 4-dinitrobenzene (CDNB) for GSTs and ethoxycoumarin-O-deethylase (ECOD) for cytochrome P450 (Eziah *et al.*, 2009) ^[11]. The mixture is incubated at 25-30 °C for 15-30 minutes to allow the enzymatic reaction to occur and produce measurable colour or fluorescence.
- **Step 3:** During the biochemical detection assay each enzyme produces a characteristic colour after incubation with its specific substrate. Esterases produce blue colour, GST (Glutathione-S-transferase) produces yellow colour and cytochrome P450 produce red to pink colour. Intensity of the developed colour directly indicates the level of enzyme activity in the insect sample darker and stronger colour reflects higher enzymatic activity.
- **Step 4:** After colour development, the intensity of the colour is measured by using a spectrophotometer or fluorometer. Higher absorbance (or fluorescence) values indicate higher enzyme activity reflecting a stronger metabolic detoxification capacity in the insect population.
- **Step 5:** After measuring the color intensity, the enzyme activity level is calculated, which is then correlated with the presence and degree of insecticide resistance in the tested insect population. Enzyme activity levels are compared between susceptible and field populations. Higher activity of enzymes indicates the insects may have developed metabolic resistance to insecticides.

Eziah *et al.* (2009) ^[11] evaluated larval populations of *P. xylostella* highest ECOD activity was observed in the WiC strain (30.72; 5.75-fold higher than susceptible HaC strain), highest α -N-OH esterase activity occurred in the WiC strain (611.20; 1.88 fold increase) and highest GST CDBN conjugation activity was recorded in the YaC strain (448.11; 1.61-fold over than susceptible HaC strain) These results indicate that the WiC and YaC strains show strongest elevation in P450, esterase and GST enzymes respectively.

Insecticide Resistance Management (IRM)

Insecticide Resistance Management (IRM) involves

designing and implementing pest control strategies that minimize selection pressure on insect populations, thereby slowing the development of resistance and preserving the long-term effectiveness of insecticides (Sarangi *et al.*, 2025) [46]. Effective IRM includes rotation of insecticides with different modes of action, integration of non-chemical methods and use of synergists. Insecticides should be applied only when pest populations reach or exceed the threshold, ensuring that unnecessary sprays are avoided. This approach reduces excessive selection pressure, slows the development of resistance and supports sustainable resistance management.

Window approach

The window approach is a simple resistance management strategy in which the entire crop period is divided into different “windows” such as the transplanting stage (W1), cupping stage (W2), head-formation stage (W3) and harvesting stage (W4) and each window is assigned a different class of insecticide with a different mode of action. By spraying only one insecticide group within a window and use other group in next window continuous rotation is achieved, reducing selection pressure on any single chemistry. The insect generations (*e.g.*, DBM Gen-1, Gen-2 and Gen-3) occur during different crop windows and by changing the insecticide group in each window every new generation is exposed to a different mode of action. This prevents the same population from getting the same insecticide again and again, which helps slow down resistance development (Chaudhari, 2020) [9].

Chaudhari (2020) [9] concluded that resistance development in *P. xylostella* to chlorantraniliprole was delayed after the use of different groups of insecticides in sequential windows. In lab CH-resistance population resistance is reduced in treatment 1 (1132.15 to 99.56), in Sabarkantha CH-Resistant population also resistance ratio is reduced in treatment 1 from 212.19 to 13.14 and in Rajkot CH-Resistant again resistance is decrease from 169.64 to 6.90. This indicates that the sequential use of different classes of insecticides effectively delayed and reduced resistance in *P. xylostella*.

Use of synergists

Synergists have been used commercially for about 50 years and played a significant role in improving insecticide performance (Bernard and Philogene, 1993) [7]. Synergists is nontoxic chemicals added to insecticides to enhance their effectiveness against insect-pests and also help to insecticide work normally by blocking detoxification enzymes; They do not actually increase the inherent potency of the insecticides. By inhibiting detoxification enzymes synergists can make resistant populations susceptible again and help prevent or delay the development of insecticide resistance.

Table 2: Different synthetic and botanical synergists

Synthetic synergists	Botanical synergists (Thomas, 2018) [56]	
PBO (Piperonyl Butoxide)	Sesame oil	Jatropha oil
DEM (DiEthyl Maleate)	Neem oil	Sunflower oil
TPP (TriPhenyl Phosphate)	Sunflower oil	Lemongrass oil
DEF (S,S,S-Tributyl phosphorotrithioate)	Castor oil	Pongamia oil

Liu *et al.* (2015) [29] concluded that the use of synergists

DEF, DEM and PBO significantly reduced the LC₅₀ values of cyantraniliprole in all three strains indicating their ability to enhance insecticide toxicity. The strongest synergistic effect was observed with DEM in both the Cya-SEL strain (SR=3.2) and UNSEL strain (SR=2.1) followed by PBO and DEF. In LAB strain PBO produced highest synergism (SR=1.5). Overall, data suggested that metabolic detoxification enzymes particularly esterases and cytochrome P450s play a major role in cyantraniliprole resistance and synergists can partially restore susceptibility. Another study conducted by Chaudhari (2020) [9] on the effect of sesame oil on cyantraniliprole resistance in *P. xylostella*, reported that after the use sesame oil with cyantraniliprole, reduced the LC₅₀ value and increased the synergistic ratio in all tested populations, indicating that the synergist enhanced the effectiveness of insecticide and delayed resistance.

Insecticidal mixture

Insecticide mixtures are considered an essential strategy for managing resistance in many insect pests (South and Hastings, 2018) [51]. Insecticide mixtures increase the level of target pest control and broadening the range of pests controlled; however, they may offer benefits for IRM when appropriately incorporated into rotation strategies with additional mode(s) of action, but generally a single mixture should not be relied upon alone (IRAC, 2012) [17]. Shabbir *et al.* (2021) [48] The results show that the FOH-DBM strain was the most susceptible baseline for both chlorantraniliprole and *Bt*, while the other three strains (*Bt*-DBM, CL-DBM and *Bt*C-DBM) exhibited high levels of resistance, particularly to chlorantraniliprole, where CL-DBM showed the highest resistance ratio (31.2) followed by *Bt*C-DBM (27.3) and *Bt*-DBM (24.2). For *Bt* alone, *Bt*-DBM displayed the greatest resistance (RR=18.7), whereas CL-DBM and *Bt*C-DBM showed only low resistance (RR=1.8 and 1.3). Importantly, when *Bt* was combined with chlorantraniliprole, LC₅₀ values and resistance ratios were markedly reduced across all resistant strains, indicating additive effect of mixture *Bt* + chlorantraniliprole is more effective in managing resistant *P. xylostella* populations than either insecticide alone.

Genome editing

Insecticides have traditionally been the primary tool for managing *P. xylostella* populations, but the species has developed resistance to nearly all major insecticide groups, making effective control increasingly difficult in agricultural environments. This growing resistance highlights the need for novel and efficient management approaches particularly genetic-based control methods. These methods involve the genetic modification of target insects followed by their reintroduction into natural populations (McFarlane *et al.* 2018). These strategies include RNA interference (RNAi) for targeted gene suppression, transgenic crops engineered to produce *B. thuringiensis* (*Bt*) toxins and CRISPR/Cas9 technology for precise genome modification all of which provide sustainable alternatives to conventional chemical pesticides. Among these, the CRISPR/Cas9 system shows exceptional potential for advancing pest management, enabling the development of gene-drive systems for population reduction and (gSIT) genetic sterile insect techniques (Asad *et al.*, 2025).

Bautista *et al.* (2009) demonstrated that across both

experiments, CYP6BG1 dsRNA-fed larvae (es + dsRNA) showed lower LC₅₀ values compared to controls at both 24 h and 48 h a significant enhancement of permethrin toxicity due to gene silencing. In experiment 1, LC₅₀ values in dsRNA-fed larvae reduce from 1722 ppm to 662 ppm at 24 h and 1034 ppm to 824 ppm at 48 h reducing the resistance ratio from 52 to 20 fold and 67 to 32 fold, respectively. Similarly, in experiment 2, dsRNA-fed larvae demonstrated substantial declines in LC₅₀ from 1601 ppm to 493 ppm at 24 h and 1068 ppm to 499 ppm at 48 h resulting in over a two fold decrease in resistance. Overall data clearly show that CYP6BG1 knockdown significantly increases permethrin susceptibility.

Conclusion

Diamondback moth (*Plutella xylostella* L.) is recognized as the most resistant globally, with documented resistance to 104 chemical compounds. Resistance in *P. xylostella* arises through both behavioural and physiological mechanisms including reduced insecticide penetration, enhanced metabolic detoxification by enzymes and target-site modifications. Detection of these resistance traits is accomplished through conventional bioassays and biochemical methods which help to identify resistance levels and underlying mechanisms. To mitigate resistance several strategies have been employed including the window strategy of rotating insecticides with different modes of action, use of synergists (PBO, DEM, TPP), mixtures of insecticides and advanced molecular approaches like RNA interference (RNAi) have shown promise in targeting resistance related genes (e.g., CYP6BG1).

Future prospects

Future research should prioritize the development of rapid, high-throughput assays for detecting resistance across multiple insecticide classes in diverse DBM populations, alongside detailed investigations into behavioral mechanisms that facilitate avoidance and reduced exposure to insecticides. Additionally, expanding genome-editing targets and enhancing the delivery efficiency of advanced molecular tools such as RNAi and CRISPR/Cas9 will be crucial for designing next-generation strategies to manage resistance in *P. xylostella*.

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