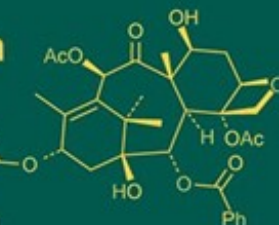
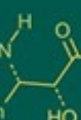
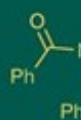
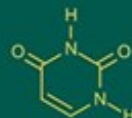
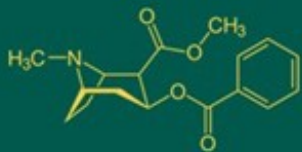


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New classes of insecticides, modes of action, comparative performance, and the shift towards genetic tools

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ABSTRACT

Insecticide discovery over the last quarter-century (2000–2026) has largely relied on structural variations around classical receptor targets. However, rapid field evolved resistance and tightening regulatory hurdles have pushed agrochemical research into a critical transition phase. This review critically evaluates two parallel paradigms: advanced synthetic chemistries (diamides, sulfoximines, mesoionics, *meta*-diamides, and chordotonal-organ disruptors) and emerging genetic platforms (sprayable dsRNA and CRISPR-Cas9 gene drives). Rather than offering a standard historical overview, this paper identifies critical research gaps at the intersection of target-site saturation, non-target site resistance (NTSR), and bio-delivery limitations. We highlight how structural cryo-EM insights and CRISPR-guided resistance forecasting can be leveraged to design next-generation chemistry, while pinpointing the exact biochemical and regulatory bottlenecks preventing genetic pest management from replacing small molecules. Ultimately, this synthesis provides a strategic roadmap for researchers seeking high-impact research topics in molecular entomology, chemical biology, and resistance forecasting.

Keywords: Insecticide discovery, Resistance forecasting, Sprayable dsRNA, CRISPR-Cas9 gene drives, Molecular entomology

1. Introduction

By the late 1990s, the agrochemical sector faced a double crisis: widespread resistance to legacy organophosphates, carbamates, and synthetic pyrethroids, paired with regulatory phase-outs driven by mammalian and environmental toxicity. Industry responded by targeting structural differences in insect ion channels and receptors, giving rise to modern IRAC-classified chemistries (Deep *et al.*, 2018) [1]. Concurrently, recent breakthroughs in sprayable RNA interference (RNAi) and CRISPR-Cas9 population suppression have introduced a fundamental shift from chemical poisoning to sequence-specific genetic control (Rodrigues, 2021; Zhao *et al.*, 2024) [2, 3].

Research Gap: Despite rapid literature growth across these disparate domains, current reviews remain fragmented, treating small-molecule pharmacology and genetic pest control as isolated disciplines. Furthermore, existing literature heavily emphasizes target-site point mutations while systematically under-reporting metabolic resistance mechanisms, cuticular penetration dynamics, and the commercial delivery bottlenecks of nucleic-acid biopesticides. A holistic synthesis that directly compares the longevity, evolutionary constraints, and field-translation hurdles of synthetic chemistry versus genetic tools is missing (Bhuvaneswari *et al.*, 2022) [4].

The primary objective of this review is to bridge this gap by evaluating the comparative performance and evolutionary vulnerabilities of 2000–2026 pest management technologies. By identifying where target-site discovery is hitting diminishing returns and where genetic platforms face physical delivery barriers, this paper serves as an actionable guide for researchers seeking unresolved, high-impact research topics in insecticide discovery, structural target site validation, and resistance forecasting (Sparks *et al.*, 2015) [5].

2. Nicotinic acetylcholine receptor insecticides after imidacloprid

The insect nicotinic acetylcholine receptor (nAChR) has been the single most productive insecticide target of the last thirty years, but not every compound that acts on it works the same way, and that distinction matters more than it might first appear (Simon-Delso, 2015; Xu, 2020) [6, 7].

2.1 Neonicotinoids - the class that started it, and its resistance debt

While first-generation neonicotinoids established nAChR as a primary target, their widespread adoption triggered severe target-site (R_{81T}) and P450-mediated metabolic resistance across hemipteran pests, necessitating chemistry that avoids these specific detoxification pathways (Matsuda *et al.*, 2020) [8].

Target-site mutations (notably R_{81T} in the nAChR $\beta 1$ subunit) combined with elevated cytochrome P450 activity have produced well-documented resistance in *Bemisia tabaci* (Gennadius), *Myzus persicae* (Sulzer), and *Nilaparvata lugens* (Stål), and cross-resistance between imidacloprid, thiamethoxam, and clothianidin is now routine in several of these species (Bass *et al.*, 2015; Nauen and Denholm, 2005; Zhang *et al.*, 2021) [9, 10, 11]. Regulatory restriction of several neonicotinoids in the EU, driven by pollinator concerns, has added a second pressure pushing the industry toward new nAChR chemistry that avoids the same detoxification pathways (Sahani *et al.*, 2021) [12].

2.2 Spinosyns - a different door on the same receptor

Spinosad, isolated from the soil actinomycete *Saccharopolyspora spinosa* Mertz and Yao, and commercialized in 1997, was the first insecticide shown not to compete with any previously known binding site - a series of receptor-binding assays found no interaction with the nicotinic, GABAergic, or other classical target sites tested (Orr *et al.*, 2009) [13]. It took another decade of receptor work to establish that spinosyns act allosterically on the $D_{\alpha 6}$ nAChR subunit, a site distinct from where neonicotinoids bind (Geng *et al.*, 2013; Salgado and Sparks, 2005) [14, 15]. Spinetoram, a semi-synthetic follow-up approved in the mid 2000s, extended the spectrum and residual activity of the class (Kirst, 2016) [16]. A 2025 twenty-five-year retrospective by Corteva researchers is a useful marker of how much has been learned since, resistance is now confirmed in *Plutella xylostella* (Linnaeus, 1758) and several thrips species, almost always through mutations at the $D_{\alpha 6}$ locus, but cross-resistance to neonicotinoids or diamides has not been observed, which keeps spinosyns genuinely useful in rotation programs (Crouse *et al.*, 2025; Sparks *et al.*, 2021) [17, 18].

2.3 Sulfoximines - built to dodge neonicotinoid resistance

Sulfoxaflor, introduced around 2013, was explicitly designed to retain nAChR activity while avoiding the metabolic and target-site mechanisms that were already compromising neonicotinoids (Sparks *et al.*, 2013) [19]. Structurally the sulfoximine group sits differently in the receptor pocket than the nitroguanidine or nitromethylene groups of neonicotinoids, and this has translated into limited cross-resistance in most field populations tested so far, although a handful of studies have found P450-mediated cross-resistance in already highly resistant *N. lugens* strains (Long *et al.*, 2021; Sparks *et al.*, 2019) [20, 21]. In practice sulfoxaflor is treated as a genuinely separate IRAC subgroup (4C) rather than a neonicotinoid variant, and that distinction has held up reasonably well under field use.

2.4 Butenolides - one new class, one commercial compound

Flupyradifurone, launched in 2014, is so far the only commercialised butenolide. Its discovery is a nice example of natural-product-inspired design, the butenolide ring was borrowed from stemofoline, a naturally occurring nAChR ligand, and grafted onto a synthetic scaffold optimised for field stability (Nauen *et al.*, 2015) [22]. Flupyradifurone acts as a partial agonist at the nAChR and, critically, is not metabolised by CYP_{6CM1} , the P450 enzyme responsible for a large share of neonicotinoid resistance in *B. tabaci*, which is why it remains active against neonicotinoid-resistant whitefly populations in several published trials (Jeschke *et al.*, 2015) [23].

2.5 Mesoionics - inhibiting the receptor instead of activating it

Triflumezopyrim, the first commercial mesoionic insecticide, works differently from every other nAChR insecticide discussed so far, instead of activating the receptor, it binds the orthosteric site and blocks it, acting as a true antagonist rather than an agonist (Cordova *et al.*, 2016) [24]. That was initially seen as a liability antagonism at this site was assumed to give poor field control - but triflumezopyrim turned out to be highly effective against planthoppers, including *N. lugens* populations already resistant to neonicotinoids (Holyoke *et al.*, 2017) [25]. A second mesoionic compound, fenmezoditiaz, was characterised

more recently and extends the class to lepidopteran pests as well (Huang *et al.*, 2025; Maiefisch *et al.*, 2025) [26, 27].

Open research opportunities in nAChR modulation

- **Allosteric Mining:** Investigating non-orthosteric binding pockets on $D_{\alpha 6}$ and orphan nAChR subunits using computational ligand docking to discover novel spinosyn mimics.
- **Antagonist Synergy:** Evaluating whether orthosteric antagonists like triflumezopyrim can suppress field populations exhibiting R_{81T} target-site resistance without inducing rapid target-site cross-selection

3. Diamides - the other major success story of the 2000s

If neonicotinoids defined insecticide discovery in the 1990s, diamides defined it in the 2000s. Flubendiamide and chlorantraniliprole, both commercialised around 2007-2008, were the first insecticides shown to activate the insect ryanodine receptor (RyR), the channel responsible for releasing calcium from the sarcoplasmic reticulum during muscle contraction (Du *et al.*, 2023; Du *et al.*, 2023) [28, 29]. Sustained RyR activation empties intracellular calcium stores, and insects essentially exhaust themselves into paralysis and death. Because the insect RyR differs enough structurally from the mammalian isoforms, diamides show a wide margin of safety in vertebrates while remaining extremely potent against lepidopteran and some coleopteran pests. Cryo-EM structures showed, at near-atomic resolution, that diamides bind within the transmembrane domain of the RyR channel, stabilized by hydrophobic interactions specific to insect isoforms, explaining why selectivity for the insect channel is so high (Lin *et al.*, 2024) [30]. The critical I_{4790M} and G_{4946E} field mutations disrupt these binding pockets through steric hindrance without compromising essential calcium transport. This structural landmark opens an active research avenue: designing sterically adapted, second-generation RyR modulators or pseudo-diamides capable of accommodating mutated residues while maintaining insect selectivity.

That structural clarity has not stopped resistance from spreading, however. Beyond target-site mutations, metabolic resistance driven by the upregulation of cytochrome P450 monooxygenases (such as CYP_{6AE} family members) and carboxylesterases as well as cuticular penetration resistance, play significant roles in field-evolved resistance across *Spodoptera* and *Helicoverpa* species. Point mutations at RyR positions I_{4790M} and G_{4946E} , (numbering varies slightly between species) are now widespread in diamondback moth, *Spodoptera* species, and *Tuta absoluta* (Meyrick, 1917) populations across Asia and South America, and resistance ratios above 100-fold have been reported for chlorantraniliprole in some rice leaf-folder populations (Richardson *et al.*, 2025) [31]. Second-generation diamides such as cyantraniliprole and tetraniliprole were developed partly to retain activity against these resistant strains, with mixed but generally encouraging results (Roditakis *et al.*, 2026) [32].

4. GABA-gated chloride channel modulators - from fipronil to the newest isoxazolines

The more interesting development is that two newer classes - meta-diamides and isoxazolines - also act on the RDL GABA receptor but bind at a site that is structurally distinct from where fipronil and the older cyclodienes bind, so there is no cross-resistance between them (Nakao and Banba, 2016; Zhao *et al.*, 2014) [33, 34]. Broflanilide was the first commercial meta-diamide (2019). Isoxazolines were first commercialised for veterinary use - fluralaner, launched in 2016 for flea and tick control in companion animals - before isocycloseram brought the chemotype into crop protection around 2022; broflanilide and the isoxazolines are grouped together as IRAC Group 30 for exactly this reason (Lümmen *et al.*, 2022; Asahi *et al.*, 2023) [36, 35]. Both compounds cause hyperexcitation and convulsions rather than the flaccid paralysis typical of GABA antagonism at the classical site, which is itself a clue that the binding mode is different. Using

CRISPR/Cas9 knock-in, researchers have since pinned down the residue responsible - a glycine at the third position of the M_3 transmembrane domain (G_3) - as essential for both broflanilide and isocycloseram activity; substituting it with the mammalian-type methionine essentially abolishes sensitivity to both compounds in *Drosophila* (Asahi *et al.*, 2023; Chen *et al.*, 2024) [35, 37]. That is a genuinely useful piece of resistance forecasting, it tells researchers exactly which mutation to watch for in field populations before resistance becomes a practical problem, something that was never available for the older GABA-acting chemistries. Field trials of broflanilide formulated for indoor residual spraying have also shown strong efficacy against pyrethroid-resistant malaria vectors, which has pushed interest in this class well beyond crop protection (Snetselaar *et al.*, 2021) [38].

Open research opportunities in GABA receptors

- **CRISPR-Guided Resistance Mapping:** Applying CRISPR-Cas9 knock-in screens across non-model crop pests to proactively identify potential resistance mutations beyond G_3 in the M_3 transmembrane domain before Group 30 compounds hit widespread field failure.

5. Sodium channel blockers - indoxacarb and the oxadiazines

Indoxacarb, launched in 2000, was the first oxadiazine and remains the only major representative of Insecticide Resistance Action Committee (IRAC) Group 22A. Unlike pyrethroids, which keep sodium channels open, indoxacarb blocks them - but only after being bioactivated inside the insect to its more potent metabolite, DCJW (Lapied *et al.*, 2001; Wing *et al.*, 2000) [39, 40]. That bioactivation step is itself part of the selectivity, DCJW blocks insect sodium currents roughly 1000-fold more potently than the parent compound blocks the equivalent channel in mammalian neurons, and the *S*-enantiomer of DCJW is far more active than the *R*-enantiomer in both insects and in vitro assays (Silver *et al.*, 2003) [41]. Resistance mechanisms reported so far lean toward metabolic detoxification - upregulation of nuclear receptor HR_96 and its downstream detoxification genes has been documented in *Spodoptera litura* (Fabricius, 1775) - rather than target-site mutation, which is a somewhat different resistance

profile from most of the other classes covered here (Zhang *et al.*, 2016) [42].

6. Disrupting the chordotonal organ - a genuinely new target family

One of the more interesting developments of the last decade has nothing to do with synapses at all. The chordotonal organs - the mechanosensory structures insects use for hearing, balance, and proprioception - turned out to be druggable. Pymetrozine, developed in the 1990s, was the first insecticide recognised to act here, working through TRPV channels (Nanchung and Inactive) in these organs rather than through any neurotransmitter receptor (Ausborn *et al.*, 2022) [43]. Afidopyropen, a pyropene insecticide that reached the market around 2018, works the same way, over-stimulating and then silencing the TRPV complex, which shuts down feeding behaviour in aphids and whiteflies within hours (Horikoshi *et al.*, 2021; Ye *et al.*, 2021) [44, 45]. Because the target is structurally and functionally unrelated to nAChRs or GABA receptors, there is essentially no cross-resistance with any of the older chemistries.

The newest entrant here is dimpropyridaz, a pyridazine pyrazolecarboxamide marketed by BASF as Axalion or Efficon, which received its own Insecticide Resistance Action Committee (IRAC) classification (Group 36) in 2023 (BASF, 2023) [46]. Electrophysiological work published the same year showed that dimpropyridaz inhibits chordotonal organ function upstream of the TRPV channels themselves, meaning it acts even earlier in the signalling pathway than pymetrozine or afidopyropen do (Spalthoff *et al.*, 2023) [47]. It is still too early for field resistance data, but the mechanism gives it a reasonable case for being a genuinely distinct addition to the toolbox rather than a re-branded analogue. Furthermore, BASF has reported selectivity toward piercing-sucking pests with low acute toxicity to key beneficial pollinators such as *Apis mellifera* Linnaeus, 1758 in its own product data, though independent ecotoxicological studies confirming this selectivity have not yet been published (BASF, 2023) [46].

7. Comparative summary

Table 1. Comparative summary of major modern insecticide classes

Table 1. pulls the classes discussed above together for direct comparison.

Class	IRAC group	Target site	First introduced	Representative compound(s)	Known resistance issues
Neonicotinoids	4A	nAChR (orthosteric, agonist)	1991 (imidacloprid)	Imidacloprid, thiamethoxam, clothianidin	Widespread; target-site (R_{81T}) and metabolic (P450) resistance in aphids, whiteflies, planthoppers
Spinosyns	5	nAChR, allosteric Site I ($\alpha 6$ subunit)	1997 (spinosad); 2007 (spinetoram)	Spinosad, spinetoram	Target-site mutations in $D_{\alpha 6}$; field resistance in <i>Plutella</i> , <i>Frankliniella</i>
Sulfoximines	4C	nAChR (distinct binding mode)	2013 (sulfoxaflor)	Sulfoxaflor	Low cross-resistance to neonicotinoids so far; some P450-based cases reported
Butenolides	4D	nAChR (partial agonist)	2014 (flupyradifurone)	Flupyradifurone	Not metabolised by CYP_{6CM1} ; resistance rare but documented in whitefly
Mesoionics	4E	nAChR (orthosteric antagonist)	2015 (triflumezopyrim)	Triflumezopyrim, fenmezoctiaz	New class; resistance selection reported in planthoppers under lab pressure
Diamides	28	Ryanodine receptor (calcium release)	2007 (flubendiamide, chlorantraniliprole)	Chlorantraniliprole, cyantraniliprole, tetraniliprole	RyR point mutations (I_{4790M} , G_{4946E}) widespread in <i>Plutella</i> , <i>Spodoptera</i> , <i>Tuta</i>
Phenylpyrazoles	2B	GABA receptor (RDL), noncompetitive	1993 (fipronil)	Fipronil	A2'N/A2'S RDL mutations; largely phased out in EU agriculture, retained in vet use

(continued on next page)

Class	IRAC group	Target site	First introduced	Representative compound(s)	Known resistance issues
Meta-diamides	30	GABA receptor (RDL), distinct allosteric site	2019 (broflanilide)	Broflanilide	No cross-resistance with fipronil; lab-selected resistance and <i>G₃</i> TMD3 mutation identified
Isoxazolines	30	GABA receptor (RDL), same group as meta-diamides	2016 (fluralaner, vet); 2022 (isocycloseram, ag.)	Fluralaner, isocycloseram	Shares <i>G₃</i> TMD3 resistance mechanism with broflanilide
Oxadiazines	22A	Voltage-gated sodium channel (blocker)	2000 (indoxacarb)	Indoxacarb	Detoxification-gene upregulation (<i>HR₉₆</i>) reported in <i>Spodoptera litura</i>
Pyridine azomethines / pyropenes	9B / 9D	Chordotonal organ (TRPV channels)	2000 (pymetrozine); 2018 (afidopyropen)	Pymetrozine, pyrifluquinazon, afidopyropen	Limited field resistance reported to date
Pyridazine amides	36	Chordotonal organ, upstream of TRPV	2023 (dimpropyridaz)	Dimpropyridaz (Axalion/Efficon)	Too new for field resistance data

8. Beyond small molecules, RNA interference as an insecticide

Everything discussed so far works by binding a protein target. RNAi-based biopesticides work by silencing the gene that makes the target in the first place, using sequence-specific double-stranded RNA (dsRNA) delivered by spraying rather than by genetic engineering of the crop. The regulatory milestone that made this a real product category rather than a laboratory curiosity was the US EPA's December 2023 registration of Ledprona, a sprayable dsRNA biopesticide targeting the Colorado potato beetle (*Leptinotarsa decemlineata* (Say, 1824)) (Rodrigues *et al.*, 2021) [2]. Mechanistically these products rely on the same RNAi pathway first described in *Caenorhabditis elegans* (Maupas, 1900) in 1998, now repurposed for pest-specific gene knockdown (Guan *et al.*, 2023) [48].

The appeal is fairly obvious, sequence specificity means a dsRNA construct can, in principle, be designed to affect only the target pest and closely related species, sidestepping the broader non-target effects that have dogged conventional insecticides. The practical obstacles are just as real.

Physiological Degradation Barriers & Nano-Carrier Engineering:

The field translation of sprayable dsRNA is strictly limited by insect physiology. While Coleopterans (e.g., *L. decemlineata*) exhibit robust systemic RNAi uptake, Lepidopteran and Hemipteran species possess high luminal pH and abundant double-stranded ribonucleases (dsRNases) in their midguts, which rapidly degrade naked dsRNA before cellular internalization can occur (Yan *et al.*, 2021) [49]. Overcoming this bottleneck represents a prime target for formulation scientists. Emerging solutions include star-polymer complexes, chitosan nanoparticles, and lipid-based carriers engineered to withstand alkaline gut environments and facilitate endosomal escape (Yan *et al.*, 2021; Dong, 2024) [49, 50].

Delivery and stability of dsRNA in the field remain difficult many insect species degrade extracellular dsRNA quickly due to midgut endonucleases, and uptake efficiency varies enormously between taxa. Additionally, commercial adoption faces substantial economic hurdles, including high synthesis costs relative to traditional small molecules, formulation challenges for UV stability, and complex regulatory approval pathways for living modified organisms in the case of CRISPR gene drives (Luo *et al.*, 2024) [51]. Off-target silencing is also a live concern; the degree of RNAi effect correlates with the mismatch rate between the dsRNA sequence and non-target transcripts, so poorly designed constructs can, in theory, affect species well outside the intended target range (Guan *et al.*, 2023) [48]. Reviews focused on Lepidoptera have catalogued which target genes have worked well so far (chitin synthase, V-ATPase subunits, and various genes) and which pest species remain stubbornly resistant to RNAi despite gene knockdown being confirmed at the molecular level

(Sharma *et al.*, 2023) [52]. Resistance to RNAi itself has already been documented in Colorado potato beetle populations under lab selection, though those same resistant lines generally remain fully susceptible to conventional small-molecule insecticides, which supports using dsRNA and chemical insecticides together rather than as strict alternatives (Luo *et al.*, 2024) [51]. Public perception research suggests farmers and consumers are, so far, cautiously open to RNAi biopesticides, though acceptance depends heavily on how the technology is explained and regulated (Tardin-Coelho *et al.*, 2025; Chen *et al.*, 2024) [53, 37].

9. Genome editing, CRISPR-Cas9 and gene drives

CRISPR-Cas9 takes the genetic approach a step further, moving from silencing a gene transiently to editing it permanently, and in the case of gene drives, spreading that edit through an entire wild population over successive generations. A CRISPR gene drive works by copying itself into the matching chromosome during meiosis - so-called "homing" - which lets an edited allele spread through a population far faster than normal Mendelian inheritance would allow, in principle reaching fixation within a handful of generations even from a small release (Zhao *et al.*, 2024) [3]. Most of the early engineering work targeted mosquito disease vectors, but in 2023 saw the first successful demonstration in a genuine agricultural pest, a split homing drive targeting the *doublesex* gene in *Drosophila suzukii* (Matsumura, 1931) (spotted-wing drosophila), a soft-fruit pest with no effective biological control options in most of its invaded range (Yadav *et al.*, 2023) [54]. The drive disrupts a female-specific exon of *doublesex*, leaving males unaffected but rendering homozygous females sterile, which is intended to collapse the population over time rather than simply killing individual insects on contact.

Since then the pipeline has broadened considerably. A 2024 study engineered a gene drive and genetic sex-conversion system in the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann, 1824), one of the most economically damaging invasive pests worldwide (Meccariello *et al.*, 2024) [55]. Parallel work is underway in the diamondback moth, *P. xylostella*, though as of the most recent published attempts the drive components have not yet achieved efficient homing in that species, which is a useful reminder that gene drive engineering does not transfer cleanly from one insect to another even when the underlying CRISPR machinery is identical (Xu *et al.*, 2022) [56]. Toxin-antidote systems - which pair a recessive lethal cassette with a rescue transgene rather than relying purely on homing - have also been explored as a way of building drives that are self-limiting and easier to contain if something goes wrong, addressing one of the most persistent biosafety criticisms of the technology (Zhao *et al.*, 2024) [3]. Ethicists and policy researchers have been fairly blunt about the regulatory gap here, gene drive organisms cross national borders in ways existing biosafety frameworks were never built to handle, and

calls for internationally coordinated oversight have been a recurring theme in the literature since at least 2018 (Resnik *et al.*, 2024) [57]. The same core CRISPR toolkit is also being explored outside gene drives entirely - for basic functional genomics work identifying new insecticide targets, and even in weed management, where gene drive concepts are being tested in invasive plant populations using comparable logic (Kumam *et al.*, 2023; Arjunan and Thiruvengadam, 2026; Komal *et al.*, 2023) [59, 58, 60].

Open research opportunities in genetic pest control

- **Homing Efficiency Barriers:** Identifying the cellular DNA repair mechanisms (e.g., non-homologous end joining vs. homology-directed repair) that impede homing drive efficiency in agricultural pests like *P. xylostella* (Karami Nejad Ranjbar *et al.*, 2018) [61].
- **Resistance Modeling:** Developing multi-locus evolutionary models to predict how wild insect populations might evolve drive-resistant target site alleles under field-selection pressure (Champer *et al.*, 2016) [62].

10. Where botanicals and nanotechnology fit into this picture

It is worth being honest about where plant-derived and nanoformulated products actually stand relative to the synthetic classes above. Neem-based azadirachtin, pyrethrum, and various essential-oil formulations remain heavily researched - a systematic review covering roughly 2,500 papers published between 1945 and 2019 found the field has grown exponentially since the 1990s, still concentrated on a handful of plant families (Meliaceae most of all). But the same review points out a real weakness, most of this research stops at laboratory screening and rarely progresses to field-scale, commercially viable products, and non-target testing is done in barely one paper in eight (Saikumar *et al.*, 2025; Turchen *et al.*, 2020; Ullah *et al.*, 2025) [64, 65, 63]. Nanoformulation - encapsulating conventional or botanical actives in silica, polymer, or lipid carriers - is being pursued mainly to fix delivery problems (poor solubility, rapid degradation, off-target drift) rather than to introduce new modes of action, and safety data on nanoparticle carriers themselves is still catching up with the pace of formulation research (Dong *et al.*, 2024; Manna *et al.*, 2023) [50, 66]. Neither category currently rivals the synthetic classes discussed above in terms of field-scale reliability, but both are reasonable components of an integrated resistance-management programme rather than stand-alone replacements.

11. Discussion - what has actually changed since 2000

Looking across all of this, a few patterns stand out. First, genuinely new target sites are rare, and the industry knows it - IRAC has assigned only a handful of new mode-of-action groups since 2000 (diamides, sulfoximines, butenolides, mesoionics, *meta*-diamides/isoxazolines, and now the pyridazine amides), and each one took roughly a decade from initial discovery work to commercial launch (Maienfisch *et al.*, 2025; Sparks and Nauen, 2015; Jeschke, 2021) [27, 5, 67]. Second, resistance still tends to catch up eventually, but the newer classes are, on average, holding up longer than the organophosphates and carbamates did, partly because target sites are being chosen with more attention to structural divergence from mammalian homologues from the start, and partly because resistance monitoring itself has become far more molecular - point mutations are now identified and tracked years before they become a field-scale problem, which was simply not possible with 1990s-era tools (Richardson *et al.*, 2025; Asahi *et al.*, 2023) [31, 35]. Third, the two genetic technologies covered here - RNAi and CRISPR gene drives - are not competing with small-molecule insecticides in any direct sense. RNAi biopesticides are close to functioning like a very selective conventional insecticide, applied the same way, with the main open questions being delivery efficiency and cost (Rodrigues *et al.*, 2021; Luo *et al.*, 2024) [2, 51]. Gene drives are a completely different kind of

tool, aimed at population-level suppression over multiple generations rather than field-season control, and they come with a regulatory and ecological risk profile that has no real precedent in agrochemical history (Resnik *et al.*, 2024; Zhao *et al.*, 2024) [57, 3]. Treating them as if they belong on the same continuum as, say, sulfoxaflor versus imidacloprid probably understates how different the underlying logic is.

12. Conclusion

The period from 2000 to 2026 expanded the insecticidal toolbox with high-affinity chemistries targeting the ryanodine receptor, GABA receptor allosteric sites, and chordotonal TRPV complexes. Yet, field-evolved target-site mutations (e.g., RyR *I*_{4790M}, GABA *G*₃) and multi-gene metabolic detoxification demonstrate that new target sites alone cannot outpace pest evolution. While genetic tools like sprayable dsRNA and CRISPR gene drives offer unprecedented sequence specificity, they do not replace small molecules due to fundamental environmental stability, delivery, and biosafety constraints. To move the field forward, researchers looking for novel topics should focus on the following key gaps:

- **1. Target-Site Discovery & Allosteric Mining:** Cryo-EM structural biology must be applied to uncharacterized insect-specific membrane proteins and orphan G-protein coupled receptors (GPCRs) to discover genuinely non-classical binding pockets before target-site saturation occurs.
- **2. Non-Target Site Resistance (NTSR) Mechanisms:** More research is needed to unravel the transcriptional networks (e.g., *HR*₉₆, CCNC pathways) regulating cytochrome P450 and esterase cross-resistance across structurally unrelated modern chemistries (e.g., diamides vs. sulfoximines).
- **3. Bio-Delivery Engineering for RNAi:** Developing smart nano-carriers (e.g., pH-responsive polymer or lipid nanoparticles) capable of protecting dsRNA from gut endonucleases and UV degradation in stubborn lepidopteran and hemipteran targets.
- **4. Resistance Forecasting via Functional Genomics:** Utilizing CRISPR-Cas9 gene editing in model species to proactively construct functional resistance maps (*G*₃ analogues) for newly registered chemistries (e.g., IRAC Group 36 dimpropridaz) prior to widespread field failure.
- **5. Hybrid Resistance Management Models:** Developing mathematical and empirical frameworks to model co-selection risks when combining sprayable dsRNA with sub-lethal doses of small-molecule chemistries in integrated pest management (IPM) programs.

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