Review

CUADb-based oligonucleotide insecticides (contact unmodified antisense DNA biotechnology) vs. RNA biocontrols (double-stranded RNA technology): newly born fraternal twins in plant protection

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Abstract: Advances in molecular genetics, nucleic acid synthesis, and bioinformatics have created novel opportunities for plant protection. DNA and RNA now serve as active insecticidal ingredients, transcending their traditional role as carriers of genetic information. Critically, nucleic acids function as tools for pest control through two fundamentally distinct mechanisms: DNA containment (DNAc), employing oligonucleotide insecticides based on contact unmodified antisense DNA biotechnology (CUADb), and RNA interference (RNAi), employing RNA biocontrols based on double-stranded RNA (dsRNA) technology. It is essential to recognize that CUADb is not a variant of dsRNA technology; these are separate approaches with unique molecular actions. Early partial understanding of how antisense RNA and DNA oligonucleotides might function emerged in the 1960s–1970s. The role of antisense RNA within dsRNA fragments, central to RNAi, was discovered in the late 1990s as an antiviral immune response in nematodes. The RNAi approach then underwent extensive development for plant protection. In contrast, the CUADb approach originated independently, manifesting as effective oligonucleotide insecticides against lepidopterans in 2008. The subsequent discovery of its DNAc mechanism revealed its fundamental importance in disrupting rRNA biogenesis and ribosome production, a process entirely distinct from RNAi. As separate classes of next-generation insecticides, the end-products of these independent technologies offer long utility duration, low carbon footprint, high selectivity, and rapid environmental biodegradation. Currently, their efficacy profiles diverge: CUADb and DNAc demonstrate optimal results against Sternorrhyncha (e.g., aphids, mealybugs, psyllids, scale insects), while dsRNA technology and RNAi show the strongest insecticidal potential against beetles from the Tenebrionidae and Chrysomelidae families. Therefore, CUADb-based oligonucleotide insecticides represent a mature and distinct platform, not merely a supplementary or embryonic stage relative to RNAi. Highly likely, both technologies will prove effective against specific insect groups and could be deployed complementarily in formulations for broader pest control, leveraging their unique mechanisms of action.

Keywords: oligonucleotide insecticides; CUAD biotechnology; DNA containment; RNA biocontrols; double-stranded RNA technology; RNA interference; next-generation insecticides; plant protection

1. Introduction

The relentless innovation in insect pest control drives continuous replacement of older chemistries (carbamates, organophosphates) by contemporary agents (neonicotinoids, diamides) (van den Berg et al., 2021; Sparks et al., 2021; Zhang et al., 2023). Yet the intractable challenge of genetic resistance ensures this cycle persists (Sparks et al., 2021; Mogilicherla et al., 2023), demanding insecticides with extended utility, heightened selectivity, enhanced biodegradability, and reduced environmental footprint. Biomolecules represent the frontier for achieving these critical attributes (Christiaens et al., 2020; Oberemok et al., 2022; Palli, 2023; Gal'chinsky et al., 2024). A paradigm shift has emerged where nucleic acids (DNA and RNA) serve as active insecticidal ingredients, enabled by advances in molecular genetics, synthesis, and bioinformatics (Oberemok et al., 2018a; Rank and Koch, 2021; Oberemok et al., 2023; Koeppe et al., 2023). This has given rise to two distinct platforms: oligonucleotide insecticides based on Contact Unmodified Antisense DNA Biotechnology (CUADb) and RNA biocontrols utilizing double-stranded RNA (dsRNA) technology (Nitnavare et al., 2021; Gal'chinsky et al., 2024). Crucially, CUADb and dsRNA/RNAi are fundamentally distinct antisense technologies. Alongside CRISPR/Cas, they represent three core approaches for insect control, each employing unique molecular mechanisms: RNAi functions through guide RNA-mRNA duplexes cleaved by Argonaute nuclease; CUADb operates via guide DNA-rRNA duplexes processed by DNA-guided rRNase such as RNase H1 (DNAc mechanism); and CRISPR/Cas utilizes guide RNA-genomic DNA complexes with Cas nuclease (Kumar et al., 2025). It is essential to emphasize that CUADb constitutes an independent technological platform—not a variant of RNAi. While all three exhibit taxon-specific efficacy enabling combinatorial strategies, their divergent molecular targets preclude universal application, ensuring each will occupy defined niches.

After two decades of development, RNAi still lacks a standardized design algorithm for reliable insecticide development. Conversely, CUADb possesses a validated design framework effective against approximately 10-15% of insect pests (Oberemok et al., 2024a), demonstrating particular efficacy against Sternorrhyncha (e.g., aphids, mealybugs) and superorder Paraneoptera in general (Oberemok et al., 2025a), whereas RNA biocontrols show strongest activity against Coleoptera. This review advances the paradigm shift toward nucleic acid-based solutions, with dedicated focus on CUADb as a transformative technology characterized by its novel DNAc mechanism and advanced insecticidal profile. These independent "fraternal twins", CUADb and dsRNA, hold synergistic potential for developing highly selective, next-generation pest control agents.

2. CUADb-based oligonucleotide insecticides

In 2008, our research group pioneered the use of short unmodified antisense DNA oligonucleotides as contact insecticides (Oberemok, 2008). Initial work on the spongy moth (*Lymantria dispar*) demonstrated that effective gene silencing using oligonucleotide insecticides (oligoRING and oligoRIBO-11) depended critically on target gene selection, specific target regions within those genes, and target gene expression levels (Oberemok et al., 2017; Oberemok et al., 2019). Subsequent research identified rRNA as a prime target, leading to the development in 2019 of oligonucleotide insecticides based on 11-mer DNA sequences complementary to rRNA (Kumar et al., 2025). rRNA is an optimal target

due to its abundance (constituting ~80% of cellular RNA) and the substantial energy investment (>60% of cellular energy) required for ribosome production and maintenance (Szaflarski et al., 2022; Deng et al., 2022). Insect rRNA comprises 28S rRNA (~3900 nt), 18S rRNA (~1920 nt), 5.8S rRNA (~160 nt), 5S rRNA (~120 nt), and mitochondrial rRNAs including 16S rRNA (~1140 nt) and 12S rRNA (~600 nt) (Deng et al., 2022). The first successful rRNA targeting was achieved against *L. dispar* 5.8S rRNA (Oberemok et al., 2019). CUADb-based oligonucleotide insecticides are designed using the DNAInsector program (dnainsector.com) or manually via GenBank sequences of pest pre-rRNA and mature rRNA. Synthesis employs the phosphoramidite method through liquid-phase or solid-phase synthesis using instruments such as the ASM-800 (BIOSSET, Russia), OligoPilot™ (Cytiva, Sweden), or PolyGen 10-Column DNA Synthesizers (Gal'chinsky et al., 2019).

These insecticides demonstrate high efficacy against sternorrhynchans and thrips, with successful targeting documented across multiple species: 28S rRNA in *Unaspis euonymi*, *Dynaspidiotus britannicus*, *Icerya purchasi*, *Ceroplastes japonicus*, *Aonidia lauri*, and *Coccus hesperidum* (Gal'chinsky et al., 2020; Useinov et al., 2020; Oberemok et al., 2022; Gal'chinsky et al., 2023; Gal'chinsky et al., 2024; Oberemok et al., 2025a); 18S rRNA in *Pseudococcus viburni* (Novikov et al., 2023); mitochondrial 16S rRNA (Oberemok et al., 2025b); and ITS2 regions in *Macrosiphoniella sanborni*, *Schizolachnus pineti* (Puzanova et al., 2023; Oberemok et al., 2024b), *Trioza alacris* (Oberemok et al., 2024c), and *Tetranychus urticae* (Gavrilova et al., 2025), indicating potential for oligonucleotide acaricides (Kumar et al., 2025). A single contact treatment at 100 ng/μl typically achieves approximately 80% mortality in sternorrhynchans within 3-14 days (Oberemok et al., 2023; Oberemok et al., 2024a). While highly effective against hemipterans and lepidopterans including *L. dispar*, efficacy is notably lower against coleopterans such as *Leptinotarsa decemlineata* (Oberemok et al., 2018b). The 11-mer length provides species specificity with a uniqueness frequency exceeding $1/4.19 \cdot 10^6$, sufficient for most agricultural applications (Oberemok et al., 2022). rRNA's dual advantages of abundance and inter-species variability make it superior to less concentrated cellular RNAs as a target. Contact delivery (CUADs) outperforms oral delivery (ODUADs) due to digestive DNases and extra-oral salivary barriers in hemipterans (Cao et al., 2018).

The 2008 discovery was unexpected, contradicting established views that unmodified oligodeoxyribonucleotides are rapidly degraded by DNases (Dias and Stein, 2002) and that rRNA resists RNase H-dependent antisense DNA (Will and Lührmann, 2001; Bachellerie et al., 2002). Prior research focused exclusively on gene downregulation, overlooking insecticidal potential (Dias and Stein, 2002; Oberemok, 2008). Oligonucleotide insecticides (termed olinscides or DNA insecticides) refute these assumptions through a two-step DNA containment (DNAc) mechanism: initial target rRNA "arrest" halts ribosome function and triggers rDNA transcription hypercompensation, followed by degradation of target rRNA by DNA-guided rRNases such as RNase H1 (Kumar et al., 2025; Gal'chinsky et al., 2024). This "genetic zipper" mechanism (Oberemok et al., 2024c) induces metabolic shifts toward lipid-based energy synthesis, enhancing ribosome biogenesis and ATP production. Ultimately, widespread kinase downregulation – including mTOR, which regulates ribosome biogenesis through mTORC1 – causes "kinase disaster" due to ATP insufficiency, while significant RNase H1 upregulation occurs throughout DNAc (Oberemok et al., 2025a).

Three years after this discovery, Wang et al. (2011) adapted the contact concept for dsRNA insecticides. Crucially, DNAc downregulates key RNAi enzymes (DICER1, Argonaute 2, DROSHA), underscoring its mechanistic distinction from RNAi (Oberemok et al., 2025a). Thus, CUADb (DNAc) and dsRNA (RNAi) represent complementary but

fundamentally distinct antisense technologies with synergistic potential in pest management (Kumar et al., 2025) (Table 1).

Table 1. Comparative overview of CUAD biotechnology and dsRNA technology, highlighting differences in active substances, mechanisms, synthesis methods, and pest targets.

Key features	CUAD biotechnology	dsRNA technology
Active substance	ssDNA	dsRNA
Mechanism	DNA containment	RNA interference
Mode of action	ssDNA:rRNA	ssRNA:mRNA
Nuclease involved	DNA-guided rRNase (such as RNase H1)	Argonaute
Synthesis	Liquid-phase oligonucleotide synthesis based on phosphoramidite chemistry	Large-scale cell-free biomanufacturing (cell-free dsRNA production)
Target pests with best outcome	Sternorrhynchans	Beetles

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A working hypothesis proposes that DNA viruses may exploit the rRNA gene expression hypercompensation observed during the initial step of the DNA containment mechanism to increase cellular ribosome numbers essential for viral replication (Oberemok et al., 2025a). Bioinformatics studies indicate viral hijacking of host genes is common (Cerio et al., 2020). Given rRNA's dominance (~80% of cellular RNA) and the significant energy investment (>60%) in ribosome production (Deng et al., 2022; Szaflarski et al., 2022), DNA viruses likely co-opted rRNA-like sequences during co-evolution (Evseev et al., 2021).

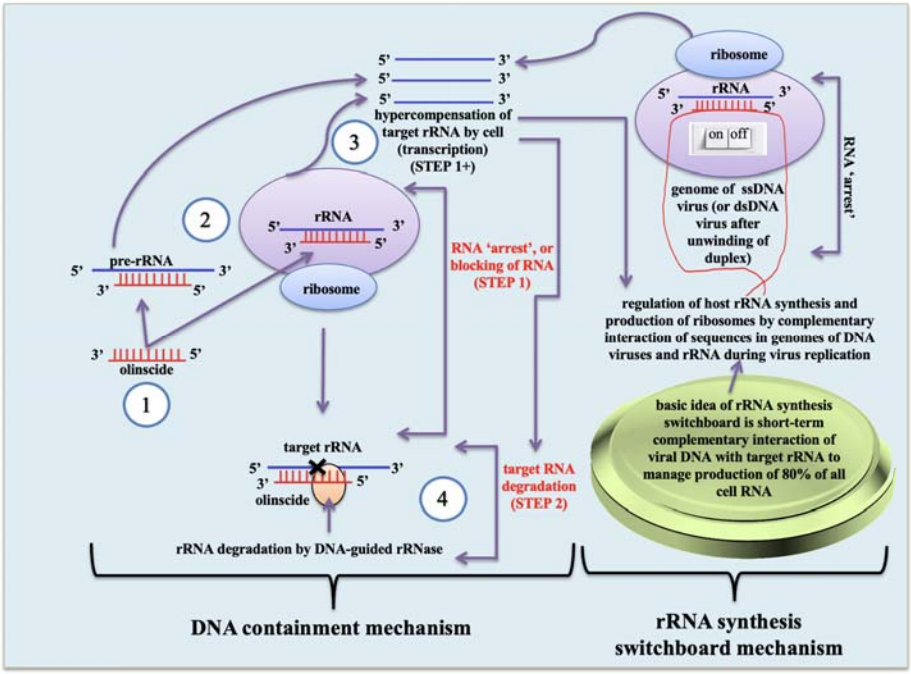

Figure 1. Figure 1. Mechanisms underlying oligonucleotide insecticides and rRNA regulation. Left: DNA containment mechanism (DNAc) for insect pest control. (1) Antisense DNA oligonucleotides complementary to pest mature rRNA or pre-rRNA are applied as insecticides. (2) Duplex formation occurs between the oligonucleotide and target rRNA/pre-rRNA. (3) In the first step of DNAc, antisense DNA arrests rRNA or pre-rRNA, leading to hypercompensation and interference with ribosome biogenesis ('arrested ribosomes'). (4) In the second step, DNA-guided rRNase

mechanism, where DNA viruses regulate host rRNA synthesis and ribosome production by introducing complementary sequences of viral DNA, influencing up to 80% of rRNA output.

Most DNA viruses replicate within the nucleus and interact with the nucleolus—a site critical for ribosome assembly and virus life cycles (Salvetti et al., 2024). Nucleoli facilitate rRNA synthesis, processing, and pre-ribosomal subunit assembly (Rawlinson et al., 2018). We hypothesize that specific DNA viral genome fragments could regulate host rRNA synthesis via an ON/OFF switch mechanism, using complementary interactions between viral DNA and host rRNA to initiate ribosome biogenesis with minimal energy expenditure (Oberemok et al., 2025a) (Figure 1). Additionally, DNAc may function within the innate immune system against ssDNA viruses (major vectors: hemipterans) and DNA viruses infecting hemipterans (Barnes and Turner, 2016; Wang and Blanck, 2021; Guo et al., 2022) by degrading viral DNA fragments (Gal'chinsky et al., 2024).

Targeting conserved regions of insect pest rRNA genes for oligonucleotide insecticide design delays target-site resistance development, as mutations occur less frequently in these areas (Sharma et al., 2014; Oberemok et al., 2019). Should resistance emerge, new effective CUADb insecticides can be designed by shifting the target site adjacent to the resistant region within the rRNA genes (Gal'chinsky et al., 2024). Notably, current CUADb-based pesticides demonstrate efficacy against approximately 30% of the most resistant arthropods (Whalon and Mota-Sanchez, 2015), showing substantial activity against Sternorrhyncha and spider mites (Kumar et al., 2025).

Oligonucleotide insecticides undergo rapid, 'painless' biodegradation in the environment (soils, water, plants) via abiotic factors (temperature, pH, salinity, UV radiation) and biotic factors (microbes, extracellular enzymes) (Barnes and Turner, 2016; Oberemok et al., 2024d). Deoxyribonucleases in tissue homogenates of *Lymantria dispar*, *Leptinotarsa decemlineata*, *Icerya purchasi*, *Dynaspidiotus britannicus*, *Aonidia lauri*, and their host plants (*Quercus pubescens*, *Solanum tuberosum*, *Pittosporum tobira*, *Laurus nobilis*) degrade olinscides within 24 hours at 27°C (Oberemok et al., 2018b; Oberemok et al., 2019; Gal'chinsky et al., 2023; Gal'chinsky et al., 2024), while *Macrosiphoniella sanborni* nucleases achieve degradation within 1 hour (Puzanova et al., 2023). Minimal negative impacts are observed on non-target organisms including *Triticum aestivum* (Oberemok et al., 2013; Nyadar et al., 2018), *Quercus robur*, *Malus domestica* (Zaitsev et al., 2015), *Manduca sexta*, *Agrotis ipsilon* (Oberemok et al., 2015), and *Galleria mellonella* (Oberemok et al., 2019). Unprecedented selectivity is evidenced by dramatic efficacy loss following single nucleotide substitutions (Puzanova et al., 2023; Gal'chinsky et al., 2024), although tolerance for non-canonical base pairs (A:C, C:A, G:U) must be considered during design (Gal'chinsky et al., 2024; Oberemok et al., 2024b). Efficacy extends to mixed pest populations (Gal'chinsky et al., 2024), and modern liquid-phase synthesis minimizes greenhouse gas emissions (nitrogen oxide, methane, carbon dioxide) compared to solid-phase methods (Gal'chinsky et al., 2023), addressing climate concerns.

In conclusion, widespread pesticide resistance necessitates novel, highly specific pesticide classes to reduce environmental impact. Oligonucleotide insecticides represent a bold advancement using unmodified antisense oligonucleotides. Mirroring the trajectory of therapeutic oligonucleotides—which overcame early challenges through sustained research and investment (TriLink BioTechnologies, 2025)—CUADb-based pesticides offer selective action, rapid biodegradation, and increasingly cost-effective production, presenting a highly promising approach (Kumar et al., 2022). Our estimates indicate the 'genetic zipper' method (CUADb) can potentially control 10-15% of insect pests using a simple, flexible algorithm (Oberemok et al., 2024c). Formulation additives (spreaders, adhesives, penetrators, UV protectants) may enhance efficacy for other insect orders, pending environmental safety assessment. Future research will undoubtedly uncover

deeper mechanistic details of DNAc and its potential. CUADb paves the way for sustainable, xenobiotic-free agriculture, positioning oligonucleotide pesticides as prospective cornerstone agents in pest control (Oberemok et al., 2025b).

3. RNA biocontrols

The foundation of RNA interference (RNAi) was established by Andrew Fire and Craig Mello's seminal discovery that antisense-strand double-stranded RNA (dsRNA) mediates sequence-specific gene silencing—a breakthrough recognized with the 2006 Nobel Prize in Physiology or Medicine (Fire et al., 1998). This pivotal work catalyzed extensive biotechnological applications in agricultural and forestry research for plant protection. RNAi operates through post-transcriptional and translational suppression of gene expression, utilizing dsRNA molecules typically exceeding 200 base pairs in length that are complementary to target genes (Tomoyasu, 2008; Svoboda, 2020). The mechanism initiates when the RNase III enzyme dicer cleaves long dsRNA into 20–30 nucleotide small interfering RNAs (siRNAs) (Carmell et al., 2002; Obbard et al., 2009; Koeppe et al., 2023). These siRNAs subsequently associate with Argonaute-family proteins to form RNA-induced silencing complexes (RISC) for cytoplasmic mRNA degradation, or RNA-induced transcriptional silencing complexes (RITS) for nuclear gene suppression (Wu et al., 2020; Müller et al., 2020). A parallel pathway involves PIWI-interacting RNAs (piRNAs; 23–36 nucleotides), germline-specific molecules that complex with PIWI-subfamily proteins to repress transposable elements through distinct silencing mechanisms (Bodemann et al., 2012; Chattopadhyay et al., 2022). Collectively, these RNAi pathways play crucial regulatory roles during insect ontogenesis (Figure 2), ultimately disrupting biosynthesis of specific proteins through targeted mRNA degradation or translational blockade.

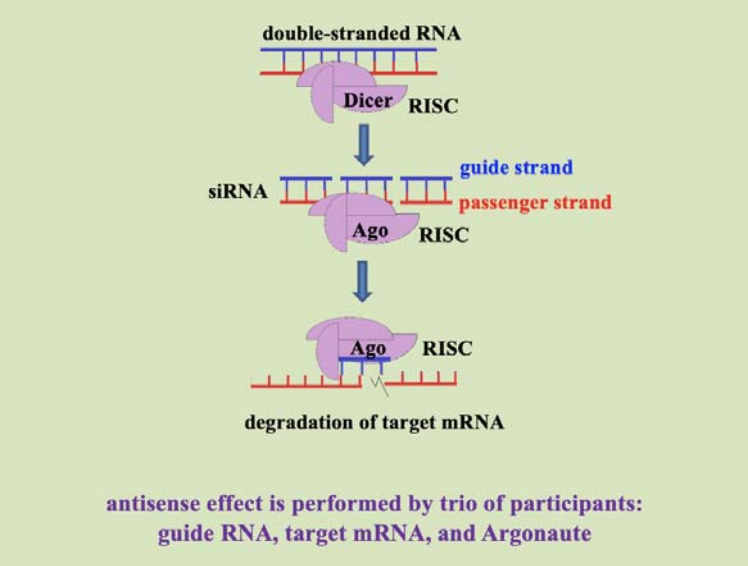

Figure 2. Main pathway of RNA interference (RNAi) utilized for the development of RNA-based biocontrols. Double-stranded RNA (dsRNA) is processed by Dicer into siRNAs, which are incorporated into the RNA-induced silencing complex (RISC). The guide strand directs Argonaute (Ago) to complementary target mRNA, resulting in mRNA cleavage and degradation. This mechanism, distinct from DNA containment (DNAc) and contact unmodified antisense DNA biotechnology (CUADb), forms the basis for RNAi-mediated pest control strategies.

RNAi efficacy exhibits profound taxonomic variation among economically significant insect pests. Coleopterans demonstrate consistently high systemic efficiency

locusts) and Blattodea (cockroaches) generally exhibit robust responses (Irles et al., 2013; Santos et al., 2014; Li et al., 2021; Hoang et al., 2022). Efficiency proves variable in Hymenoptera and Hemiptera due to limited systemic spreading in piercing-sucking insects, attributed primarily to deficient RNA-directed RNA polymerase (RdRP) activity (Christiaens et al., 2014; Jain et al., 2020, 2021). Lepidopterans and dipterans typically display low susceptibility (Miller et al., 2008; Terenius et al., 2011). Consequently, commercial RNAi biopesticide development remains largely confined to coleopteran targets (Bramlett et al., 2020; Rodrigues et al., 2021; Koo et al., 2024), with target mRNAs spanning genes governing development, detoxification, and reproduction (Liu et al., 2020) (Table 1).

Two primary delivery strategies exist: genetically modified (GM) plants expressing dsRNA, and topical application of formulated sprayable products (Ivashuta et al., 2015). GM approaches undergo protracted regulatory evaluation under biotechnology frameworks, whereas sprayable formulations—classified as biochemical pesticides—offer potential time and cost advantages, with registration timelines dependent on final product composition (Dietz-Pfeilstetter et al., 2021; Galli et al., 2024). Currently, few RNAi products are commercially deployed. The landmark SmartStax® PRO corn (US EPA-registered 2017) combines two Bt insecticidal proteins (Cry3Bb1 and Cry34/35Ab1) with DvSnf7 dsRNA targeting vesicle transport in corn rootworm (Head et al., 2017; EPA, 2024). This triple-mode-of-action pyramid achieves 99% suppression of western (*Diabrotica virgifera virgifera*) and northern (*D. barberi*) corn rootworm larvae, though dsRNA-induced mortality manifests slower than Bt toxicity (≥5 days post-ingestion) (Head et al., 2017). The targeted Snf7 protein functions within the ESCRT-III pathway governing transmembrane protein sorting and degradation (Vaccari et al., 2009; Bolognesi et al., 2012), with documented absence of cross-resistance between DvSnf7 RNAi and Bt toxins (Moar et al., 2017; Khajuria et al., 2018; Reinders et al., 2023). Experimental GM crops demonstrate efficacy against diverse pests including fall armyworm (*Spodoptera frugiperda*; Xiong et al., 2013), aphids (Mao et al., 2014), Colorado potato beetle (*Leptinotarsa decemlineata*; Guo et al., 2018), and spider mites (*Tetranychus urticae*; Shen et al., 2017).

The first exogenous sprayable dsRNA biopesticide, Calantha™ (ledprona; IRAC Group 35), was commercialized by GreenLight Biosciences in 2023 against Colorado potato beetle (Pallis et al., 2023; GreenLight Biosciences, 2025; Narva et al., 2025). Applied at 9.4 g/ha, it induces near-complete feeding cessation within 2-3 days and achieves 90-95% mortality in 1st-2nd instar larvae within 11-26 days. Operational constraints include strict larval-stage specificity, necessity for sequential applications, and recommended rotation with alternate modes of action. Proof-of-concept successes encompass numerous taxa: brown planthopper (*Nilaparvata lugens*; Zha et al., 2011), African sweet potato weevil (*Cylas puncticollis*; Prentice et al., 2017), tomato pinworm (*Tuta absoluta*; Camargo et al., 2016), oriental fruit fly (*Bactrocera dorsalis*; Sun et al., 2023), cotton mealybug (*Phenacoccus solenopsis*; Arya et al., 2021), diamondback moth (*Plutella xylostella*; Chen et al., 2021), fall armyworm (*Spodoptera frugiperda*; Li et al., 2024; Bera et al., 2025), white-backed planthopper (*Sogatella furcifera*; Mao et al., 2014; Guo et al., 2025), desert locust (*Schistocerca gregaria*; Schellens et al., 2022), and potato psyllid (*Bactericera cockerelli*; Lu et al., 2024)—though none have advanced beyond experimental stages.

Significant challenges impede broader adoption: 1) Target gene selection lacks consensus due to compensatory cellular responses including protein stability, functional redundancy among paralogs, and transcriptional feedback loops (Ulrich et al., 2015; Buer et al., 2024; Cedden et al., 2024); 2) Efficacy varies substantially with developmental stage, tissue type, and delivery method; 3) The Spray-Induced Gene Silencing (SIGS) paradigm suffers mechanistic ambiguity—whether dsRNA uptake occurs primarily via integument

penetration or oral ingestion remains unresolved, confounding optimization efforts (Dalakouras et al., 2021); 4) Specificity limitations: Off-target effects may arise from as few as 15 contiguous base matches (Powell et al., 2017; Chen et al., 2021) or sequence-independent immune activation (Hirai et al., 2004; Brutscher et al., 2017), compounded by formulants (nanocarriers, adjuvants) potentially enhancing non-target exposure (Castellanos et al., 2019; Reinders et al., 2023); 5) Rapid environmental degradation occurs via microbial nucleases in soil/phyllosphere, UV radiation, and precipitation runoff (Dubelman et al., 2014; Parker et al., 2019), with insect-derived nucleases in hemolymph (e.g., *Spodoptera litura*, *Manduca sexta*) and saliva (*Acyrthosiphon pisum*) degrading dsRNA within 0.3–3 hours (Christiaens et al., 2014; Garbutt et al., 2013; Peng et al., 2018). While dsRNA production costs have decreased through cell-free platforms like GreenLight's enzymatic NMP→NTP→dsRNA pipeline (Cunningham et al., 2020; Taning et al., 2020), RNA biocontrols remain challenged by slower lethality and shorter residual activity than conventional insecticides. Sprayable dsRNA offers theoretical advantages over GM crops but requires deeper mechanistic understanding of RNAi pathways and environmental safety validation. Presently, dsRNA insecticides demand optimized design algorithms and expanded target pest spectra, whereas CUADb-based oligonucleotide insecticides primarily require broader validation across additional pest groups (Oberemok et al., 2024d).

4. Conclusion

Although both CUAD biotechnology and double-stranded RNA technology utilize nucleic acids as active ingredients, they represent fundamentally distinct insecticidal approaches with divergent mechanisms and applications (Oberemok et al., 2018a). Oligonucleotide insecticides employ short, unmodified antisense DNA fragments delivered exclusively via contact, operating through the DNA containment (DNAc) mechanism to degrade target ribosomal RNAs in pests. The deoxyribose backbone of these DNA fragments confers greater resistance to environmental hydrolysis than RNA-based counterparts (Thorp, 2000), while their compact size (~11 nucleotides) enables exceptional targeting precision. Ribosomal RNA—constituting approximately 80% of cellular RNA—serves as the optimal target due to its abundance and metabolic significance (Oberemok et al., 2019, 2023; Gal'chinsky et al., 2024; Oberemok et al., 2025a), with current efficacy being most pronounced against sap-feeding sternorrhynchan insects (Gal'chinsky et al., 2024; Oberemok et al., 2024a). In contrast, RNA biocontrols utilize longer double-stranded RNA molecules delivered through either contact or oral routes, functioning via the RNA interference (RNAi) pathway to degrade messenger RNAs. The ribose backbone of these molecules increases susceptibility to environmental hydrolysis (Williams et al., 2016), while their extended length (typically >200 bp) and subsequent enzymatic dicing into smaller fragments complicate selectivity control, necessitating careful risk assessment for non-target organisms (Roberts et al., 2015; Christiaens et al., 2022; Rank and Koch, 2021; Liu et al., 2020). Both classes benefit from cost-effective synthesis: liquid-phase phosphoramidite methods for DNA oligonucleotides (Gal'chinsky et al., 2023; Oberemok et al., 2024b) and cell-free enzymatic production for dsRNA (Taning et al., 2020). Notably, while RNAi mechanisms were characterized years before practical dsRNA insecticides emerged, the DNAc mechanism was discovered following persistent applied research with antisense oligodeoxyribonucleotides in pest control. These complementary technologies will likely demonstrate optimal efficacy against specific pest taxa, offering extended utility through careful design and potential for synergistic integration in multi-target formulations. Their unprecedented safety profiles for non-target organisms and ecosystems—combining high selectivity, rapid environmental biodegra-

dation, and innovative mechanisms—position nucleic acid-based insecticides as indispensable biomolecules for sustainable agriculture's future.

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