

Pear Stigmatic Exudate microRNAs as Next-Generation Anti-Fire Blight Biocontrols

Cross-Kingdom Mechanisms Targeting *Erwinia amylovora*, Technical Barriers, and Translational Pathways for IPM

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BACKGROUND AND OBJECTIVE

- **Background:** Fire blight is a destructive bacterial disease affecting pome fruits. Current reliance on copper and antibiotics is limited by resistance risks, regulation, and inconsistent field performance. Pear stigmas serve as the primary infection site and secrete exudates containing regulatory small RNAs.
- **Objective:** To synthesize evidence regarding stigma-derived plant microRNAs (miRNAs) as bio-inspired antimicrobials against *E. amylovora* and map the steps for translation into integrated pest management (IPM).

MECHANISTIC CONCEPT

Hypothesis: Plant miRNAs in stigma exudate may enter bacterial cells to interfere with virulence networks or destabilize membranes, lowering colonization success.

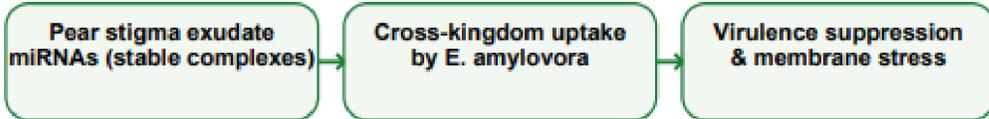


Figure 01: Figure. Conceptual cross-kingdom miRNA action at the infection court (stigma)

IMPLEMENTATION CHALLENGES

Stigma-derived miRNAs show promise for agricultural use, but successful field translation depends on overcoming barriers in production, delivery, and safety validation.

Technical Barrier	Why it limits deployment	Practical Direction
RNA degradation (RNases, UV, heat)	Short residence time reduces active dose at stigma surface.	Protective carriers (e.g., polysaccharide films); apply at bloom.
Low yield & variable composition of stigma exudate	Hard to standardize active ingredient and batch consistency.	Use synthetic or plant-produced miRNA mimics; define QC metrics.
Uncertain uptake and cellular targeting in bacteria	Mechanism may be indirect or strain-dependent.	Track uptake (fluorescent tags); validate with transcriptomics.
Non-target & phytotoxicity concerns	Impacts on pollinators and beneficial microbiota must be excluded.	Tiered ecotoxicology; field assays; select narrow-sequence candidates.
Regulatory pathway uncertainty	Plant-derived RNAs fall under evolving RNA biopesticide rules.	Early dialogue with regulators; align with RNA biopesticide guidance.

KEY FINDINGS

- **Microenvironment:** Stigmatic exudates are chemically rich environments where miRNAs accumulate during peak infection susceptibility.
- **Homology:** Candidate miRNAs show homology to plant defense and stress regulators, supporting their role in antimicrobial activity.
- **Cross-Kingdom Regulation:** In vitro assays suggest specific miRNAs can reduce bacterial virulence-associated expression and increase bacterial stress.

TRANSLATION TO PRACTICE

- **Discovery:** Profile stigma miRNAs across cultivars with contrasting susceptibility and disease pressure.
- **Validation:** Confirm uptake and target engagement in *E. amylovora* using reporter assays, transcriptomics, and mutants.
- **Formulation:** Protect RNAs using biopolymers or lipid/clay carriers and optimize stigma residence time.
- **Field Fit:** Integrate with bloom sprays, forecasting, and biologicals; test under rain, UV, and variable nectar loads.

PROPOSED ROADMAP

The proposed roadmap for stigma-targeted miRNA biocontrols outlines a comprehensive process, starting with profiling stigma miRNAs, selecting candidates and predicting targets, followed by uptake and target validation, carrier formulation, and culminating in orchard field trials.

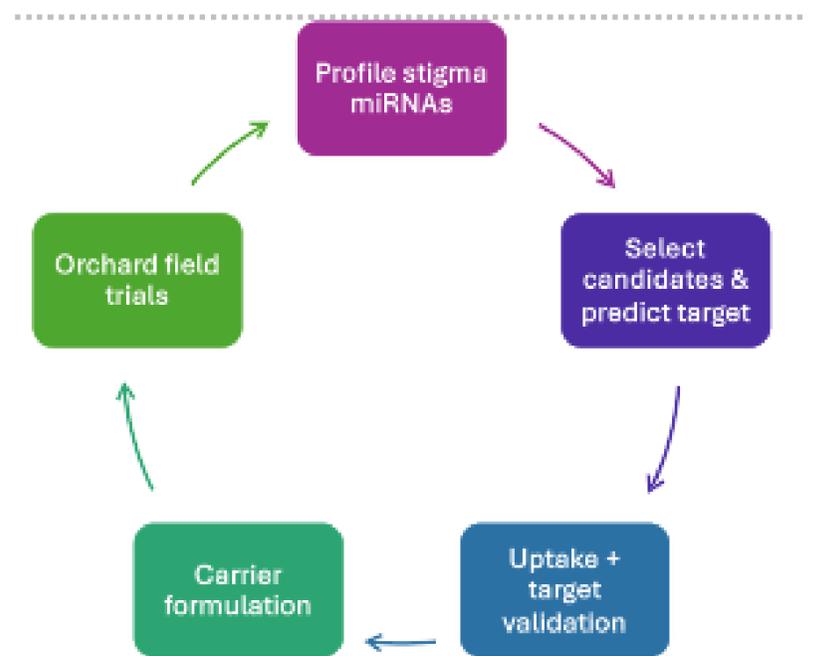


Figure 02: Proposed validation-to-field roadmap for stigma-targeted miRNA biocontrols.