Volatile Profiling Reveals Conserved VOC Responses Associated with Early *Pseudomonas syringae* pv. *aptata* Colonization of Beet Seeds

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Introduction

- *Pseudomonas syringae* pv. *aptata* (Psa), the causal agent of bacterial leaf spot, is a genetically diverse, seedborne pathogen. Its ability to colonize seeds asymptomatically facilitates its undetected introduction into crop ecosystems.
- Genetically distinct bacterial pathogens can produce distinct volatile fingerprints that reflect differences in their metabolism and physiological state (García-Barceló et al., 2026; Fitzgerald et al., 2020).
- Beyond serving as metabolic fingerprints, microbial VOCs mediate biological interactions, influence virulence, and enable non-destructive detection of seedborne pathogens in asymptomatic seeds (Wang et al., 2023; Gaudioso et al., 2025).”

Objectives

1. Characterize conserved and strain-specific volatile signatures associated with early Psa colonization of beet seed.
2. Identify candidate VOC biomarkers for early detection and bioactive compounds with potential biocontrol activity.

Experimental Workflow and Methods

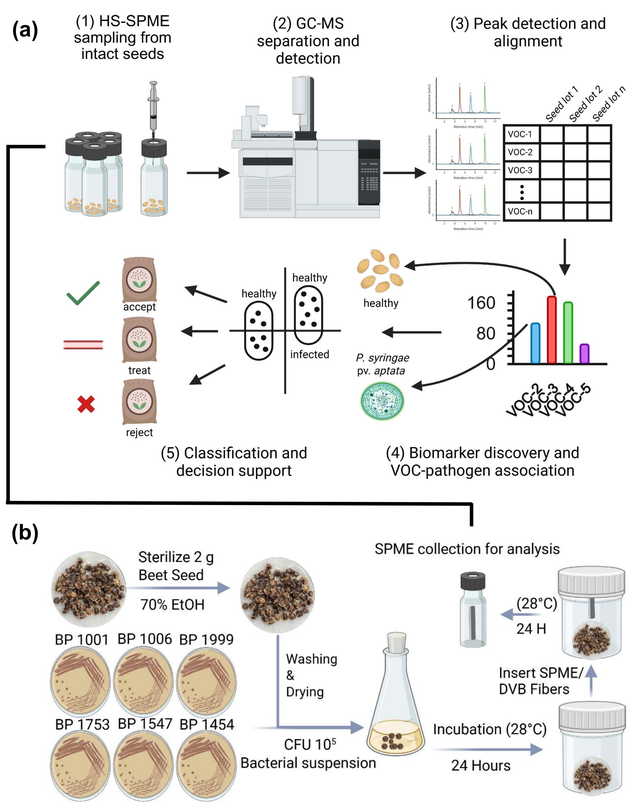

Figure 1 Experimental Workflow and Methods. (a) HS-SPME–GC–MS workflow for VOC collection, detection, and biomarker discovery in intact beet seeds. Identified VOC signatures can distinguish healthy from Psa-colonized seeds and support accept, treat, or reject decisions. **(b).** Experimental workflow for Psa inoculation and VOC collection. Surface-sterilized beet seeds were inoculated with six Psa strains, incubated at 28°C for 24 h, and sampled using SPME/DVB fibers for GC–MS analysis.

Results

Psa Strains Produce Distinct Volatilomic Fingerprints

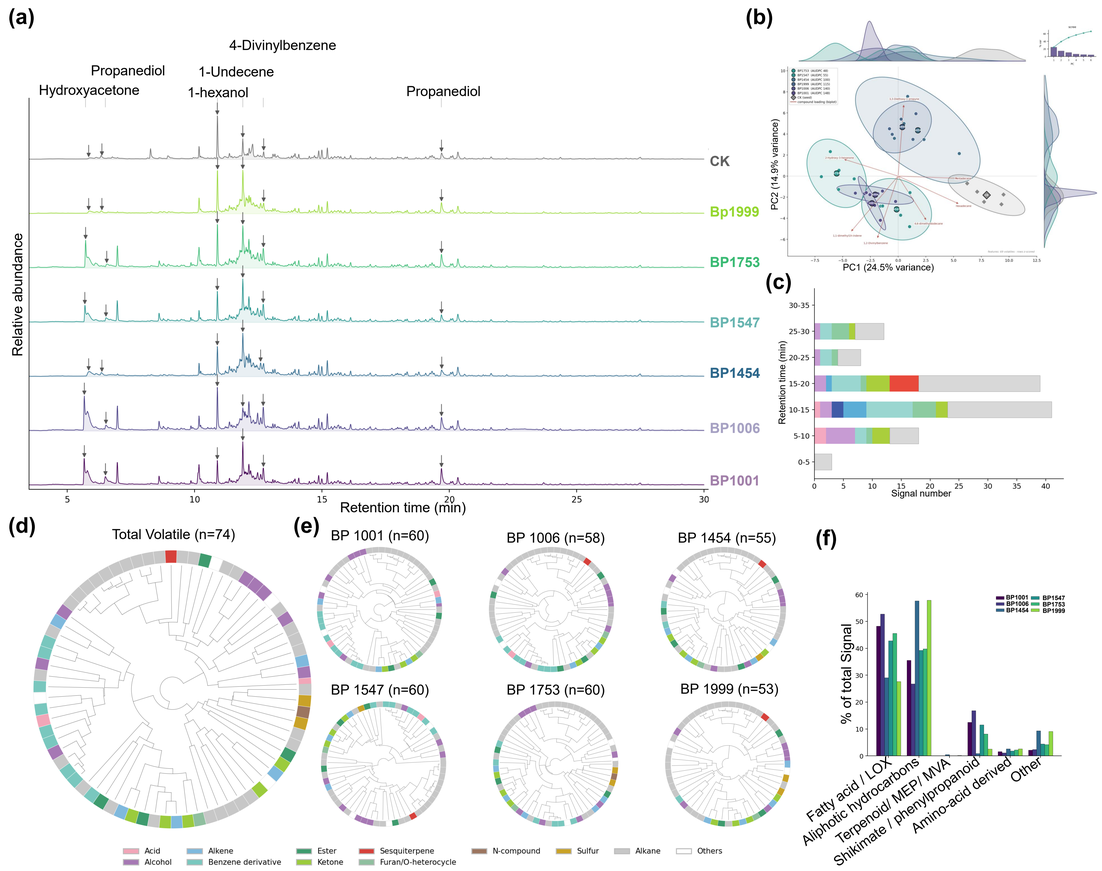

Figure 2. Strain-dependent VOC fingerprints of Psa-colonized beet seed (a) Stacked GC–MS chromatograms comparing CK and six Psa strains (b) PCA biplot showing treatment-specific clustering and marginal distributions. (c) VOC abundance and chemical-class distribution across retention-time intervals. (d–e) Classification of all 74 VOCs and strain-specific subsets. (f) Relative contributions of putative biosynthetic pathways to total signal. Overall, Psa altered VOC identity, abundance, and biochemical origin in a strain-dependent manner. CK, uninoculated seed control.

Virulence-Ranked VOC Signatures Associated with Psa

Figure 3. Virulence-Ranked VOC Signatures Associated with Psa. AUDPC values from Osabutey et al. (2025) were used for correlation analysis with VOC abundance. VOC abundance shifted with strain virulence: six compounds increased with AUDPC (r = 0.57–0.84), whereas seven were enriched in milder strains (r = −0.48 to −0.80). These opposing patterns reveal distinct volatile signatures associated with aggressive and mild Psa strains.

Discussion

- Psa inoculation altered the Pablo seed volatilome. PCA separated the seed control from inoculated samples, while strain-specific clustering showed variation among Psa strains.
- The 74 detected VOCs covered diverse chemical classes, with fatty acid/LOX and aliphatic hydrocarbon signals contributing the largest proportions.
- 1-Undecene and 3-methyl-2-heptanol increased after inoculation, although their abundance varied among strains. In contrast, 1-hexanol and 2-ethylhexanoic acid decreased, indicating bacterial consumption, transformation, or altered seed emissions.

Extracted-ion signal

Candidate Biomarker

Candidate Biocontrol

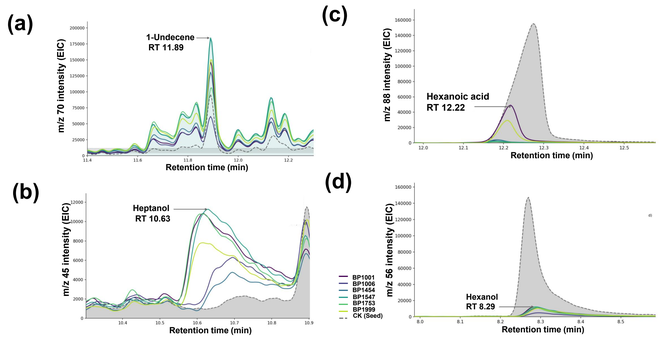

Figure 4. Extracted-ion chromatograms of candidate VOCs. (a–b) 1-Undecene and heptanol were enriched in Psa-inoculated seeds, supporting their use as candidate biomarkers. (c–d) Hexanoic acid and hexanol were depleted relative to CK and selected for further biocontrol evaluation.

Key findings

1. Psa colonization produced a clear, strain-dependent shift in the beet seed volatilome, revealing a multi-VOC signature separating inoculated seeds from uninoculated controls.
2. A multi-VOC signature, including 1-undecene and 3-methyl-2-heptanol, showed potential for early detection of Psa-colonized seed.
3. These findings support headspace volatilomics as a promising tool for identifying Psa-colonized seed and prioritizing compounds for biocontrol testing.

Future Directions

- Validate candidate VOC biomarkers in a Psa-resistant beet genotype to assess their robustness across host genetic backgrounds.
- Evaluate identified VOCs for biocontrol activity against Psa and their ability to reduce seed colonization and disease development.

Citations

1. García-Barceló et al. (2026). doi.org/10.1038/s41598-026-58199-2
2. Wang et al. (2023). doi.org/10.1038/s41396-023-01356-6
3. Gaudioso et al. (2025). doi.org/10.3390/agronomy15020497
4. Osabutey et al (2025). APS Plant Health.

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