Policy Research Journal
ISSN (E): 3006-7030 ISSN (P) : 3006-7022 Volume 4, Issue 3, 2026

SPATIAL METABOLOME–TRANSCRIPTOME INTEGRATION TO DECODE NUTRIENT COMPETITION BETWEEN PLANTS AND NECROTROPHIC FUNGI

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DOI:http://doi.org/10.5281/zenodo.22642403

Keywords
Spatial transcriptomics, Mass Spectrometry Imaging, spatial metabolomics, plant–pathogen interactions, necrotrophic fungi, Spatial Flux Balance Analysis, nutrient competition, metabolic reprogramming, phytoalexins, multi-omics integration

Article History
Received: 15 January 2026
Accepted: 28 February 2026
Published: 14 March 2026

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Abstract

Plant–necrotrophic fungal interactions represent a spatially dynamic molecular battlefield where host plants and pathogens compete for metabolic control over infected tissues. Traditional bulk omics approaches homogenize these intricate spatial architectures, obscuring localized metabolic fluxes and intercellular signaling events critical for understanding nutrient competition. This review synthesizes recent advances in spatial multi-omics technologies that enable high-resolution mapping of host–pathogen metabolic interactions. Spatial transcriptomics platforms, including array-based sequencing (10x Visium, Stereo-seq) and imaging-based methods (MERFISH, CosMx SMI), provide in situ gene expression profiles at resolutions ranging from subcellular to 55 µm, while Spatial MetaTranscriptomics (SmT) enables simultaneous host and fungal transcript detection. Concurrently, Mass Spectrometry Imaging techniques such as MALDI-MSI, DESI-MSI, and AFADESI-MSI facilitate untargeted spatial mapping of primary metabolites, phytoalexins, and phytotoxins across infection fronts. The integration of these complementary datasets through computational frameworks like SpatialMETA (deep generative modeling) and Spatial Flux Balance Analysis (spFBA) bridges resolution mismatches and translates transcriptomic data into predictive metabolic flux maps. Integrated spatial analyses reveal that infected tissues organize into concentric micro-niches necrotic cores exhibiting fungal nutrient depletion, lesion interfaces characterized by host cell-wall invertase and phytoalexin accumulation forming defensive perimeters, and distal zones undergoing systemic metabolic priming. This spatial framework illuminates how host plants rewire carbon and nitrogen metabolism to starve invading pathogens while necrotrophic fungi employ detoxification mechanisms to dismantle chemical barriers. Despite technical challenges including resolution disparities, tissue permeabilization optimization, and metabolite identification limitations, spatial

metabolome–transcriptome integration promises transformative insights for engineering climate-resilient crops with enhanced metabolic defense barriers.

1. Introduction

Plant–fungal interactions represent a dynamic, spatially constrained molecular arms race in which the host plant and invading pathogen continually compete for metabolic control over infected tissue. Among fungal pathogens, necrotrophs such as *Botrytis cinerea* and *Sclerotinia sclerotiorum* employ aggressive infection strategies characterized by the secretion of cell-wall-degrading enzymes, phytotoxins, and reactive oxygen species to trigger localized host cell death, thereby liberating cellular contents for fungal nutrient acquisition (Peyraud et al., 2017). Despite the destructive nature of these pathogens, host defense activation is rarely uniform across the affected organ. Instead, plant tissues establish distinct spatial micro-niches comprising a necrotic infection core, an adjacent hypersensitive or defense-active interface (lesion border), and distal, symptomless tissue undergoing systemic metabolic priming (Silva et al., 2015).

Historically, investigating the metabolic competition between plants and necrotrophic fungi relied on bulk transcriptomic and metabolomic analyses. While bulk approaches provide extensive inventories of gene expression and chemical accumulation, the homogenization of whole tissues inevitably dilutes microscale spatial gradients, masking localized metabolic fluxes and intercellular signaling events (TAN et al., 2009). The advent of single-cell RNA sequencing (scRNA-seq) enabled the resolution of transcriptomic heterogeneity among individual cells. However, scRNA-seq requires enzymatic protoplasting a process that destroys the physical architecture of plant tissues, triggers mechanical stress responses, and strips away the spatial context required to understand localized host–pathogen interactions (Shaw et al., 2021).

To address these limitations, spatially resolved omics technologies have emerged as transformative tools in plant pathology. Spatial transcriptomics (ST) enables the in situ quantification of thousands of transcripts while retaining spatial coordinates within intact tissue architectures. Concurrently, spatial metabolomics powered by Mass Spectrometry Imaging (MSI) facilitates the untargeted, spatial mapping and quantification of primary nutrients and secondary defense metabolites directly from sectioned tissues (Barmukh et al., 2025).

The cross-modal integration of spatial transcriptomics and spatial metabolomics provides a comprehensive analytical framework for decoding plant–pathogen metabolic interactions. By bridging spatial gene expression profiles with local metabolite distributions, multi-modal spatial integration uncovers how host plants reorganize local carbon and nitrogen flux to starve or encapsulate pathogens, and conversely, how necrotrophic fungi manipulate host metabolic networks to extract vital nutrients (Deng et al., 2026).

2. Technological Foundations of Spatial Multi-Omics in Phytopathology

Decoding nutrient competition at the host–pathogen interface requires specialized analytical platforms capable of capturing nucleic acids and small molecules from heterogeneous tissue sections without compromising spatial integrity. The spatial multi-omics workflow begins with intact cryo-sectioned plant-pathogen tissue, which is processed along parallel transcriptomic and metabolomic arms. Spatial transcriptomics utilizes sequencing or imaging arrays to generate discrete gene count matrices, while spatial metabolomics uses mass spectrometry imaging to produce continuous spectral intensity maps (Cassat et al., 2018). These complementary datasets are subsequently aligned through computational integration frameworks, such as probabilistic deep learning or spatial constraint-based flux balance analysis, to yield high-resolution maps of metabolic sinks, phytoalexin barriers, and host-pathogen nutrient exchanges across lesion micro-niches (Li & Tang, 2024).

2.1 Spatial Transcriptomics and Spatial Metatranscriptomics (SmT)

Spatial transcriptomics platforms applied to plant tissues are generally categorized into array-based sequencing approaches and in situ hybridization or imaging-based technologies. Array-based techniques such as 10x Visium, Spatial Enhanced Resolution Omics Sequencing (Stereo-seq), and high-density barcoded capture arrays transfer mRNA from fresh-frozen tissue sections onto glass slides coated with spatially barcoded reverse-transcription primers. Once permeabilized, transcripts hybridize to capture probes, creating spatially barcoded cDNA libraries for high-throughput sequencing (Yao et al., 2025).

To concurrently capture pathogen dynamics, Spatial metaTranscriptomics (SmT) was introduced, using multimodal arrays containing poly-d(T) capture probes alongside targeted ribosomal RNA primers targeting bacterial 16S and fungal ITS/18S regions. SmT enables the simultaneous mapping of host transcriptomic reprogramming and fungal colonization patterns across tissue sections at resolutions as fine as 55 µm. In parallel, targeted imaging-based methods including Multiplexed Error-Robust Fluorescence In Situ Hybridization (MERFISH), CosMx Spatial Molecular Imager (SMI), Xenium In Situ, and PHYTOMap offer single-cell or subcellular resolution by sequentially hybridizing fluorescent probes to pre-selected candidate genes (Saarenpää et al., 2024).

Applying these transcriptomic platforms to plant–fungal systems poses technical challenges. Plant cell walls and fungal chitin walls present rigid mechanical barriers that hinder uniform tissue permeabilization, often leading to lateral transcript diffusion or incomplete RNA release. Additionally, tissue-specific autofluorescence and varying enzymatic digestion efficiencies complicate spatial transcript recovery at the infection interface, requiring specialized cryo-sectioning and permeabilization protocols tailored to infected plant tissues (Cole et al., 2021).

2.2 Spatial Metabolomics via Mass Spectrometry Imaging (MSI)

Spatial metabolomics employs Mass Spectrometry Imaging (MSI) to map the spatial distribution and chemical abundance of endogenous metabolites in situ without requiring chemical labeling or prior extraction. The principal ionization methods utilized in plant research include Matrix-Assisted Laser Desorption/Ionization (MALDI-MSI), Desorption Electrospray Ionization (DESI-MSI), and Airflow-Assisted DESI (AFADESI-MSI) (Min et al., 2024).

MALDI-MSI requires coating the tissue section with an organic matrix that absorbs laser energy to ionize target analytes. Recent advancements, such as MALDI-2 (laser-induced post-ionization) paired with Fourier-transform ion cyclotron resonance (FT-ICR) or Orbitrap mass analyzers, have significantly enhanced sensitivity and spatial fidelity, enabling metabolite mapping at sub-micrometer to 10-µm spatial resolution (Chughtai & Heeren, 2010).

In contrast, DESI-MSI and AFADESI-MSI operate under ambient conditions by directing an electrically charged solvent spray onto the tissue surface, desorbing analytes with minimal sample preparation and no matrix requirement. AFADESI-MSI exhibits high sensitivity for polar compounds, efficiently mapping primary energy metabolites (such as hexoses, sucrose, and TCA cycle intermediates) and amino acids alongside specialized defense compounds (such as phenylpropanoids, flavonoids, and terpenoids) across host–pathogen infection fronts (Sampath , 2024).

Table 1: Comparison of Spatial Transcriptomics and Spatial Metabolomics Platforms in Plant–Pathogen Systems.

Technology Category	Specific Methods	Spatial Resolution	Target Molecules	Key Advantages in Phytopathology	Technical Limitations

Sequencing-based Spatial Transcriptomics	10x Visium, Stereo-seq, SmT	10–55 µm (down to sub-micron for Stereo-seq)	Whole transcriptome (poly-A mRNA); Dual host-fungal rRNA/mRNA (SmT)	Unbiased, genome-wide discovery; simultaneous detection of host and fungal transcripts.	Resolution often spans multiple cells; requires tissue fixation and permeabilization optimization.
Imaging-based Spatial Transcriptomics	MERFISH, CosMx SMI, Xenium, PHYTOMap	Sub-cellular to 1 µm	Targeted mRNA panels (tens to thousands of genes)	High spatial resolution; accurate single-cell boundary assignment in plant tissues.	Limited to pre-selected target genes; high cost; optical interference from cell wall autofluorescence.
Matrix-Assisted MSI (Metabolomics)	MALDI-MSI, MALDI-2-MSI	1–10 µm	Lipids, secondary metabolites, phytoalexins, small peptides	Ultra-high spatial resolution; broad chemical coverage; enhanced sensitivity with MALDI-2.	Requires uniform matrix deposition; potential matrix interference in low mass-to-charge (m/z) range.
Ambient Mass Spectrometry Imaging	DESI-MSI, AFADESI-MSI	20–100 µm	Primary metabolites (sugars, amino acids, organic acids), polar secondary compounds	Minimal sample preparation; no matrix needed; high throughput for polar metabolic profiling.	Lower spatial resolution compared to MALDI; potential surface spot dragging or analyte displacement.

3. Methodological and Computational Frameworks for Integration

Combining spatial transcriptomics and spatial metabolomics datasets presents computational challenges arising from fundamental differences in data structure, spatial resolution, and sample preparation requirements. Data processing begins with raw spatial transcriptomic count matrices and continuous mass spectrometry intensity spectra, which undergo co-registration across matched or adjacent tissue sections to establish spatial coordinate alignment. Once preprocessed, these multi-modal inputs are channeled into three computational integration modalities (Vandereyken et al., 2021): probabilistic joint embeddings (e.g., deep generative models like SpatialMETA), direct spatial correlation mapping of biosynthetic pathways, and constraint-based metabolic modeling using Spatial Flux Balance Analysis (spFBA). These integrated workflows ultimately output spatial flux enrichment scores, delineate metabolic sink-source transitions, and resolve host-pathogen defense frontiers (Jin et al., 2026).

3.1 Overcoming Multi-Modal Heterogeneity and Resolution Mismatch

The primary obstacle in spatial multi-omics integration stems from data structure disparities: spatial transcriptomics yields discrete molecular count matrices characterized by high sparsity, whereas spatial metabolomics generates

continuous mass spectrometry signal intensity vectors across a continuum of mass-to-charge (m/z) ratios. Furthermore, a spatial resolution mismatch exists between technologies; high-density transcriptomic slides or imaging platforms often capture data at single-cell scale (1–10 µm), whereas ambient spatial metabolomics platforms operate at 20–100 µm pixel resolutions (Ge et al., 2025). Finally, sample preparation protocols conflict: transcriptomics requires tissue fixation, permeabilization, and enzymatic treatment, whereas metabolomics demands rapid cryo-freezing to prevent enzymatic degradation and metabolite diffusion, precluding the collection of both data modalities from a single physical section (Giacomello & Lundeberg, 2018).

To resolve these discrepancies, computational frameworks utilize adjacent co-registered tissue sections and advanced statistical alignment algorithms. Frameworks like SpatialMETA utilize Conditional Variational Autoencoders (CVAEs) to project unpaired or paired spatial transcriptomics and spatial metabolomics data into a shared, low-dimensional latent space. SpatialMETA employs modality-specific decoders and tailored loss functions to correct for batch effects, bridge resolution mismatches, and preserve spatial morphology (Isik et al., 2026).

Additionally, point-to-point spatial mapping workflows align high-resolution optical images of stained tissue sections, enabling spatial co-localization analysis between transcript expression (such as biosynthetic genes) and metabolite intensities (such as pathway end-products) (Murakami & Heintz, 2022).

3.2 Predictive Metabolic Modeling: Spatial Flux Balance Analysis (spFBA)

While co-localization analyses reveal spatial correlations, gene expression alone does not equal metabolic flux due to post-transcriptional regulation, substrate limitations, and thermodynamic constraints. To translate spatial transcriptomic data into functional metabolic outputs, Spatial Flux Balance Analysis (spFBA) integrates spatial transcriptomics with genome-scale metabolic network reconstructions (Wang et al., 2026).

In the spFBA workflow, transcript abundances at each spatial coordinate (spot or cell) are processed through Gene-Protein-Reaction (GPR) logical rules to compute a Reaction Activity Score (RAS) for every metabolic reaction j at spot s:

$$RAS_{j,s} = f(\text{mRNA}_{g,s})$$

Where GPR logic resolves boolean AND relations (protein complexes) using minimum expression values, and OR relations (isoenzymes) using sum expression values (Maspero et al., 2024).

Next, Flux Variability Analysis (FVA) establishes the global upper ($\mathbf{v}^{\max}$) and lower ($\mathbf{v}^{\min}$) bounds for each reaction within a steady-state metabolic model defined by the stoichiometric matrix $\mathbf{S}$:

$$\mathbf{S} \cdot \mathbf{v} = \mathbf{0}, \quad \mathbf{v}^{\min} \leq \mathbf{v} \leq \mathbf{v}^{\max}$$

The spot-specific $RAS_{j,s}$ values are then applied as local constraints to scale the lower ($lb_{j,s}$) and upper ($ub_{j,s}$) flux boundaries for each spot s:

$$lb_{j,s} = v_j^{\min} \cdot \left(\frac{RAS_{j,s}}{\max_s(RAS_{j,s})} \right)$$

$$ub_{j,s} = v_j^{\max} \cdot \left(\frac{RAS_{j,s}}{\max_s(RAS_{j,s})} \right)$$

By sampling the constrained solution space across all spatial coordinates, spFBA generates Flux Enrichment Scores (FES) for individual metabolic reactions. This spatial flux modeling predicts localized reaction rates, directional nutrient uptake and secretion, and metabolic sink-source transitions across the host–pathogen lesion landscape (Lularevic et al., 2019).

Table 2: Computational Tools and Integration Strategies for Spatial Multi-Omics.

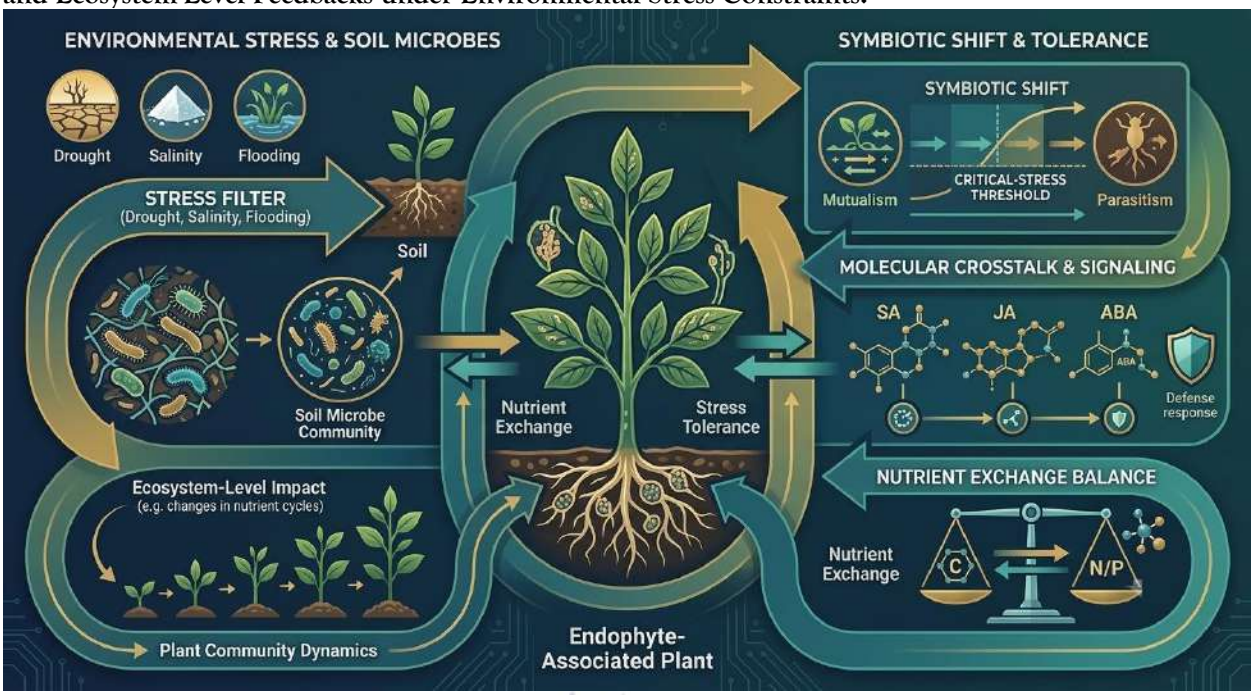
Computational Tool / Framework	Input Modalities	Algorithmic / Mathematical Model	Primary Function in Plant–Pathogen Studies
SpatialMETA	Spatial Transcriptomics (ST)	Conditional Variational Autoencoder (CVAE) deep generative model.	Modality fusion, cross-modal prediction, batch correction, and spatial

	+ Spatial Metabolomics (SM)		cluster identification across non-identical tissue sections.
Spatial Flux Balance Analysis (spFBA)	Spatial Transcriptomics + Genome-Scale Metabolic Models (GEMs)	Constraint-based linear programming; Gene-Protein-Reaction (GPR) mapping; FVA corner sampling.	Calculates localized Reaction Activity Scores (RAS) and Flux Enrichment Scores (FES) to map reaction fluxes and metabolic sink/source shifts.
Point-to-Point Spatial Correlation	Matched AFADESI-MSI / MALDI-MSI + ST	Non-linear image registration; pixel-to-spot interpolation; bivariate spatial correlation.	Directly links biosynthetic enzyme transcript abundance with target metabolite accumulation in defined tissue zones.
Spatial metaTranscriptomics (SmT) Pipeline	Dual-capture spatial sequencing arrays	Spatial demultiplexing; rRNA read alignment to host/fungal databases; spatial network clustering.	Maps inter-kingdom spatial correlations between host defense response transcriptomes and microbial pathogen hotspots.

4. Decoding Spatial Micro-Niches of Nutrient Competition

The spatial integration of transcriptomic and metabolomic data reveals that plant–necrotroph interactions are characterized by highly structured, concentric micro-niches. Rather than serving as an unstructured battlefield, infected leaves display distinct spatial zones where nutrient acquisition strategies and metabolic defense barriers conflict. Spatially, this zonation transitions from the necrotic core (characterized by dense fungal hyphae, depleted host sugars, and high phytotoxin concentration), through the lesion interface or border zone (where host cell-wall invertases, hexose transporters, and secondary phytoalexin accumulation form a defensive perimeter), to the distal uninfected tissue (where host cells undergo systemic metabolic priming and nitrogen reallocation) (Amin, 2026).

Figure 1. Conceptual Framework of Endophyte–Host Plant Symbiotic Dynamics, Molecular Signaling, and Ecosystem-Level Feedbacks under Environmental Stress Constraints.



4.1 Primary Metabolic Rewiring and Carbon/Nitrogen Sinks

Necrotrophic fungi rely on extracting host primary metabolites to sustain hyphal expansion and spore development. Integrated spatial profiling demonstrates that pathogens like *Botrytis cinerea* manipulate host tissue architecture to convert the infection site into a metabolic sink.

Within the necrotic core, spatial metatranscriptomics (SmT) captures high densities of fungal transcripts encoding hexose transporters, amino acid permeases, and cell-wall-degrading enzymes. Spatial metabolomics shows a corresponding depletion of host primary nutrients specifically glucose, fructose, sucrose, and essential amino acids confirming intense fungal consumption within dead or dying cells (Corwin et al., 2016).

At the lesion interface or border zone, the host plant attempts to counter fungal nutrient hijacking by reprogramming its primary metabolic pathways. Spatial transcriptomics reveals strong, localized induction of host cell-wall invertases (CWINVs) and hexose transporters (Sugar Transport Proteins, STPs) in cells immediately adjacent to fungal hyphae. CWINVs hydrolyze apoplastic sucrose into glucose and fructose, retrieving sugars back into host cells to prevent fungal acquisition while fueling the high energetic cost of local plant defense (Sánchez et al., 2026).

Application of spFBA to this interface reveals a localized shift toward glycolytic flux and tricarboxylic acid (TCA) cycle activity, accompanied by elevated Reaction Activity Scores (*RAS*) for ATP-generating pathways. Concurrently, spatial metabolomics highlights the hyper-accumulation of specific amino acids such as L-glutamine, L-proline, and organic acids like (*R*)-2-methylmalate in cotyledon and leaf border cells, serving as metabolic precursors for defense proteins and nitrogen scavenging (Deng & Walther, 2020).

In the distal uninfected zone, host tissues undergo systemic metabolic reprogramming, repressing photosynthetic gene expression while activating amino acid transport networks to reallocate resources away from the expanding fungal front (Wajid et al., 2026).

4.2 Spatial Zonation of Defense Metabolites and Counter-Defense

The spatial defense perimeter established by host plants is anchored by the localized synthesis and accumulation of secondary defense metabolites, including phytoalexins, flavonoids, and phenolic acids (Fahlberg et al., 2019).

Spatial metabolomics maps reveal that antimicrobial phytoalexins are strictly compartmentalized in a narrow ring surrounding the infection interface, forming a chemical barrier against advancing necrotrophic hyphae. Spatial transcriptomics demonstrates that genes encoding key enzymes in these secondary metabolic pathways such as phenylalanine ammonia-lyase (PAL), hydroxycinnamoyltransferase (HCT), 3-hydroxy-3-methylglutaryl-CoA reductase (HMGR), and 1-deoxy-D-xylulose 5-phosphate synthase (DXS) are upregulated specifically in host cells lining the lesion border, rather than uniformly throughout the leaf (Maia et al., 2024).

Table 3: Micro-Niche Mapping of Plant–Necrotroph Interactions Revealed by Integrated Spatial Omics.

Lesion Spatial Zone	Host Transcriptional & Flux Signature (ST / spFBA)	Pathogen Transcriptional Signature (SmT)	Metabolomic Profile (Spatial MSI)	Primary Biological Function
Necrotic Core	Cell death pathway induction; shutdown of host primary metabolic GRNs.	High expression of cell-wall degrading enzymes (CWDEs), hexose transporters, phytotoxins.	Depleted host sugars/amino acids; high accumulation of fungal secondary toxins.	Active fungal proliferation and nutrient extraction from dead host tissue.
Lesion Interface (Border Zone)	Strong upregulation of CWINVs, STPs, PAL, HCT, HMGR; high spFBA glycolytic flux.	Effector gene secretion; localized expression of phytoalexin detoxification enzymes.	Hyper-accumulation of phytoalexins, phenolic acids, L-glutamine, L-proline.	Host containment wall establishment; primary-to-secondary metabolic flux shift.
Symptomless Periphery	Repression of photosynthesis; induction of systemic acquired resistance (SAR) genes.	Minimal to absent pathogen reads.	Nitrogen remobilization intermediates; baseline primary sugars; primed signal precursors.	Systemic metabolic priming and nutrient reallocation away from the lesion.

Necrotrophic fungi counter this defensive perimeter through spatially targeted detoxifying mechanisms. SmT profiling reveals that fungal hyphae at the lesion front express specific ABC transporters and cytochrome P450 monooxygenases that degrade or efflux host phytoalexins. The spatial overlay of transcriptomics and metabolomics captures this chemical arms race in real time, demonstrating how necrotrophs attempt to dismantle the host chemical barrier to advance into healthy tissue (Gay et al., 2023).

5. Methodological Challenges and Future Horizons

Despite the unprecedented insights offered by spatial metabolome–transcriptome integration, several technological and computational hurdles must be addressed to fully realize its potential in plant pathology. A major bottleneck involves spatial resolution disparities and multi-modal co-

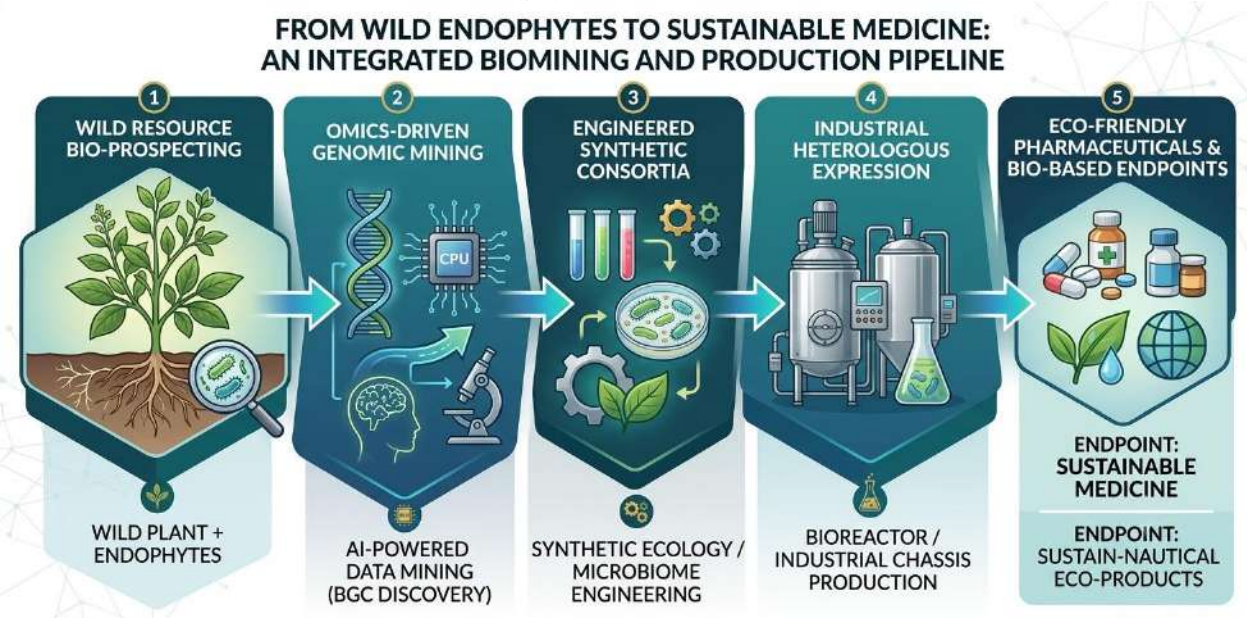
registration across non-identical tissue sections. While single-cell spatial transcriptomics platforms achieve near-subcellular resolution, ambient mass spectrometry imaging platforms like DESI and AFADESI operate at coarser resolutions ranging from 20 to 50 µm (Nobori, 2025). Consequently, aligning continuous metabolite intensities with discrete gene count matrices requires non-linear image registration and probabilistic deep generative modeling, which can introduce smoothing artifacts at narrow cellular borders. Advanced ionization techniques such as MALDI-2 post-ionization and cryo-nanoscale secondary ion mass spectrometry (nanoSIMS) are emerging to push spatial metabolomics toward true sub-micron, single-cell resolution, facilitating seamless alignment with high-density transcriptomic arrays (Baião et al., 2025).

Mechanical properties of infected plant tissues further complicate sample preparation and analyte extraction. The presence of rigid, pectin-rich plant cell walls combined with tough, chitinous fungal hyphae creates non-uniform physical barriers during tissue cryo-sectioning and permeabilization (Koshani et al., 2025). Over-permeabilization causes horizontal diffusion of soluble mRNA molecules and small metabolites across the slide surface, whereas under-permeabilization reduces transcript capture efficiency and leads to incomplete fungal RNA extraction. Standardizing tissue-specific permeabilization matrices and developing specialized cryo-embedding media that preserve both nucleic acid integrity and metabolite spatial localization represent critical methodological priorities (Saxena, 2025).

From a chemical perspective, untargeted spatial metabolomics faces persistent compound identification challenges. Mass spectrometry imaging identifies analytes based primarily on mass-to-charge (m/z) ratios, which often cannot discriminate between structural isomers or stereoisomers without tandem mass spectrometry (MS/MS) fragmentation. On-tissue MS/MS derivatization, coupled with high-field ion mobility mass spectrometry and localized chemical standard arrays, will be required to unambiguously annotate novel plant phytoalexins and fungal phytotoxins directly within tissue micro-niches (Zhang et al, 2024).

Finally, current spatial multi-omics technologies provide static, single-time-point snapshots of destructive host-pathogen interactions. To capture the dynamic flux of nutrient competition, future research must integrate spatial multi-omics with live-cell biosensor imaging, stable isotope tracing, and real-time functional single-cell analyses. Furthermore, incorporating spatial proteomics alongside spatial transcriptomics and metabolomics will complete the central dogma spectrum in situ (Biteen et al., 2016). Combining these multi-layered spatial datasets with predictive constraint-based models, such as spatial Flux Balance Analysis (spFBA), will empower researchers to construct digital twin platforms of infected plant organs, driving the rational design of climate-resilient crops with engineered metabolic defense barriers against necrotrophic pathogens (Kundu & Tanti, 2026).

Figure 2. An Integrated Multi-Stage Pipeline for Bioprospecting, Mining, Engineering, and Scale-Up Production of Endophyte-Derived Secondary Metabolites.



Conclusion

The integration of spatial transcriptomics and spatial metabolomics has revolutionized our understanding of plant–necrotrophic fungal interactions by resolving the spatially structured metabolic competition that bulk analyses previously obscured. This multi-modal approach has revealed that infected tissues organize into functionally distinct concentric micro-niches, each characterized by unique transcriptional programs, metabolic fluxes, and host–pathogen exchange dynamics. The lesion interface emerges as a critical defensive frontier where host plants deploy localized primary metabolic reprogramming upregulating cell-wall invertases and hexose transporters alongside secondary metabolite barriers of phytoalexins and phenolic compounds to contain advancing fungal hyphae. Concurrently, necrotrophic pathogens employ spatially targeted detoxification mechanisms and nutrient acquisition strategies that actively dismantle these host defenses. Computational frameworks such as SpatialMETA and spFBA have proven essential for bridging resolution mismatches between transcriptomic and metabolomic datasets, enabling predictive modeling of localized reaction rates and metabolic sink-source transitions. However, significant challenges remain, including tissue permeabilization optimization, compound identification limitations in untargeted metabolomics, and the static nature of current spatial technologies. Future advances must focus on achieving sub-micron spatial resolution for metabolomics, developing standardized protocols for infected plant tissues, and integrating live-cell biosensing with stable isotope tracing to capture dynamic nutrient fluxes in real time. Ultimately, spatial multi-omics integration will drive the rational design of crop varieties with engineered metabolic barriers, contributing to sustainable disease management in agricultural systems facing escalating pathogen pressures under climate change.

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