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In Brief

Arbuscules up close: spatiotemporal and single cell translatomics in rice and arbuscular mycorrhizal symbiosis

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Arbuscular mycorrhizal (AM) symbiosis is a widespread, ancient, mutualistic relationship between plants and fungi that supports nutrient acquisition in more than 80% of plant species (Brundrett and Tedersoo 2018; Sportes et al. 2021). Central to the plant-AM symbiotic relationship is the formation of arbuscules. Arbuscules are finely branched fungal structures that form inside root cortical cells and serve as sites for nutrient exchange (Fig. 1a; Luginbuehl and Oldroyd 2017). Although arbuscules are morphologically well described, their cellular heterogeneity and developmental dynamics have remained difficult to resolve because colonized individual roots contain a mosaic of fungal structures at different stages.

In recent work, **Chancellor and colleagues (Chancellor et al. 2026)** bring plant-AM symbiosis into sharp focus by combining dual-species spatial transcriptomics with AM stage-specific TRAP-*seq* (translating ribosome affinity purification RNA sequencing) (Fig. 1b, c) in rice (*Oryza sativa*) roots colonized by the AM *Rhizophagus irregularis*. Using a targeted spatial panel, the team profiled both plant and fungal transcripts directly in root sections at single-cell resolution, then layered on translating-ribosome capture to follow gene activity across early, middle, and late phases of the symbiosis and arbuscule development.

The spatial data confirmed that AM colonization rewires the root, and about 65% of genes exhibited differential expression between the AM colonized and noncolonized roots. Further, it was found that vesicles and intraradical hyphae displayed much higher transcript abundance for fungal genes such as *RiEF1α* than arbuscules did. This was consistent with microscopy results indicating that fungal nuclei were abundant in vesicles and hyphae but less so in arbuscule branches. Thus far, the team had challenged the assumption that arbuscules function as self-contained transcriptional units and therefore proposed that adjacent fungal compartments act as biosynthetic hubs that support the specialized functions of arbuscules.

The plant response was equally dynamic. Of the 15 informative rice cell-type markers, 73% were reduced in colonized roots, indicating that AM colonization reshapes cell identity at the transcriptional level. Even more strikingly, morphologically similar arbuscules and their respective host cells within the same root section displayed differential transcriptional profiles. Key arbuscule-associated genes, including phosphate- and nitrogen-related genes, along with lipid transfer genes varied markedly from cell to cell. By now, the message is clear: There is no single transcriptional signature of a "mature" arbuscule.

This conclusion was reinforced further by single-cell analyses. Segmentation of colonized roots produced more than 3,000 cells grouped into 6 clusters. However, these clusters did not map onto distinct developmental stages. The team observed that cells containing morphologically similar arbuscules could fall into different transcriptional states, underscoring how strong local context could shape symbiotic function. Spatial mapping revealed that nutrient transport is more nuanced than a simple on or off symbiotic switch. For instance, the gene *OsPT11* remained restricted to arbusculated cells, while other phosphate transporter genes displayed broad or shifted localization patterns. Further the gene *OsSWEET12* appeared redistributed into colonized cortical cells without a major change in total abundance.

To capture changes across the arbuscule's life cycle, the team used a stage-specific TRAP-*seq* driven by defined promoters active at early (*OsAM1*), middle (*OsPT11*), or late (*OsARK1*) stages in colonized cells. TRAP-*seq* revealed that the marker transcripts were significantly enriched in the matched TRAP fractions relative to the constitutive control. Further, the TRAP analyses exposed the scale of developmental reprogramming. Arbuscules in the early stage had 1,588 enriched and 4,041 depleted transcripts, the middle stage showed 1,652 enriched and 1,957 depleted transcripts, while the late stage had 1,402 enriched and 106 depleted transcripts. The overlaps among these gene sets mirrored arbuscule progression. Arbuscules in the early and middle stages shared the largest fraction of differentially captured

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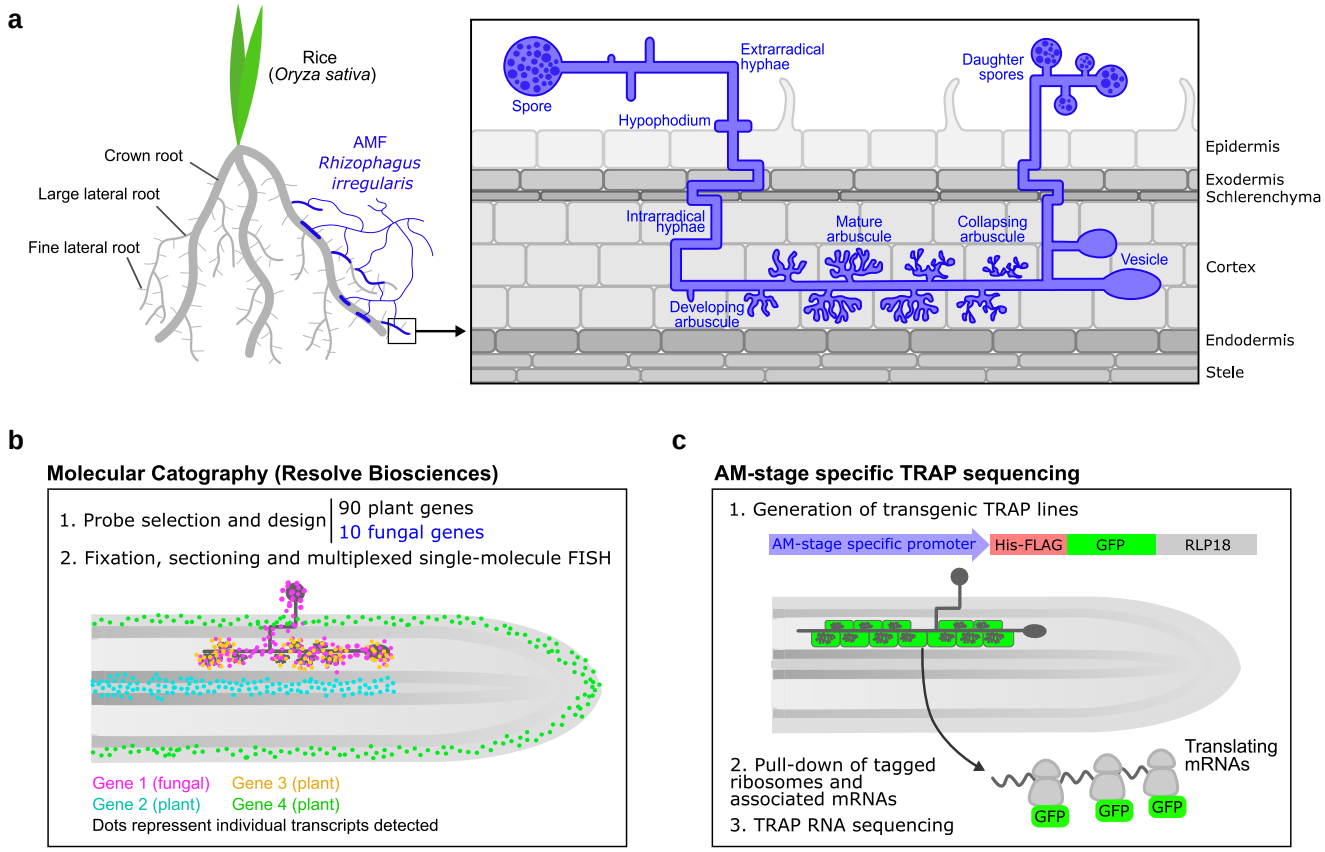

Figure 1 a) Simplified depiction of rice-arbuscular mycorrhizal symbiosis; macroscopic interaction (left) and microscopic fungal structures outside and within the root (right). Next, overview of high-resolution approaches: molecular cartography, illustrating gene-specific FISH signals (a); and TRAP-*seq* (c), using AM-stage promoters to profile ribosome-bound transcripts via tagged RLP18, (adapted from Chancellor et al. (2026) Fig. 1).

genes (34.7%), whereas overlap between early- and late-stage arbuscules dropped to about 9.7%. This highlights a substantial shift in cellular priorities over time. The early and middle stages were marked by suppression of defense and cell-wall-related pathways, consistent with fungal accommodation. By contrast, late-stage cells reactivated immune, oxidative, and cell wall-remodeling programs, matching the transition toward arbuscules degeneration.

Collectively, by integrating complementary approaches of spatial transcriptomics and AM-stage specific translatomics, the authors recast that arbuscules are not static exchange structures but are short lived, highly individualized interfaces shaped by local transcriptional and translational controls. This study strengthens our understanding of AM symbiosis in rice and establishes a powerful framework for dissecting how plant cells cooperate and exchange nutrients, as well as the eventual fungal withdrawal at one colonized cell at a time. As similar spatial and translatomic tools are applied across species and environmental contexts, the long-standing black box of symbiotic function may finally yield its most intimate secrets.

Recent related articles in *The Plant Cell*:

- Leng et al. (2023) demonstrated that arbuscular mycorrhiza (AM) induce the kinases (K) AMK8 and AMK24, which interact with receptor-like kinase gene KINASE3 to regulate AM symbiosis in *Lotus japonicas*.
- Araújo et al. (2025) examined whether the SPARK receptor-like kinase (RLK)/receptor-like cytoplasmic kinase (RLCK), known to be essential for arbuscular mycorrhiza (AM) formation in monocot and dicots, also functions in legume AM symbiosis. The authors showed that in the legume *Aeschynomene evenia*, the AeRLCK2 receptor contributes to nodulation but not required for AM symbiosis.

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Data availability

No new data were generated or analyzed in support of this research.

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