Plant Communications
Commentary

CellPress
Partner Journal

Proteomics-based multi-omics reveals m^{6}A-mediated regulation of the plant proteome

Proteins are fundamental to nearly all biological processes, making protein-level investigation essential for a deeper understanding of plant biology. Recent advances in mass spectrometry (MS)-based proteomics have enabled precise, high-throughput analyses of plant protein expression, modification, and interaction patterns (Sang et al., 2025). These developments are transforming our ability to dissect complex regulatory networks and are increasingly being integrated into multi-omics frameworks for gene discovery and trait analysis. Notably, two recent studies have produced the most comprehensive pan-tissue, proteomics-centered multi-omics atlases to date in rice and soybean—two of the world's most important crops—revealing a key role for N^6-methyladenosine (m^{6}A) in the post-transcriptional regulation of the plant proteome (Li et al., 2024; Yang et al., 2025).

A proteome view of plant biology

As the primary orchestrators of cellular function, proteins display only weakly positive genome-wide correlations with mRNA levels (Walley et al., 2016; Sang et al., 2025). This highlights the need to integrate proteomics with transcriptomics to more accurately capture cellular regulatory networks. MS-based proteomics has become a powerful tool for large-scale, high-resolution analysis of plant proteomes across diverse species (Sang et al., 2025).

Comprehensive proteomic landscapes have now been established in several plant species (Figure 1A), including *Arabidopsis thaliana* (Mergner et al., 2020), *Oryza sativa* (rice; Li et al., 2024), *Zea mays* (maize; Walley et al., 2016), *Glycine max* (soybean; Yang et al., 2025), *Triticum aestivum* (wheat; Zhang et al., 2025), *Pyrus bretschneideri* (pear; Wang et al., 2023), and *Medicago truncatula* (Marx et al., 2016). Despite these efforts, the number of identified proteins remains substantially lower than that of transcripts in all species examined. Several factors likely contribute to this discrepancy. First, MS-based workflows often group multi-copy proteins when unique peptides are unavailable for distinction. Second, signals from high-abundance proteins can mask those from low-abundance proteins. For instance, high-abundance proteins such as Rubisco constitute a dominant proportion of the protein pool in plant tissue proteomes, reducing detection sensitivity and overall proteome coverage, and potentially introducing bias into quantitative analyses. Third, limited sampling across tissues may restrict the detection of tissue-specific or low-abundance proteins. For example, the number and abundance of identified proteins vary considerably among tissues and developmental stages in *Arabidopsis*, rice, and soybean (Mergner et al., 2020; Li et al., 2024; Yang et al., 2025). While such variation reflects genuine tissue-specific biological functions, it may also arise from technical variation or sampling limitations, underscoring the need for broader sampling to ensure comprehensive proteome representation. In addition, proteomic datasets are highly sensitive to differences in sample preparation, data acquisition, and computational pipelines, making cross-study integration and meta-analysis particularly challenging.

Despite technical limitations, two recent pan-tissue, proteomics-centered multi-omics studies (Figure 1B) have quantified 15 174 and 12 855 proteins across 14 tissues in rice and soybean, respectively (Li et al., 2024; Yang et al., 2025). These represent the most comprehensive proteomic datasets reported for these species and provide valuable resources for examining correlations between mRNA and –protein expression. In both crops, most genes lacking detectable protein evidence are expressed at low transcript levels, indicating that genes with higher mRNA abundance are more likely to be translated. Notably, as observed in humans, many genes in rice and soybean show weak or insignificant correlations between protein and RNA levels. Among those with significant correlations, most display positive associations, whereas only a small fraction show negative correlations (Jiang et al., 2020; Li et al., 2024; Yang et al., 2025) (Figure 1C). This discordance between transcript and protein abundance is particularly evident in soybean, where approximately 72.5% of detected proteins show insignificant correlations, compared with about 50% in rice and humans. Moreover, mRNA–protein correlations vary considerably across tissues within both species (Li et al., 2024; Yang et al., 2025). Together, these findings suggest that the regulation of protein abundance differs markedly across tissues and species, reflecting the combined influences of post-transcriptional regulation, translation efficiency, transcript stability, and protein turnover. This highlights the importance of extending proteomic analyses across a wider range of plant species to better capture this regulatory diversity.

m^{6}A-mediated post-transcriptional regulation of the plant proteome

The steady-state abundance of a protein is governed by multiple regulatory layers, including transcriptional, post-transcriptional, translational, and post-translational processes. Among these, m^{6}A—the most prevalent internal modification of mRNA—has emerged as a key regulator of post-transcriptional control (Shen et al., 2023; Zhang et al., 2024). To investigate the causes of discordance between RNA and protein levels, pan-tissue maps of the m^{6}A methylome were generated using m^{6}A-seq2 in rice and soybean (Li et al., 2024; Yang et al., 2025). These analyses revealed a prominent role for m^{6}A in determining protein abundance in both species (Figure 1C). Totals of 270 375 and 304 638 m^{6}A peaks were identified in rice and soybean, respectively—numbers which, on average, are comparable to those detected across *Arabidopsis* tissues (Zhang et al., 2024)

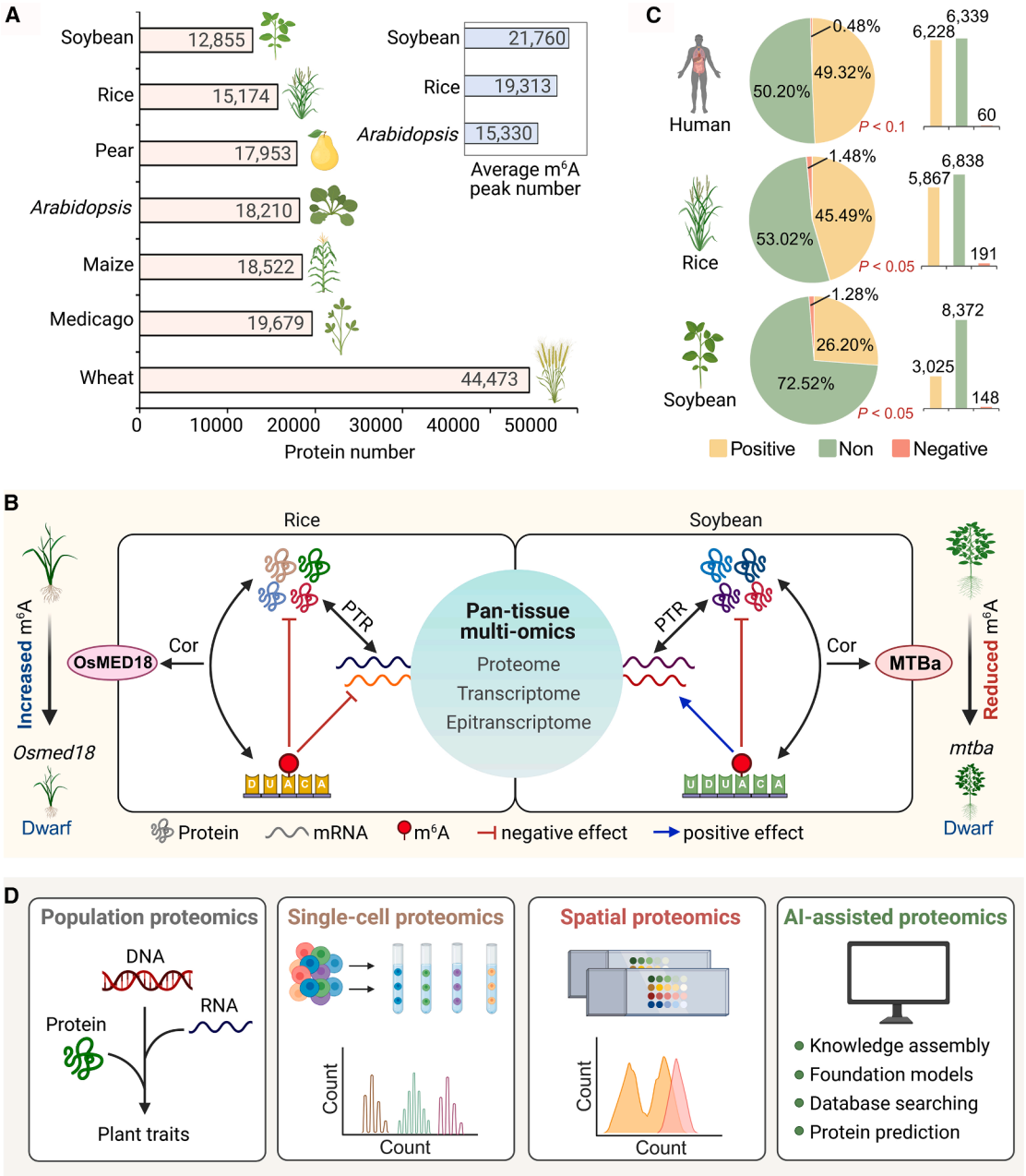

Figure 1. A mass spectrometry (MS)-based proteome view of plants.
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(A) Summary of identified protein numbers across various species, including *Arabidopsis*, rice, maize, soybean, wheat, pear, and *Medicago truncatula*. The inset shows the average number of m^{6}A peaks detected across different tissues in *Arabidopsis*, soybean, and rice.
(B) Systematic multi-omics atlases integrating the proteome, transcriptome, and m^{6}A epitranscriptome in rice and soybean. In both species, m^{6}A exhibits a repressive effect on protein abundance. In rice, m^{6}A levels are negatively correlated with mRNA abundance, whereas a positive correlation is observed in soybean. Correlation analyses between the proteome and the m^{6}A epitranscriptome identified novel m^{6}A regulators in both species. Abbreviations: Cor, correlation; PTR, protein-to-RNA ratio.
(C) Discordance between RNA and protein levels in humans, rice, and soybean. Pie charts indicate the proportion of each classification, whereas bar charts display the corresponding counts. Categories: Negative, negative correlation; positive, positive correlation; non, no correction.
(D) Potential future research directions in MS-based plant proteomics. Schematics depict conceptual distributions of protein abundance at single-cell and spatial levels (axes are not to scale).

(Figure 1A). The enriched sequence motifs DUACA (D = A/G/U) in rice and UDUACA in soybean suggest conserved m^{6}A features, consistent with the canonical RRACH motif (R = G/A and H = A/U/C) previously reported in *Arabidopsis* (Zhang et al., 2024).

Notably, the correlation between m^{6}A modification and mRNA abundance displays opposite trends in rice and soybean (Li et al., 2024; Yang et al., 2025). In rice, m^{6}A levels are negatively correlated with transcript abundance, whereas in

soybean, the correlation is positive (Figure 1B). Despite these contrasting patterns, genes bearing m^6A modifications consistently exhibit lower protein-to-RNA ratios than unmodified genes across tissues in both species, indicating a translationally repressive role of m^6A (Li et al., 2024; Yang et al., 2025). Supporting this conclusion, luciferase (LUC) transcripts with m^6A in rice protoplasts exhibit significantly higher RNA levels but similar protein output compared with unmethylated controls or those co-expressed with the Fat mass and obesity–associated protein (FTO), a known m^6A demethylase (Li et al., 2024). Furthermore, among factors such as RNA abundance, m^6A modification, codon usage, gene length, and exon number, m^6A ranks as the second- and third-strongest contributor to protein abundance in rice and soybean, respectively (Li et al., 2024; Yang et al., 2025), highlighting its importance in regulating protein abundance. Although m^6A generally represses protein abundance in both rice and soybean, tissue-conserved m^6A has been shown to promote mRNA translation in *Arabidopsis* (Zhang et al., 2024), suggesting that its function may vary across species and contexts. Additional studies will be required to clarify these differences. It is also worth noting that while these analyses explore mRNA–protein relationships, the presence of multi-copy genes remains a technical challenge, especially in polyploid plants, in which distinguishing homologous transcripts and proteins is difficult.

Despite the observed correlations between m^6A and translation, the precise mechanisms through which m^6A modulates protein synthesis remain poorly understood in plants. In *Arabidopsis*, the m^6A reader EVOLUTIONARILY CONSERVED C-TERMINAL REGION 8 selectively sequesters m^6A-modified mRNAs into phase-separated cytosolic condensates, providing temporary storage that may help maintain a translation-ready mRNA pool in the cytoplasm (Wu et al., 2024). Similarly, in tomato, the m^6A reader SlYTH2 forms phase-separated cytosolic condensates with its m^6A-modified targets and translational regulators, thereby modulating protein synthesis (Bian et al., 2024). Moreover, m^6A has been reported to exert both positive and negative effects on translation of specific transcripts, highlighting its intricate and context-dependent regulatory roles on translation (Shen et al., 2023). Comprehensive proteomic analyses targeting diverse m^6A writers, erasers, and readers will be crucial to further elucidate the multifaceted roles of m^6A in shaping the plant proteome.

Integrating proteomic and m^6A landscapes in gene discovery

Correlation analyses between m^6A levels and protein abundance have led to the identification of potential genes involved in m^6A regulation, such as *OsMED18* in rice and *MTBa* in soybean (Li et al., 2024; Yang et al., 2025) (Figure 1B). Loss-of-function mutants of these newly identified candidates further confirm their regulatory roles in m^6A modification and their contribution to plant growth and development (Figure 1B). In rice, the mediator complex protein OsMED18 exhibits a negative association with m^6A levels across tissues. Mutants lacking *OsMED18* display shorter roots and shoots along with significantly elevated m^6A levels compared with the wild type (Li et al., 2024). In soybean, the putative m^6A writer MTBa exhibits a positive correlation between protein abundance and m^6A levels across organs. Mutants of *MTBa* show dwarf phenotypes accompanied by greatly reduced m^6A levels (Yang et al., 2025). Together, these findings highlight the power of proteome-integrated multi-omics approaches for identifying functional regulators like m^6A-related genes, providing valuable targets for further exploration of post-transcriptional control mechanisms in plants.

Future research directions for proteomics-based studies

Proteome-integrated multi-omics approaches have strong potential for identifying novel regulatory genes and providing valuable insights for crop breeding. However, comprehensive proteomic studies in plants remain limited compared with transcriptomic and genomic efforts, highlighting the need for broader and deeper exploration of the plant proteome to fully harness its potential for crop improvement. Ideally, proteomics-based multi-omics studies should generate paired proteomic, transcriptomic, m^6A-seq, and other omics data from the same biological samples, as demonstrated in recent works on rice and soybean (Li et al., 2024; Yang et al., 2025). Future research in plant proteomics must also overcome key technical challenges. The high abundance of proteins such as Rubisco, together with the presence of complex cell wall components, can obscure the detection of low-abundance regulatory proteins, representing a major bottleneck in plant proteomics (Sang et al., 2025). With continued advances in MS, plant proteomics is poised to address central questions in plant biology (Figure 1D). Future directions include (i) population proteomics, which can be integrated with genome- or transcriptome-wide association studies to link genetic variation to phenotypic traits by functional protein differences; (ii) single-cell proteomics, enabling cellular-level resolution of protein expression that extends beyond transcript-level insights; (iii) spatial proteomics, which retains the tissue context while mapping protein distributions, providing insights into tissue organization and intercellular interactions; and (iv) AI-assisted proteomics, where AI can analyze complex datasets, predict protein functions, and model protein dynamics—paving the way for virtual plant cell simulations. Additional areas, such as post-translational modification proteomics, protein–protein and protein–ligand interaction studies, and protein turnover analysis, also hold strong promise for deepening our understanding of plant molecular systems. Collectively, these approaches are expected to transform proteomic knowledge into powerful tools for accelerating crop improvement.

FUNDING

This work was supported by the National Research Foundation Competitive Research Programme (NRF-CRP22-2019-0001) and by intramural research funding from the Temasek Life Sciences Laboratory.

ACKNOWLEDGMENTS

No conflict of interest declared.

Received: September 7, 2025
Revised: October 2, 2025
Accepted: October 29, 2025
Published: November 1, 2025

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https://doi.org/10.1016/j.xplc.2025.101583

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