

## A Multi-omic Atlas of the Model Legume *Medicago truncatula*

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Legumes develop a complex symbiotic association with soil bacteria named rhizobia. This symbiosis culminates in the development of root nodules inside which the rhizobia fix atmospheric nitrogen and transfer it to their plant host. Over the last decades, a very close relative of alfalfa, *Medicago truncatula* has emerged as a key model legume to study associations with rhizobia but also many biological questions pertaining to legume biology. We developed a large-scale, quantitative proteomic, phospho-proteomic, and acetyl-proteomic study to document the global expression in 6 major tissues (stem, leaf, flower, buds, roots, and nodules) of *Medicago truncatula*. Included in this comparison are nodule tissues that were harvested at 10, 14, and 28 days post rhizobial inoculation, permitting the assessment of the dynamic protein and post-translational modification (PTM) level changes that accompany the progress in nodule development. This time-course data not only encompass the proteins within *Medicago* nodules, but also the bacterial proteins, for which we identified and quantified over 3,000 proteins, 140 phosphorylation sites, and 60 acetylation events. Combined with *Medicago*-specific identifications, this study quantified more than 20,000 proteins, more than 21,000 sites of phosphorylation, and more than 500 sites of acetylation within the two species. The 10 million MS/MS spectra that were collected throughout our tissue-specific analyses were searched against a consensus database comprising 4 *Medicago* protein sequence repositories (UniProt, Ensembl, RefSeq, and JCVI), independent *Medicago* gene predictions (compiled in-house using Augustus), and a rhizobial protein database (UniProt). This rich dataset includes uncharacterized and novel proteins, allowing us to assess and cluster tissue-specific proteins, PTMs, and PTM motifs, enabling the elucidation of tissue-specific protein functionality. We have included the gene atlas expression data (Benedito *et al.* 2008) to generate a multi-omics picture of *Medicago*. Collectively, these data comprise the most holistic proteomic analysis of a legume to date, an incomparable resource for the study of symbiotic nitrogen fixation and legume biology.