



A regulatory network-based approach dissects late maturation processes related to the acquisition of desiccation tolerance and longevity of *Medicago truncatula* seeds

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In seeds, desiccation tolerance (DT) and the ability to survive the dry state for prolonged periods of time (longevity) are two essential traits for seed quality that are consecutively acquired during maturation. Using transcriptomic and metabolomic profiling together with a conditional-dependent network of global transcription interactions, we dissected the maturation events from the end of seed filling to final maturation drying during the last 3 weeks of seed development in *Medicago truncatula*. The network revealed distinct coexpression modules related to the acquisition of DT, longevity, and pod abscission. The acquisition of DT and dormancy module was associated with abiotic stress response genes, including late embryogenesis abundant (LEA) genes. The longevity module was enriched in genes involved in RNA processing and translation. Concomitantly, LEA polypeptides accumulated, displaying an 18-d delayed accumulation compared with transcripts. During maturation, gulose and stachyose levels increased and correlated with longevity. A seed-specific network identified known and putative transcriptional regulators of DT, including ABSCISIC ACID-INSENSITIVE3 (*MtABI3*), *MtABI4*, *MtABI5*, and APETALA2/ ETHYLENE RESPONSE ELEMENT BINDING PROTEIN (*AtAP2/EREBP*) transcription factor as major hubs. These transcriptional activators were highly connected to LEA genes. Longevity genes were highly connected to two *MtAP2/EREBP* and two basic leucine zipper transcription factors. A heat shock factor was found at the transition of DT and longevity modules, connecting to both gene sets. Gain- and loss-of-function approaches of *MtABI3* confirmed 80% of its predicted targets, thereby experimentally validating the network. This study captures the coordinated regulation of seed maturation and identifies distinct regulatory networks underlying the preparation for the dry and quiescent states.

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