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The search of somatic embryogenesis regulators in *Medicago truncatula* among the MtNF-Y family

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Motivation and aim: Somatic embryogenesis (SE) is a regeneration technique whereby plants form bipolar embryo-like structures from somatic cells that are capable of developing into new plants. SE is of great importance for plant biotechnology, where it is used to obtain artificial seeds, as a model in fundamental research, and in plant transformation as a way of regeneration. For most leguminous plants, SE initiation protocols have not been developed and regeneration as well as transformation is challenging. Initiation of SE is possible through the utilisation of morphogenetic regulators. NF-Y are conserved eukaryotic transcription factors consisting of three subunits, NF-YA, B and C. In plants, each subunit is encoded by a gene family, and combinations of different subunits can regulate different processes in plant development. The aim of our study is to identify such regulators among transcription factors NF-Y involved in legumes embryo development.
Materials and methods: We used *Medicago truncatula* as a model species and detected *MtNF-Y* genes with high expression levels during SE by qPCR analysis and analyzing transcriptomic databases. We also utilized yeast two-hybrid system to search for proteins among MtNF-YA, MtNF-YB, MtNF-YC families capable of forming protein complexes. We also used Agrobacterium transformation and *in vitro* cultivation of plant explants to evaluate SE capacity in plants with changed functions of NF-Y genes. CRISPR/Cas9 technology was implemented to obtain edited plants with gene loss of function.
Results: We identified several proteins from the MtNF-Y family that are able to form complexes during SE. Coexpression of genes from two such complexes initiates SE in a non-embryogenic *M. truncatula* line. Overexpression of one of the *NF-YA* genes increases callus mass. Plants with overexpression of a gene from the *NF-YB* family demonstrate accelerated somatic embryo development, while edited plants with loss of function of this gene have delayed embryo development.
Conclusions: Data on *NF-Y* genes involved in SE can be used to evaluate gene coexpression effects on the regeneration process in *M. truncatula* of the remaining MtNF-Y complexes and validate them as morphogenetic regulators in other legumes.
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