

tection of Ogu-INRA *cms* gene and two for detection of *Rfo* gene, was tested. Both markers used for detection of Ogu-INRA *cms* gene produced similar and comparable results, as was the case with two markers, Bn-RFO-AS2 and BolJon, used for detection of *Rfo* gene.

Keywords: *Brassica napus* L., marker, Ogu-INRA *cms* system

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Study of chicory germacrene A synthase function in guaianolide biosynthesis through amiRNA-induced gene silencing

PP1-3

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Germacrene A synthase (GAS) catalyzes an important step in guaianolide biosynthesis converting farnesyl pyrophosphate to germacrene A – universal sesquiterpene precursor in chicory. The function of two genes coding for GAS – *CiGASlo* and *CiGASsh*, was studied by gene silencing. Chicory was transformed with amiRNA constructs designed to specifically silence either of the two genes, and regenerated transgenic plants were tested for changes in gene expression and sesquiterpene lactones content. Silencing was successful in most of the clones, which had reduced gene expression. However, the amiRNA constructs were not specific for *CiGASlo* or *CiGASsh*, so that both genes were silenced in most transgenic plants. To verify that the gene silencing had impact on sesquiterpene lactone levels, three representative guaianolide oxalates (lactucin-15-oxalate, 8-deoxilactucin-15-oxalate and lactucopicrin-15-oxalate) were identified and quantified by UPLC-MS/MS in roots and shoots of transgenic and control plants. Data analyses showed unequal segregation of targeted compounds in the plant tissues, and implied that the synthesis of these compounds highly depends on the expression of GAS genes in the shoots and that these metabolites are pooled in the shoots, while the root guaianolide oxalates content is regulated on a level different from GAS expression. Nevertheless gene silencing reduced the targeted metabolites levels significantly. It was concluded that both genes are involved in guaianolide biosynthesis, and that their complex pattern of expression in different parts of the plant determines the amount of guaianolides and their derivatives.

Keywords: germacrene A synthase, silencing, amiRNA, chicory, guaianolides

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