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ESTABLISHMENT OF A PROTOCOL FOR TRANSIENT TRANSORMATION OF ARTEMISIA ANNUA PLANTS

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Artemisia annua is an herbaceous plant belonging to the *Artemisia* genus, one of the largest and most widely diffused genus of the family Asteraceae (Compositae). Also known as sweet wormwood, *Artemisa annua* plants have been used for more than 2000 years in traditional Chinese medicine thanks to its beneficial properties as an antipyretic, antiseptic, antispasmodic, carminative, stimulant, tonic, and stomachic. In modern times, *Artemisia annua* earned great importance due to its strong antimalarial activity. Artemisinin, a sesquiterpene lactone, is the bioactive compound responsible for this action against malarial disease and it has been isolated from *Artemisia annua* leaves. Nowadays artemisinin-based combination therapies have been recommended from WHO as treatments of malarial disease. Unfortunately, artemisinin yield from plants is rather low, ranging from 0,01% to 1,5 %. Because of this, many different biotechnological approaches have been tried to increase Artemisinin yield, from genetic engineering in plants to cell cultures. In order to develop a metabolic engineering approach in *Artemisia annua* plants to boost artemisinin production, we set up a protocol for transient transformation. Transient expression, performed via *Agrobacterium tumefaciens* infiltration, allows us to investigate in a short time the effect of the manipulation of the genes involved in the biosynthetic pathway of artemisinin. We decided to use whole plants for agroinfiltration. Plants of *A. annua* variety #20931, with high artemisinin content from Thailand, have been used in these experiments. In order to evaluate the efficiency of the agroinfiltration we used the GFP (green fluorescent protein), a reporter gene. Infiltrations have been performed with the *Agrobacterium* strain EHA105, which is known to be the most effective for *A. annua* transformation. Different parameters as plant age, growth condition (in vivo or in vitro plants), environmental conditions (greenhouse or growth chamber) and the OD of the agrobacterium solution have been evaluated. A preliminary analysis of the results has been carried out with microscopy techniques and western blot analysis.