

GENETIC MANIPULATION OF THE NOVEL CROP WOLFFIA GLOBOSA (LEMNACEAE)

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Wolffia globosa belongs to Lemnaceae family (duckweed), the smallest aquatic monocotyledonous plants that include five genera (*Spirodela*, *Lemna*, *Wolffiella*, and *Wolffia*) with diverse morphologies and living habits. These plants primarily propagate through vegetative reproduction. *W. globosa* has garnered interest due to its economic value and potential to mitigate resource constraints and environmental issues. It is characterized by rapid growth rate, ease of cultivation, direct contact with water, and adaptability to environmental changes. In addition it has favourable qualitative and quantitative nutritional profiles with an average dry weight/fresh weight ratio of 4%; total protein content of 20%-40%, starch content of 10-20%, fat content of 1-5%, and fiber content of approximately 25%. Notably, the essential amino acid content meets the requirements of preschool-aged children according to standards of the World Health Organization. The macro- and microelements (minerals) contents not only depends on the cultivation conditions but also on the genetic background of the species. Furthermore, *W. globosa* do not show detectable anti-proliferative or cytotoxic effects, and is consumed as a food source in several Asian countries. In addition, *W. globosa* has shown a high relative growth rate even under microgravity, making it suitable to be used in space as astronauts food. Due its high biomass and starch accumulation and low lignin content, *W. globosa* is adequate as bioenergy feedstock to produce bioethanol and biogas. Transgenic *W. globosa* lines expressing a protective edible vaccine antigen against fish vibriosis has been obtained, which indicated that this species could serve as a bioreactor to produce edible vaccines and many other high value-added molecules like antibodies, pharmaceutical proteins, and industrial enzymes. Our goal was to establish

an alternative genetic transformation procedure that did not rely on regeneration events but instead genetic modified meristematic cells. This approach was primarily due to the fact that obtaining transgenic plants in this species takes between 4-6 months. Therefore, we carried out indirect genetic manipulation by co-cultivating plants with *Agrobacterium tumefaciens* using the LB4404 strain carrying the 35S-GUS-INT binary vector. The transformation frequency was approximately 0.2% of the total co-cultured fronds. Further experiments are underway to improve transformation efficiency to overproducing high-value metabolite as anthocyanins also by genome editing