World Academy of Science, Engineering and Technology
International Journal of Agricultural and Biosystems Engineering
Vol:19, No:06, 2025

Function of GIGANTEA Genes in the Commercial Potato Cultivar ‘Désirée’

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Abstract : GIGANTEA (GI) is a plant-specific, circadian clock-regulated, nuclear protein involved in diverse processes from flowering to stress responses. In the obligate short-day tuberising Andigenum Group potatoes, GI is indirectly involved in determination of the time of tuber initiation. The goal of our study was to get information on the function of GI in the day-length independent tuberising commercial potato cultivar ‘Désirée’, a tetraploid plant carrying two GI genes, one on chromosome 4 (GI.04) and another one on chromosome 12 (GI.12). Functional analysis of the two GI genes was attempted by targeted mutagenesis using the CRISPR-Cas9 system. Two sets of mutants were generated. The mutations were mapped at nucleotide level and the plants grown in a greenhouse. GI is located in the nucleus and interacts with at least five proteins. Three out of them, two photoreceptors and FKF1, bind on GI close to the nuclear localisation (NLS) signal to the so called LOV domain. In Andigenum Group potatoes, FKF1 interacts not only with GI but form a triplex with CDF1, a positive regulator of tuberisation, and transport it to proteasomes, where CDF1 is degraded. Three GI.04 and three GI.12 null mutants were selected from the first set of mutagenesis. Although, the deletions did not reach the NLS and LOV domain in any of the six mutants all GI. 04 and two GI.12 mutants were shorter than the control suggesting that both GIs are involved in vegetative growth regulation and the deleted region might be important in terms of conformation or stability of the proteins. From the second set of mutagenesis, three null mutants carrying mutations in one of the two GI genes and three mutants carrying mutations in both GI genes were selected for detailed analysis. Deletions in the GI mutants of this set disrupted the NLS and extended to the LOV domain. Nevertheless, none of the single GI gene mutations influenced the time of tuberisation or the tuber number and yield, whereas one of the GI.04 and all GI.12 mutants were shorter than the ‘Désirée’ control. Furthermore, all GI.12 mutants showed early senescence. The early senescence of mutants carrying mutations in both GI genes was even more pronounced, resulting in substantial yield loss in one of the double mutants. These results raise the possibility that the two GI genes can substitute each other in term of tuberisation or they are not involved in it, the yield loss is due to the early death of the plants. To distinguish between the two possibilities yeast two-hybrid experiments were initiated to detect the interaction between the GI proteins and FKF1 and between FKF1 and CDF1 originated from ‘Désirée’.

Keywords : gene editing, tuberisation, senescence, Solanum tuberosum
Conference Title : ICPB 2025 : International Conference on Plant Biology
Conference Location : Vienna, Austria
Conference Dates : June 19-20, 2025