“Molecular farming” for production of nutraceuticals and pharmaceuticals using technology that builds public confidence

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Most pharmaceuticals and nutraceuticals necessary for preventing lifestyle-related illness and fostering longevity and health are derived from plants and herbs, to which serious attention is currently paid. Genetic engineering of plants may improve plants to allow them to accumulate more functional substances in a shorter period than by ordinary plant breeding. Strategies employed for the production of genetically modified (GM) crops are premised on (1) the avoidance of gene transfer in the field, (2) the use of genes derived from edible organisms such as plants, and (3) preventing the appearance of herbicide-resistant weeds. To this end, we developed a novel vector system fused a vector for chloroplast transformation with acetolactate synthase (*ALS*) and *aadA* excision system following transformation.

We generated a series of mutated ALS (mALS) genes and introduced constructs with *mALS* and the aminoglycoside 3’-adenyltransferase gene (*aadA*) into the tobacco chloroplast genome. We have so far reported that plants harboring transplastomic mALSs of G121A, A122V, P197S and W574L/S653I are specifically tolerant to PC, IM, SU/PC and all ALS herbicides, respectively. The task that *aadA* should be excised from chloroplast genome in the transplastomic plants remains with these vectors. Therefore, we improved an original vector for chloroplast transformation by introducing a 400-bp *rbcL* fragment between *aadA* and *mALS*. The transplastomic lines with this vector generated variegation during growth with spectinomycin for 2 weeks after growth in the absence of antibiotic. We confirmed the excision of *aadA* from cells in white variegation parts of the transplastomic lines. This novel vector system is the first trial that satisfies all required premises of transformation technology for GM crops.