

研究论文

农杆菌介导的马铃薯试管薯遗传转化体系的优化及反义class I patatin基因的导入

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摘要 用两个马铃薯栽培品种"鄂马铃薯3号"和"甘农薯2号"的试管薯为供体材料,建立了一种农杆菌介导的简单、快速和高效的遗传转化系统.在含有75 mg/L卡那霉素的选择培养基上,2~3周可产生抗性芽,4~5周获得完整的转基因植株.筛选出了试管薯遗传转化的优化条件,特别是在再生培养基中加入2 mg/L玉米素,两个品种的转化频率分别高达45.5%和43.9%.周期短(4~5周)、一步培养和转化频率高,使该转化体系能够广泛用于马铃薯转基因的研究.用含有反义class I patatin基因的表达载体pBSAP转化两个品种,共获得120株卡那霉素抗性植株.PCR、PCR-Southern和Northern杂交分析证明,此反义基因已整合到马铃薯基因组中并在转基因植株中正常转录.反义基因的表达导致部分转基因植株的试管结薯株率和单株结薯数降低.结果表明,该class I patatin基因可能参与了块茎形成的调控.

关键词 [马铃薯](#) [根癌农杆菌](#) [转化](#) [试管薯](#) [反义patatin基因](#)

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An Efficient Protocol for Agrobacterium-mediated Transformation with Microtuber and the Introduction of an Antisense class I patatin Gene into Potato

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Abstract A simple, rapid and high efficient protocol was established for Agrobacterium-mediated transformation with microtuber of two potato cultivars, E-potato 3 and Gannongshu 2. Adventitious buds differentiation occurred after 2-3 weeks on selective medium containing 75 mg/L kanamycin, and transgenic plants could be obtained in 4-5 weeks. By combining the best treatments, especially using 2 mg/L zeatin riboside (ZR) in the shoot induction medium, this protocol yielded transformation efficiency of 45.5% and 43.9% from E-potato 3 and Gannongshu 2, respectively. Short duration (4-5 weeks), one-step culture and high efficiency made this system well suited for wider production of transgenic potato plants. In total 120 positive plants with root on kanamycin-containing medium were obtained using the vector pBSAP carrying an antisense class I patatin gene. Transformants were confirmed by PCR, PCR-Southern blot and Northern hybridization analysis. Expression of the antisense class I patatin gene resulted in a significant reduction of tubers production in transgenic plants. The results suggested that the class I patatin gene was possibly involved in regulating tuber formation.

Key words [Potato](#) [Agrobacterium tumefaciens](#) [Transformation](#) [Microtuber](#) [Antisense patatin gene](#)

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