Development of a multiple gene knockout system using mobile group Ⅱ intron in *Clostridium acetobutylicum*

조창희, 장유신, 이종민, 이상엽†
한국과학기술원
(leesy@kaist.ac.kr†)

Clostridium acetobutylicum is one of the industrial microorganisms for the efficient production of various chemicals. However, due to the lack of efficient genetic manipulation tools, strain improvement has been rather slow. Fortunately, mobile group Ⅱ intron was successfully applied to gene knockout of *Clostridium* species. Since the knockout system based on the mobile group Ⅱ intron was constructed in a replicable plasmid, curing of the plasmid was required prior to the disruption of the next gene. We developed a multiple gene-knockout system that does not require marker pop-out process by using a mobile group Ⅱ intron *Ll.ltrB* and two different antibiotics markers. By using this strategy, a quintuple knockout mutant has been developed, recently. [Development of Systems Metabolic Engineering for Biorefineries from the Ministry of Science, ICT and Future Planning (MSIP) through the National Research Foundation (NRF) of Korea (NRF-2012-C1AAA001-2012M1A2A2026556); and the Advanced Biomass R&D Center (ABC) of Global Frontier Project funded by the Ministry of Science, ICT and Future Planning (ABC-2010-0029799).]