A novel approach for high quality plasmid DNA isolation from E. coli through mechanical cell-lysis at elevated temperature

Muhammad Wajid Ullah, Waleed Ahmad Khattak, Mazhar-ul-Islam, 유보완, 조호정, 박중 곤* 경북대학교 (parkjk@knu.ac.kr*)

The circular double stranded plasmid DNA is used as a molecular cloning vector that functions to drive the replication of recombinant DNA sequences within host organism. Currently, different approaches are used for the isolation of plasmid DNA that uses alkaline cell-lysis, but the procedures are often laborious and time consuming. Also the obtained plasmid DNA is mostly contaminated with chromosomal DNA. We have successfully isolated pure plasmid DNA from E. coli by a simple and rapid isolation method at higher alkaline pH and elevated temperature that involves the mechanical cell-lysis through bead-beating to release the plasmid DNA. The efficiency of bead-beating method was evaluated through CFU and microscopic analysis and the quantity and quality of isolated DNA was checked through spectrophotometer and agarose gel electrophoresis, respectively. The obtained plasmid DNA is of good quality and free from chromosomal DNA contamination. This novel approach of plasmid isolation can be used on a preparative scale to isolate sufficient quantities and good quality of plasmid DNA.