Magnetic beads-based DNA extraction: different techniques compatible with PCR and microfluidic systems

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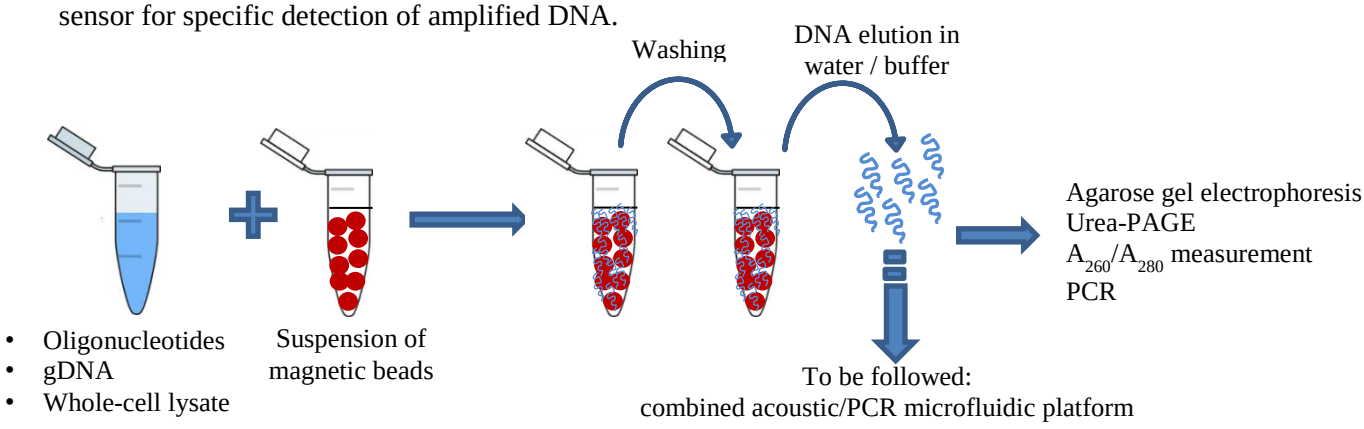
Different approaches for DNA extraction with the use of magnetic beads were investigated. Due to the subsequent application, the main effort was made on the methodologies which are easily transferred to the microscale format and which are compatible with the PCR reaction. Fast DNA adsorption and elution together with efficient extraction was also demanded. Such approaches typically rely on solid-phase extraction (SPE) methods based on the "bind-wash-elute protocols". The most widely applied solid supports for SPE of DNA are silica based[1]. Then, DNA adsorption on positively charged surfaces [(2)] or solid-phase reverse immobilization under the presence of PEG [(3)] is often used as well.

In this study, magnetic particles with different terminal functional groups (silanol, carboxyl and amine) and with size between 1 and 3 µm served as a solid phase for the aforementioned SPE methods. The DNA extraction efficiency from three types of materials differing in the complexity – 19 bases long oligonucleotide with Cy3 dye or gDNA of *Salmonella enterica* serotype Typhimurium pre-isolated or from whole-cell lysate – was monitored and compared. Moreover, specific isolation through the hybridization between the oligonucleotides immobilized on the beads and complementary sequences of the isolated DNA was performed as well. UV/VIS measurement, agarose electrophoresis, urea-PAGE and PCR were utilized for quantitative and qualitative evaluation of eluted DNA. Extraction efficiency and elution volume were another important evaluation criteria. It was confirmed that the suitability of the chosen method and particular protocol depends on the length of DNA to be isolated. The advantages and disadvantages of all methods were discussed. In future, the on-chip extraction is going to be transferred into microscale format with integrated micro-PCR module and sensor for specific detection of amplified DNA.



General scheme of methods based on the principle of solid-phase extraction used in this study for bacterial DNA extraction and purification.

References:

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