

Partition of plasmid DNA in polymer-salt aqueous two phase systems

ABSTRACT

The partition of plasmid DNA (pDNA) in polyethylene glycol (PEG)–phosphate aqueous two-phase systems (ATPS) is presented. A high molecular weight (HMW) and a low molecular weight (LMW) polymer, PEG-1450 and -300, were used in combination with dipotassium hydrogen phosphate. The experimental results demonstrated that the plasmid pTX0161 displays a varied partition behaviour in PEG–phosphate ATPS. In HMW PEG (PEG-1450–phosphate systems), pDNA partitioned to the bottom phase only. In LMW PEG (PEG-300–phosphate systems), pDNA partitioned to all of the phases with respect to the phase composition, system temperature and concentration of lysate used in the ATPS. In systems with volume ratios higher than one, pDNA was mainly recovered in the top phase. For volume ratios between 0.5 and 1, pDNA mainly partitioned to the interface. In systems with volume ratios below 0.5, most of the pDNA was recovered in the bottom phase. For temperatures between 4 and 25°C, the partition to the top phase decreased whereas partition to the interface steadily increased. At 25°C, over 80% of pDNA was recovered in the interface. The partition to the bottom phase increased steadily with increasing temperatures up to 40°C and the partition to the interface decreased. At 20°C, the recovery of pDNA in the interface gradually increased and reached a maximum at 60% (w/w) lysate with 80% recovery recorded. At 25°C, over 80% of pDNA was recovered in the interface from lysate concentrations greater than 35% (w/w). At 30°C, the top phase preference changed to an interface preference between 0 and 20% (w/w) lysate.

Keyword: Partition behaviour; Plasmid DNA (pDNA); Aqueous two-phase systems (ATPS); High molecular weight (HMW); Low molecular weight (LMW).