In vitro protein synthesis in cell-mimetic PNIPAM hydrogels

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The gene expression is the most important and essential reaction in living organisms to survive. To gain a deeper understanding of the basic biochemical reactions in cells, in vitro translation (IVT) has been investigated to mimic the cell. However, conventional studies on IVT are usually performed in dilute, homogeneous bulk solutions which do not reflect the cell-sized confinement. Therefore, the protein expression inevitably differs between in vitro and in vivo. To resolve this issue, the droplet-based microfluidics was employed to reflect the cell-like environment properly. We fabricated DNA containing poly(N-isopropylacrylamide)(PNIPAM) hydrogel in a microfluidic device. The PNIPAM shell reflected in vivo environment. By co-encapsulating DNA and E. coli lysates in NIPAM droplets, we studied protein expression in a defined reaction volume and various DNA concentrations. This proposed platform here can be utilized not only to study the effects of volume on protein expression but also to determine the practical DNA concentration for the protein synthesis in cell-mimetic environments.