Expression of functional aglycosylated full-length antibody in a cell-free protein synthesis system derived from *Escherichia coli*

오린석, 김동명[1,*]
서울대학교; [1]충남대학교 공과대학 정밀공업화학과
(dmkim@cnu.ac.kr*)

In this study, we attempted to produce functional full-length antibodies using E.coli cell-free protein synthesis system. Proper folded immunoglobulin G (IgG) required correct intermolecular disulfide bonds between two heavy chains and two light chains, parts of the IgG. To obtain functional antibody, we used disulfide cell-free system containing glutathione buffer (GSSG/GSH), E.coli protein disulfide isomerase (DsbC) and chaperone (GroEL/ES), oxidized glutathione (GSSG) treated E.coli cell extracts for efficient formation of disulfide bonds [3]. In this condition, however, heavy chain produced small quantity and insoluble form. To address this problem, initial 5 amino acid residues of chlorampenicol acetyltrasferase (CAT) are fused to N-terminal of wild type heavy chain. The engineered heavy chain can be expressed as enough as assembled with light chain to construct heterotetrameric protein, full-length IgG. Although the expressed full-length IgG was not glycosylated, selectively binds to the desired antigen (Bst2). This study was shown that E.coli derived disulfide cell-free protein synthesis system can be used as a simple, rapid, generally applicable method for the production of full-length IgG.