Enhanced batch dilution HRP refolding process: Effect of ILs and metal cofactors (Ca_2+ &Heme)

엄도영, 배상우, 구윤모†
인하대학교
(ymkoo@inha.ac.kr†)

During the last decade,there have been a lot of attempts to produce the recombinant protein via microorganisms. However, the development of protein refolding process to convert inclusion bodies(IBs) into active form of protein is still a key challenge in the production of recombinant proteins. Horseradish Peroxidase(HRP) is one of the representative metalloenzyme containing heme group and two calcium ions in their tertiary structure. It is well known that the stability of HRP is mainly controlled by these metal ions.In this study, HRP was selected as a model protein to elucidate the effect of calcium, heme cofactors, and ionic liquids(ILs) ([xMIM][Cl], x= ethyl ~ octyl) on the batch dilution refolding of HRP.When hemin, calcium chloride, and [EMIM][Cl] were added to the refolding buffer instead of urea, HRP refolding yield was remarkably improved more than 3.5 times compared with the case of the urea-containing conventional refolding buffer. The refolding yield was proportionally decreased with increasing alkyl chain length of [xMIM][Cl]. In temperature-dependent HRP refolding, the highest refolding yield was observed at 4℃.