

## SUMMARY

### Synthesis of difficult-to-express proteins by Escherichia coli expression systems (大腸菌発現システムを用いた難発現タンパク質の合成)

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In summary, through these studies it can be concluded:

1. Co-expression of MnP with DnaK-DnaJ-GrpE in SHuffle T7 Express followed by ATP maturation (ATP, CK, CP, and heme) can result in comparable specific activity with commercial MnP.
2. Expression of MnP, HRPs, and Fab in CyDisCo system was not significantly increased the solubility of target protein. However, the activation of T7HRP in activation buffer (CaCl<sub>2</sub>, hemin, GSSH in potassium phosphate) can increase the activity compared to not activated HRP.
3. Insertion of SKIK tag on zipbodyzyme N terminal is not only increase its solubility, but also its activity as confirmed by one-step ELISA. The base and acid addition to zipbodyzyme format containing Jun/Fos showed higher antigen-binding and enzymatic activity than base and base extension of Jun/Fos.