Enhanced Cell-free protein synthesis system

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Abstract

Cell-free protein synthesis systems provide rapid and convenient tools for expression of proteins. Previously considered only for laboratory-scale synthesis, this approach is now being considered as a promising alternative to conventional in vivo expression systems. Unfortunately, conventional cell-free systems have suffered from poor yields of protein synthesis, mainly as a result of their short reaction periods. In general, in an *Escherichia coli* batch cell-free system, protein synthesis lasts no more than 60 min. The reason for the early halt of protein synthesis has remained unanswered. Recently, we found that the concentration of inorganic phosphate (Pi) increases significantly during the incubation. The inhibitory effect of accumulating phosphate was relieved significantly by adding more magnesium. As a result, when the reaction was fed with magnesium acetate at 60 min, the final yield of chloramphenicol acetyl transferase (CAT) in the cell-free system reached 900 ug/mL after 3hr reaction.

References

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