A FACILE SCHEME FOR BIOSYNTHESIS OF PEPTIDES WITH NO LENGTH CONSTRAINTS

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While peptide drugs have become a viable class of biomedicines, efficient peptide expression and purification remains a critical technological need. We previous discovered that a number of self-assembling peptides such as 18A (EWLKAFYEKVLEKLKELF) and ELK16 (LELELKLKLELELKLK), when fused terminally to a target protein, can drive the target protein into active protein aggregates *in vivo*. A simple and rapid scheme for expression and purification of recombinant proteins using *Escherichia coli* was thus devised, by inserting a self-cleavable intein like Mxe GyrA between the self-assembling peptide and the target protein. In this scheme, the fusion protein is first expressed in the form of active aggregates, then separated by centrifugation upon cell lysis. Subsequently, the DTT-mediated intein self-cleavage reaction releases the target protein into solution. These self-assembling peptides together with the associated inteins constitute a set of cleavable self-aggregating tags (cSA), and provide an efficient route for the production of proteins with modest purity. More recently, this scheme has been applied to the biosynthesis of peptides, in particular those with lengths greater than 20 amino acids. A more efficient intein has also been engineered to afford the generation of authentic N-termini for the peptides. We believe this scheme will facilitate the development of more peptide drug candidates, and also lowering the costs of production of peptides of any length.