# Terminal specific PEGylation of rhEGF using intein technology

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PEGylation of biopharmaceutical proteins is important to increase in serum circulation stability and to minimize antigenicity. The most preferable method of PEGylation is a site- or terminal-specific, mono-PEGylation. We propose the terminal-specific mono-PEGylation method using intein-mediated fusion protein technology. By exploiting the affinity tagging domain (chitin binding domain) of a fusion protein, we were able to immobilize the C-terminus of the fusion protein to chitin matrix, exposing the N-terminus for solid-phase PEGylation. The distinct advantage of inteins is self-splicing ability by simple changes in pH and/or temperature without the need for cleavage proteins or reagents. By this way, we could present an integrated process for expression-refolding-PEGylation-purification. Rh-EGF (recombinant human epidermal growth factor) was used as a model protein. The PEGylation site was determined by mass spectrometry, and the bioactivity of the modified rhEGF was compared with the native and randomly PEGylated EGF using NRK cell culture assay.