

Single step protein purification and site-specific bioconjugation

Sortase-mediated expressed protein ligation and bioconjugation for molecular imaging and targeted therapeutics

Inventor

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STAGE OF DEVELOPMENT

Proof of principle and *in vitro* testing

INTELLECTUAL PROPERTY

PCT pending ([PCT/US2014/30208](#))

REFERENCE MEDIA

Warden-Rothman R et al.
[Analytical Chemistry](#), 2013, 85(22), 11090-11097.

DESIRED PARTNERSHIPS

- License
- Co-development

APPLICATIONS

- Facile, site-specific conjugation of proteins to surfaces, drugs, imaging agents, and nanoparticles
- Efficient production of targeting ligands to deliver therapeutics and imaging agents
- Quantitative flow cytometry
- PEGylation of biologics to improve circulation

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Overview

The Tsourkas lab has developed a novel conjugation technique that allows any single-chain recombinant protein to be site-specifically modified at the C-terminus during protein expression and purification procedures. Combining the concept of expressed protein ligation with the co-expression of the sortase enzyme, recombinant protein purification and conjugation are performed simultaneously. There are no additional purification steps to separate out unconjugated proteins, enabling facile and efficient functionalization of purified conjugated protein. Conjugation of protein to therapeutic agents, imaging agents, or linkers for subsequent conjugations, including click chemistry groups, can be performed in a site-specific manner. The system has been used successfully to express and conjugate a number of proteins, including EGFP, affibodies, single chain antibody fragments, extracellular matrix binding domains, and cytokines, with the conjugated peptides including visible and near-IR fluorophores and biorthogonal reactive groups.

Advantages

- Purification step linked to conjugation of protein
- No labor-intensive separation of conjugated proteins from unconjugated proteins
- Stoichiometric, site-specific, biocompatible conjugation
- Flexible, efficient, cost-effective system
- Avoids chemistry based on amines and thiols, expanding classes of proteins that can be expressed

