Exploring Innovative Liquid-Phase Peptide Synthesis Strategies with Silane-Containing Molecules

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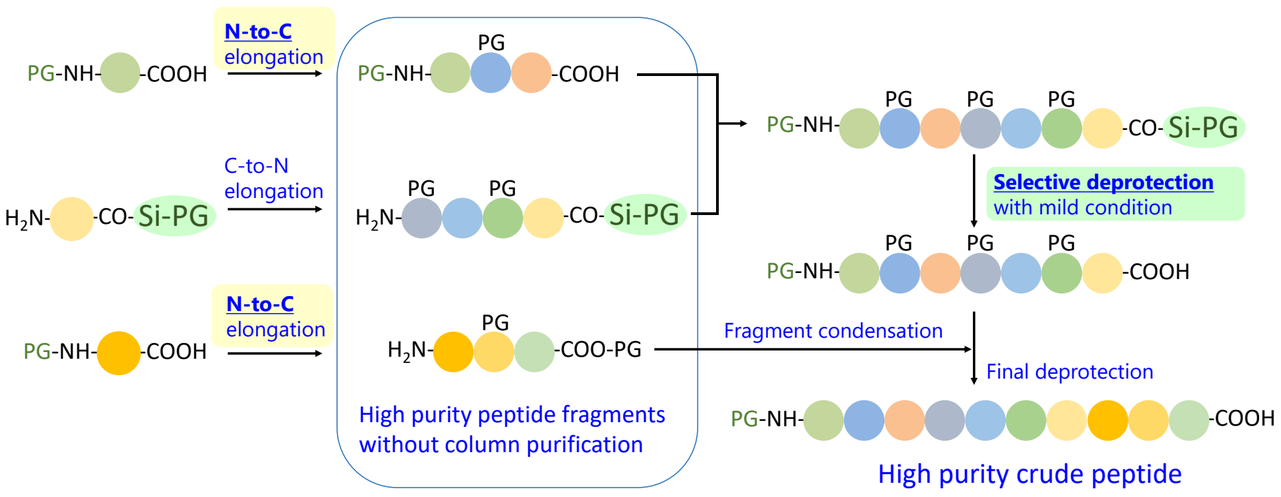
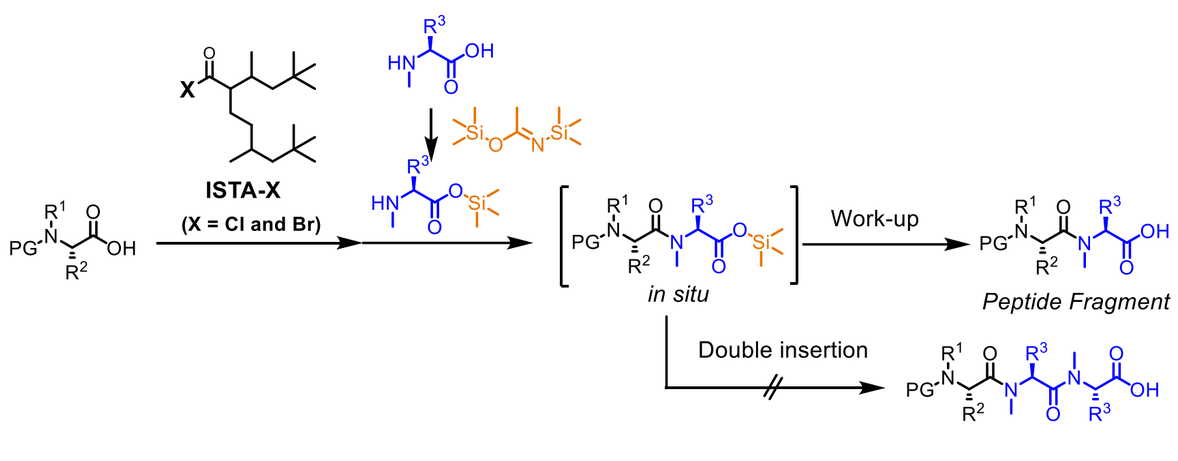
Introduction

Convergent liquid-phase peptide synthesis (LPPS) is a powerful method for producing short and medium-sized peptides. However, traditional LPPS is limited by side reactions, necessitating an extended period for process development. To address this challenge, we have developed a novel coupling method, new protecting groups, and novel solubilizing tags for LPPS, utilizing silane-containing molecules.



N-to-C coupling (R-Coupling)[1]

Our newly developed coupling method 'R-Coupling' employs isostearyl halide (ISTA-X) and a silylation reagent to facilitate peptide elongation in a non-conventional N-to-C direction.

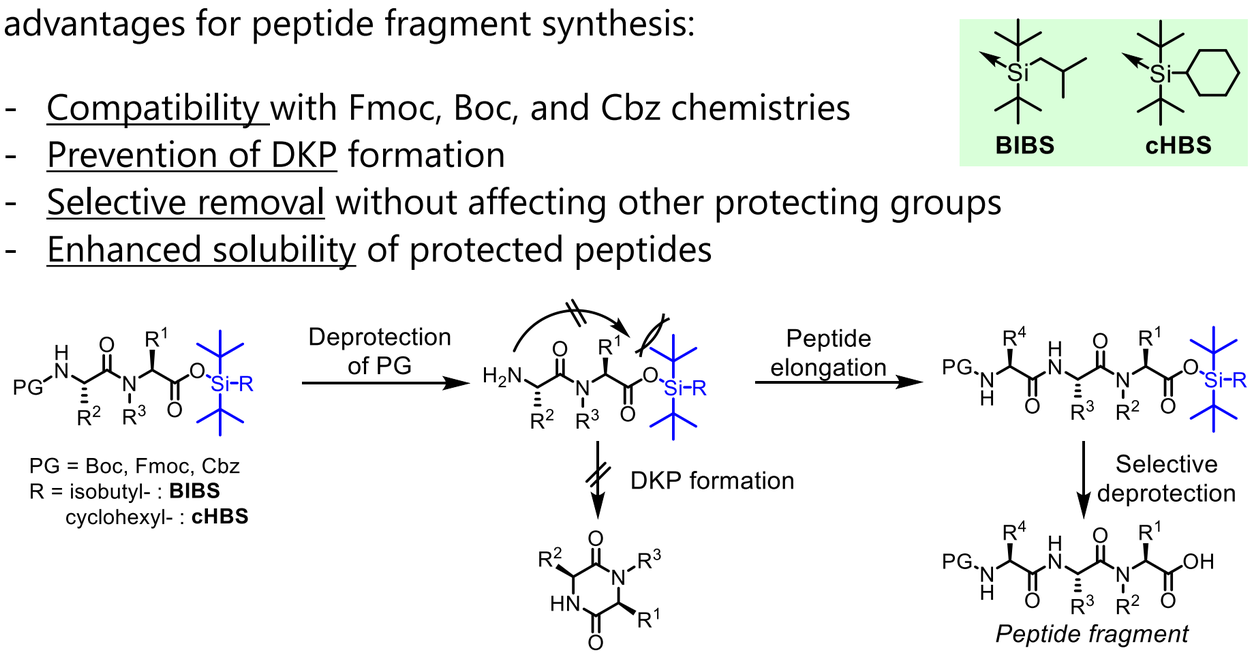


This inverse approach significantly reduces production time by eliminating deprotection steps, thereby minimizing overall synthesis steps. R-Coupling enables the rapid synthesis of high-quality peptide fragments, including those containing *N*-methyl amino acids.

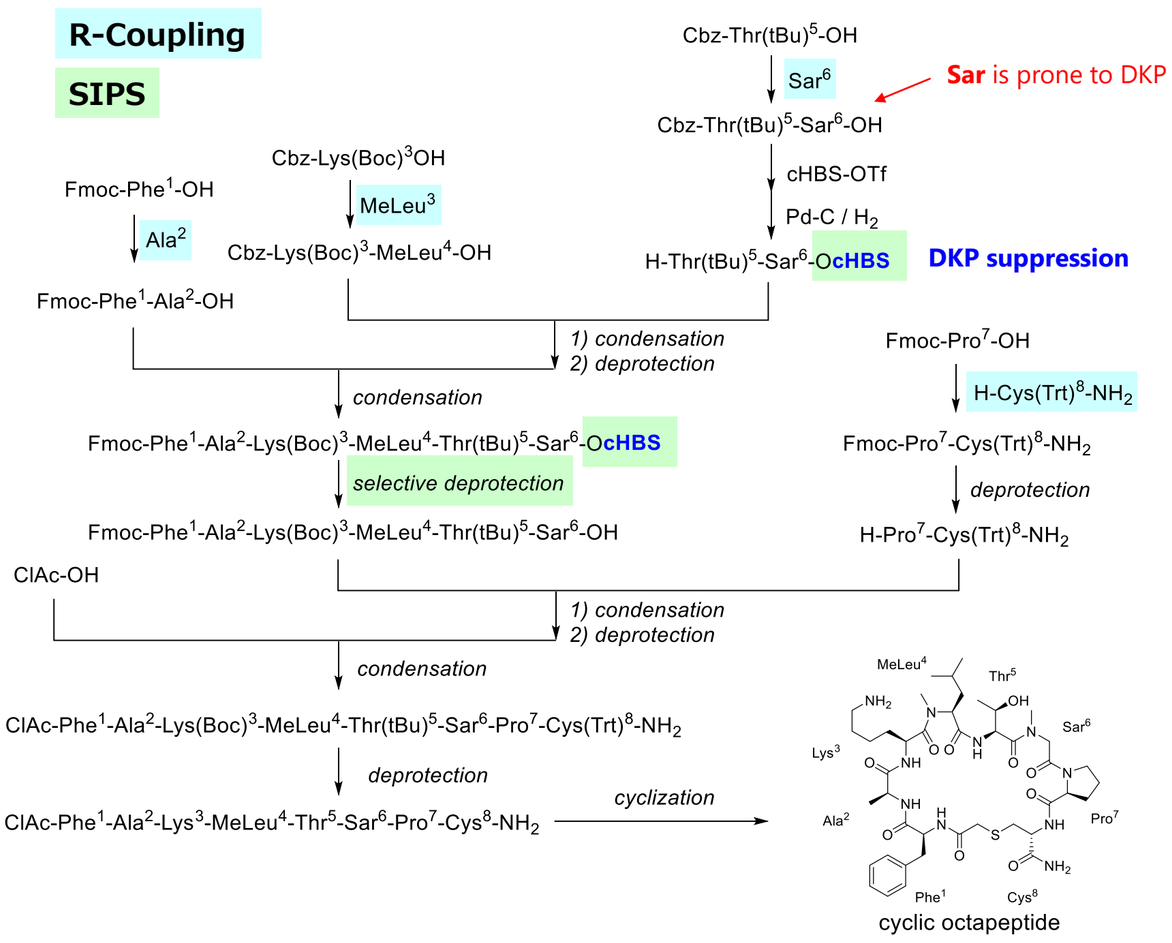
Silyl ester protecting group (SIPS)[2]

Traditional LPPS process development is time-consuming and labor-intensive, primarily due to the limitations of C-terminal carboxylic acid-protecting groups. Commonly used methyl and ethyl esters are prone to epimerization during basic deprotection, while diketopiperazine (DKP) formation is a significant challenge when proline or *N*-methyl amino acids are at the C-terminus. Our novel silyl-based protecting groups offer several advantages for peptide fragment synthesis:

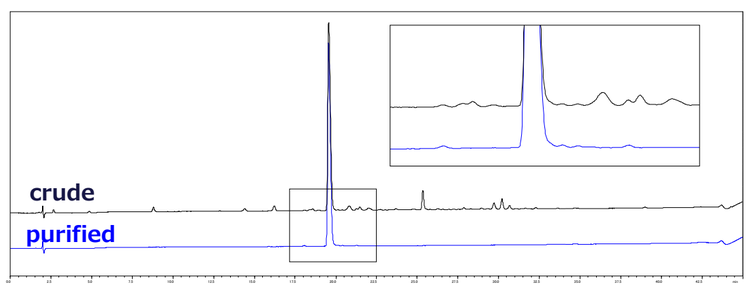
- Compatibility with Fmoc, Boc, and Cbz chemistries
- Prevention of DKP formation
- Selective removal without affecting other protecting groups
- Enhanced solubility of protected peptides



Example of cyclic peptide synthesis using R-Coupling and SIPS[2]



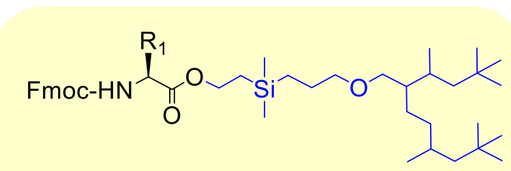
Intermediate peptide fragments bypassed column chromatography purification. The synthetic process was refined to minimize impurity generation near the main peak, enabling highly efficient purification through impurity management.



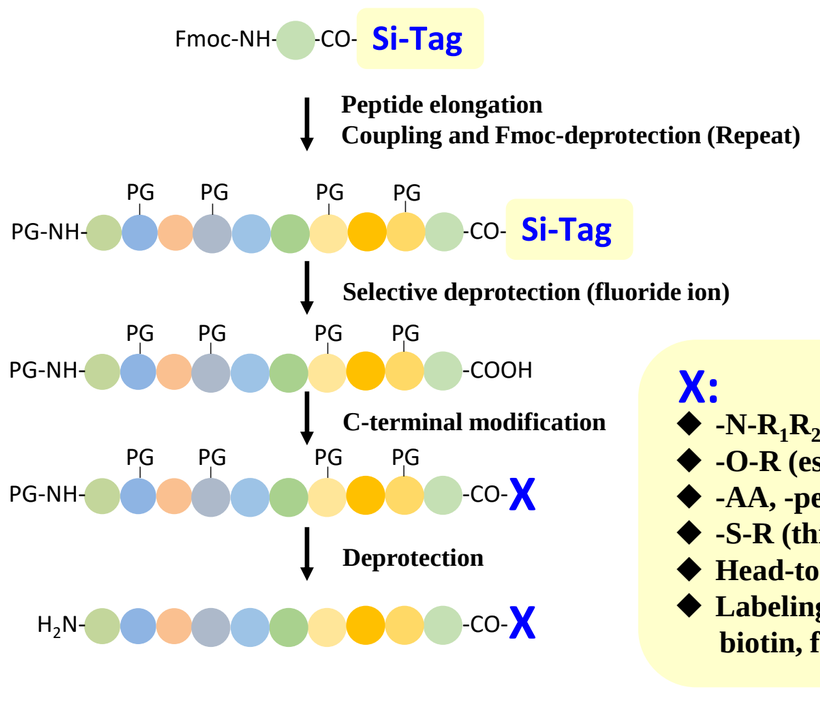
Alkyl silane solubilizing tag

Tag-assisted LPPS efficiently extends peptide chains sequentially. We developed a novel, alkyl-silane-based solubilizing tag (FMSE) for this process.

FMSE imparts exceptional solubility to peptides during synthesis. Uniquely, FMSE is selectively removable without affecting other protecting groups, generating peptide fragments.



This property enables diverse C-terminal modifications, including amidation, esterification, thioesterification, and conjugation with biotin, fluorophores, or chelators. Additionally, the C-terminal free peptide fragment serves as a versatile building block for fragment condensation, producing larger peptides.



Summary

We developed a novel convergent LPPS platform, SYNCSOL, employing silane-containing molecules. SYNCSOL incorporates N-to-C coupling, a new protecting group, and a detachable solubilizing tag. This platform efficiently produces high-quality peptide fragments, enabling fragment condensation and C-terminal modifications. SYNCSOL enhances LPPS flexibility and promises to improve peptide manufacturing productivity.

Reference: 1. Kurasaki, H. *et al*., (2020), *Org. Lett*. **20**, 8039–8043. 2. Nagaya, A. *et al.*, (2021) *Org. Process Res. Dev.* **25**, 2029–2038.