Synthesis of highly congested α-amino acids and peptides

A method for the synthesis of sterically congested α-amino acids and peptides using amino acid Schiff bases and hydrocarbons is developed. A superior helix-stabilizing effect of highly congested unnatural α-amino acids is demonstrated by circular dichroism measurements of synthesized peptides.

This is a summary of:
Tsuji, T. et al. α-Amino acid and peptide synthesis using catalytic cross-dehydrogenative coupling. *Nat. Synth*. https://doi.org/10.1038/s44160-022-00037-0 (2022).

Published online:
14 March 2022

The question

α,α-Disubstituted α-amino acids are widely observed in nature and are fundamental building blocks in medicinal and organic chemistry. The introduction of α,α-disubstituted α-amino acids into peptides leads to hydrophobicity, conformational constraints and peptidase tolerance, thus increasing their target specificity, binding activity and bioavailability (that is, the percentage of an administered dose of a drug that reaches the systemic circulation). Amino acid Schiff bases are some of the most used substrates for the synthesis of various natural and unnatural α-amino acids (Fig. 1a)[1]. However, the synthesis of highly congested α-amino acids bearing α,β-contiguous tetrasubstituted carbon centres, which might further enhance the above peptide properties, is uncommon because their steric hindrance has precluded the development of efficient and general synthetic methodologies. We previously developed a method for forming highly congested unnatural α-amino acids, but this method was inadequate for preparing mid-sized peptides for drug-development studies owing to the narrow substrate scope[2]. Furthermore, the effects of introducing highly congested α-amino acids into peptides remain unexplored.

The solution

Our previous method for synthesizing congested α-amino acids used an amino acid Schiff base as a persistent radical platform[2]. In this reaction, copper-mediated single-electron transfer involving an amino acid Schiff base and an α-bromocarbonyl was proposed to generate a persistent azaallyl radical species that couples with the tertiary alkyl radical formed from the α-bromocarbonyl[3]. However, the preparation of α-bromocarbonyls is cumbersome. We hypothesized that tertiary alkyl radical species could instead be generated from hydrocarbon feedstocks to couple with the persistent azaallyl radical to construct contiguous tetrasubstituted carbon centres.

We thus developed a copper-catalysed dehydrogenative Csp^3–Csp^3 bond-forming reaction[4] of amino acid Schiff bases that uses a hydrocarbon feedstock — typically an alkylarene — as the alkylating reagent to synthesize unnatural α-amino acids bearing α,β-contiguous tetrasubstituted carbons (Fig. 1b). This approach allows for the synthesis of various previously inaccessible unnatural α-amino acids and shows broad substrate scope. Moreover, the catalytic method can be used to modify peptides to form densely substituted dipeptides that bear up to four tetrasubstituted carbon centres (Fig. 1c), which are challenging to synthesize using conventional peptide coupling. Following modification, our congested α-amino acids can be incorporated into peptide synthesis, allowing us to prepare heptapeptides bearing a highly congested unnatural α-amino acid. Through circular dichroism measurements of these heptapeptides, we revealed that highly congested unnatural α-amino acids have a greater helix-stabilizing effect than less sterically congested analogues.

Future directions

We have demonstrated a method for preparing sterically congested α-amino acids and modifying peptides to deliver densely substituted dipeptides. Our mechanistic studies revealed that the reaction efficiency can be further increased by using an enhanced Brønsted basic copper catalyst, namely copper(I) *tert*-butoxide. Moreover, irradiation with a blue light-emitting diode allows for milder reaction conditions, offering the potential for the development of enantioselective catalysis. Nevertheless, at this stage, our method is limited to the synthesis of racemic α-amino acids, and realizing enantioselective catalysis will require a new strategy.

Our approach provides a path to explore the effects of highly congested α-amino acids in peptides. Indeed, our study confirmed the superior helix-stabilizing effect of highly congested unnatural α-amino acids, indicating their potential usefulness for the development of mid-sized peptides and functional peptide materials

We are continuing to investigate new peptide coupling reactions for highly congested α-amino acids and are also applying our original peptides bearing a sterically congested α-amino acid for the formation of mid-sized peptides, which have advantages in drug discovery research over small molecules and antibodies. We are now confirming the drug potential of the unnatural α-amino acid-containing peptides that we have synthesized.

Ryo Yazaki and Takashi Ohshima, Kyushu University, Fukuoka, Japan.

EXPERT OPINION

"Overall, this is an innovative work that will be of interest to the synthetic community. The use of alkylarenes as the coupling partner removes the need to prefunctionalize one of the coupling partners; this is a significant advance. The products, which would be difficult to prepare by other routes, are interesting and potentially useful."
Patrick Walsh, University of Pennsylvania, Philadelphia, USA.

FIGURE

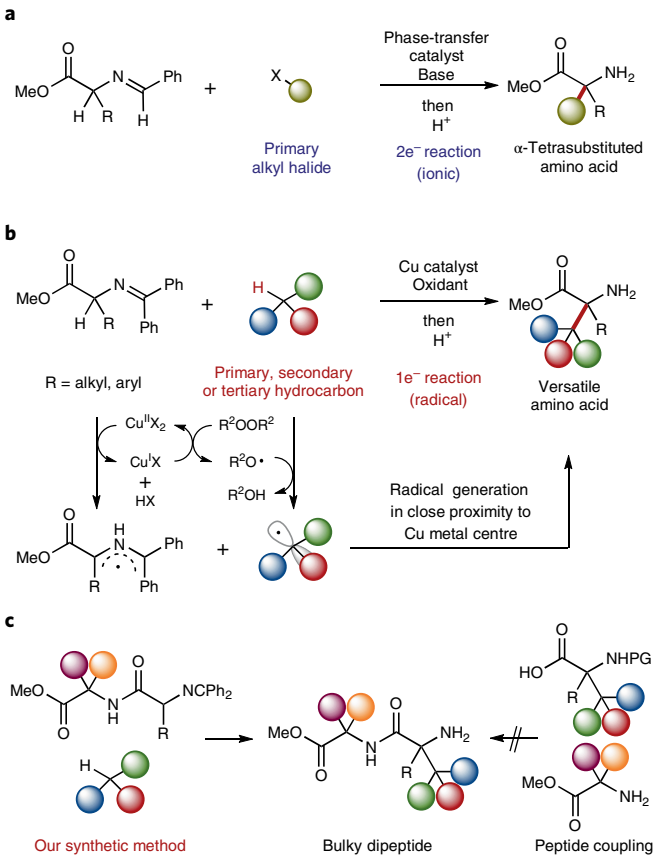

Fig. 1 | Amino acid Schiff bases for the synthesis of α,α-disubstituted amino acids and peptides. a, Conventional amino acid synthesis using amino acid Schiff bases under phase-transfer conditions. **b**, Our method of cross-dehydrogenative coupling of amino acid Schiff bases with a hydrogen feedstock to afford sterically congested unnatural α-amino acids. **c**, Synthesis of bulky dipeptides through alkylation of a peptide Schiff base. Credit: © 2022, Tsuji, T. et al.

BEHIND THE PAPER

We have been investigating bottom-up drug discovery based on organic synthesis and are especially interested in the development of mid-sized peptide drugs, which are expected to be a new modality for drug development targeting protein–protein interactions. Our interest in exploring the effects of highly congested α-amino acids on peptide properties motivated our search for a versatile method to prepare such α-amino acids. We thus sought a method that shows broad substrate generality and can be applied to peptide synthesis by focusing on radical chemistry to overcome the steric hindrance and increase the functional group tolerance. The key to success was finding that commonly used amino acid Schiff bases can serve as a radical cross-coupling partner. **R.Y.**

REFERENCES

1. O'Donnell, M. J. Benzophenone Schiff bases of glycine derivatives: versatile starting materials for the synthesis of amino acids and their derivatives. *Tetrahedron* **75**, 3667–3696 (2019).
A review article that presents the utility of amino acid Schiff bases.

2. Matsumoto, Y. et al. Amino acid Schiff base bearing benzophenone imine as a platform for highly congested unnatural α-amino acid synthesis. *J. Am. Chem. Soc.* **142**, 8498–8505 (2020).
This paper reports a synthetic method for preparing highly congested α-amino acids using amino acid Schiff bases bearing benzophenone imine.

3. Li, M. et al. Transition-metal-free chemo- and regioselective vinylation of azaallyls. *Nat. Chem.* **9**, 997–1004 (2017).
This paper reports pioneering work on cross-coupling using azaallyl radical species.

4. Li, C.-J. Cross-dehydrogenative coupling (CDC): exploring C–C bond formations beyond functional group transformations. *Acc. Chem. Res.* **42**, 335–344 (2009).
A review article that presents the utility of cross-dehydrogenative coupling.

FROM THE EDITOR

"This method by Yazaki and Ohshima particularly stood out as it allows the operationally simple functionalization of α-amino acid derivatives with hydrocarbons that are commonly unreactive. The copper-catalysed process enables the construction of α-amino acid derivatives with sterically congested carbon centres. The method has already been applied to the synthesis of helix-stabilized peptides and has the potential to become the go-to method for the synthesis of functionalized α-amino acids." **Thomas West, Associate Editor, *Nature Synthesis***