Late-Stage Peptide Diversification via Twisted Amide Bond Formation

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Peptides occupy a biomolecular niche between that of small molecules and proteins. Given that they can function not only as substrates in biochemical pathways, but as signaling molecules, peptide therapy presents a unique opportunity for medicine to closely mimic natural pathways.[1] As new challenges, such as mutant viral and bacterial strains with resistance arise, the need for modification of these peptide therapeutics has increased. In recent years, the use of acyclic twisted amides as reactants for cross-coupling reactions has been investigated. These twisted amides feature a distorted amide bond that has reduced double bond character and thus allows for the bond to be broken and replaced with various new bonds.[2]

We sought to find a method to selectively introduce these twisted amides into peptides and test their reactivity toward cross-coupling and nucleophilic acyl substitution reactions to introduce new functionalities. Amide bonds form the backbone of peptides and thus are a desirable synthetic handle for modification. Therefore, methodology to selectively modify peptides using these amide bonds leads to more possible peptide derivatives.

We started this process by investigating oxazolidinone and thiazolidinone, derivatives of serine and cysteine, respectively, using density functional theory (DFT) calculations to determine whether these structures exhibited a twisted amide bond. DFT is a computational quantum mechanical modeling method used to investigate the electronic structure of large molecules. From these initial calculations, we determined four potential moieties for twisted amides applicable to peptides: the aforementioned oxazolidinone and thiazolidinone structures in addition to their thiocarbonyl analogs.

Following the calculations, we moved to the synthesis of these moieties on small molecules before moving to the synthesis of these moieties on tripeptides. Following each step of synthesis, each product was purified using silica gel chromatography and confirmed by mass spectrometry and nuclear magnetic resonance (NMR). For work on small molecules, the methyl ester derivative of either serine or cysteine was used as a starting point. These small molecules with added carbonyl or thiocarbonyl groups were then reacted with benzoyl chloride to yield the desired potential twisted amide functionality. These potential twisted amides were tested as reactants in cross-coupling and nucleophilic acyl substitution reactions.

Work on small molecules gave positive results as these molecules featuring twisted amide moieties were successfully used in transamidation, esterification, Suzuki-Miyaura coupling, and Friedel-Crafts acylation. This result supports the formation of twisted amides as a method to introduce modifications to peptides.

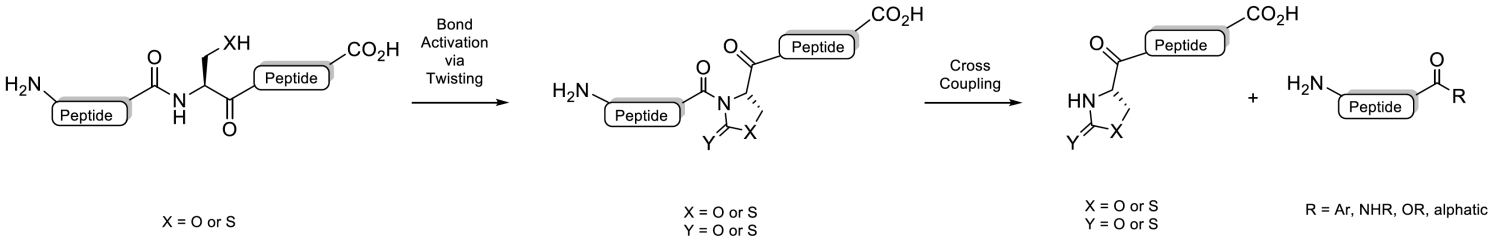

Figure 1. General scheme for peptide diversification using twisted amides.

The introduction of twisted amides to tripeptides is ongoing. Once these twisted peptides are formed, they will be tested as reactants in the previously mentioned coupling reactions to form modified peptide derivatives. Positive results in these tests support using the introduction of twisted amides to peptides as a method for late-stage diversification.

Statement of Research Advisor

Xavier has developed a new pathway for the synthesis of peptides in solution on a large scale. He also developed a new method for activating the unactivated, highly stable amide bonds for various cross-coupling reactions by twisting them.
– *Monika Raj, Chemistry and Biochemistry*

References

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[2] Liu, C.; Szostak, M. Twisted Amides: From Obscurity to Broadly Useful Transition-Metal-Catalyzed Reactions by N–C Amide Bond Activation. *Chem. – Eur. J.* **2017**, 23 (30), 7157–7173. https://doi.org/10.1002/chem.201605012.