Computational Design and Structural Analysis of Novel Peptidine Oligomers

Key words: Peptidomimetics, rotamer library, foldamers, rational drug desing.

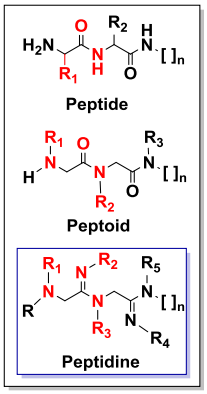
Background and Significance

Peptides are essential endogenous molecules with intriguing structures that enable them to have innumerable biorelevant functions. Although peptides have been designed as potential pharmaceutical agents, their poor bioavailabity and poor *in vivo* stability makes them bad drug candidates. This fact led to the study of peptidomimetics. One result from these efforts is peptoids, which contain a non-canonical peptidic backbone. Peptoids appear to be resilient to degradation, but their structures possess more degrees of freedom and thus pay a higher entropic penalty for target binding when compared to peptides (1).

Peptidines are a novel class of oligomers, structurally derived from peptides and peptoids (see **figure 1**). These molecules consist of repeating units of N- substituted amidines, a functional group found in drugs such as the histamine receptor antagonist cimetidine and ranitidine (Zantac) (2). The unique structure of peptidines enables the duplication of the amount of side chain; this structural feature could confer a unique secondary structure.

Peptidine synthesis is a straightforward process that consists of the iterative addition of imidoyl chloride and primary amines in sequence. To the present fifteen trimers, three tetramers and one pentamer have been synthesized in the Spiegel lab at Yale University (an example of one of this successful synthesis is shown in **figure 2**). However, studies regarding peptidine's structure are lacking. Gaining insight into peptidine structure will allow further investigation to evaluate these novel oligomers as key molecules for the development of peptidine based, more efficient therapeutics.

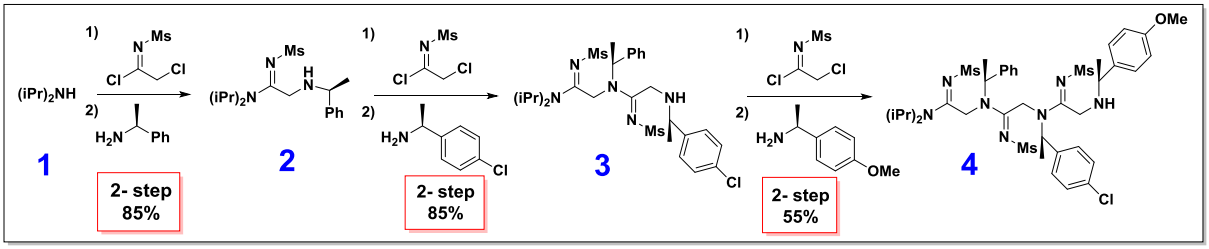
Figure 1. Oligomer-based biopolymers and mimetics. Depicted are structures of peptide, peptoid and the proposed peptidine oligomers. Monomer Backbone differences are shown in red.



[ ]$_n$= monomer sequence

I propose to study peptidines structure using an array of powerful techniques such as computational modeling software, X-ray Crystallography and Nuclear Magnetic Resonance (NMR). *I hypothesize that a) peptidines will have characteristic secondary structures similar to those found in peptides, although their secondary structures will be more rigid than peptoids and peptides due to their higher functional density; furthermore b) peptidine structure will vary with changes in pH, temperature and solvent, but c) by varying computationally the degree of steric hindrance on each side chain I will be able to control peptidine folding patterns in different chemical environments and thus control their function and selectivity once synthesized.*

Figure 2. Former synthesized peptidine trimer. Peptidines show higher functional density compared to peptides; this characteristic will enable the formation of rigid secondary structures.



Aim 1: <u>To determine experimentally former synthesized peptidine's structures using X-ray crystallography and multidimensional NMR spectroscopy.</u>

I will use X-ray crystallography to determine the electron density of former crystalline peptidines at Yale X-ray crystallography facility. Electron density data will allow me to characterize chemical bonds, electronic properties, dihedral angles and finally the mean position of atoms in each oligomer. To supplement crystallographic studies, I will use H^1 NMR NOESY for the characterization of peptide secondary structure (3), these studies will be held at Yale west campus NMR facility. Unlike other 2D NMR techniques (ex. COSY and TOCSY) NOESY detects spin polarization caused by through space dipolar interactions of atoms within 5 Å of distance. Thereby, using this data I can assign and analyze the sequence of interactions through a single oligomer which will lead me to its secondary structure. Using the same technique I will determine how these interactions change with variations in pH, temperature and solvent.

Aim 2: <u>To determine computationally peptidine's energetically favored conformations</u>

Recent expansions to the ROSETTA algorithm software allow the study of non-canonical backbones (4). In order to do molecular simulations using this program it is necessary to create a rotamer (energetically favored rotational conformers) library (5). To construct the library, dihedral and torsional angles from side chains and backbone must be determined. Using computational software like Chimera, I can predict torsional angles for a set of side chains. This data will be incorporated to ROSETTA along with dihedral angles to do molecular modeling (Density functional theory (DFT) and molecular mechanics (MM) calculations will be used in case ROSETTA software do not recognizes peptidine primary structure). I will then be able to predict peptidine structural preferences with various side chains. Intramolecular interactions can be studied for different sets of side chains this will be useful to determine folding patterns. Also, intermolecular binding activity will be studied to determine the degree of peptidomimetics in this new type of oligomers by modeling with biological receptors. This data will shed light on peptidine's function and its relation to their structure.

Intellectual merit

Studying the structures of peptidines will be a worldwide innovation in the field of peptidomimetic. It will contribute to the fundamental understanding on the relation between structure and function in oligomers that contain intriguing secondary structures that allow them to perform by efficient chemical mechanism *in vivo*. This infromation will allow the design of molecules with the desired secondary structures in order to improve and control their binding selectivity and function. Fundamentally the prediction of peptidine's biological interactions will be useful to establish their potential use as better drug leads.

Broader impact

Because peptidines are expected to have a more rigid structure, in comparison with peptoids and peptides, they will pay a lower entropic penalty for target binding. These novel oligomers are thus potential candidates to develop more efficient drugs by rational drug design to treat a wide variety of diseases. Robust peptidine based libraries can aid in the development of novel therapeutics that will be expected to have better binding efficiency, higher selectivity and vastly improved bioavailability, compared to those of peptoids and peptides.

References

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