# Exploring protein-protein interaction inhibitors and peptide nanostructures

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## Introduction

This report summarises our group's investigations in the area of chemical biology principally based around two key peptide secondary structure motifs; α-helices and β-sheets. The α-helix is the most abundant secondary structural feature found in proteins and, as such, plays an important role in a significant number of protein-protein interactions (PPIs), many of which are implicated in disease development and progression. Proteomimetics can act as inhibitors of PPIs by matching the special orientation of key binding residues on the native α-helix.

β-sheet peptide nanostructures posses unique physical properties due to their complex internal architecture. Examples include amyloid fibrils which have been identified as key players in the etiology of numerous debilitating and terminal diseases. Covalent cross-linking has been proposed as an effective method for extracting structural information from such supramolecular peptide structures.

## α-Helix mimetics as PPI inhibitors

Aromatic oligoamides based on three different scaffolds; 2-*O*-alkylated, 3-*O*-alkylated and *N*-alkylated (Figure 1a), reported from our group act as effective inhibitors of the oncogenic p53-*h*DM2 interaction. The more recently designed 2-*O*-alykated trimers were compared to the original 3-*O*-alkylated regioisomers. Initial molecular modelling studies on the two scaffolds indicated a significant difference in backbone curvature (Figure 1d.) which was confirmed by both solution and solid-state conformational studies. This difference in curvature arises due to the different internal hydrogen bonding patterns within the trimers where the 2-*O*-alkylated compounds are able to form 6-membered hydrogen bonding rings whereas the 3-*O*-alkylated compounds are forced to twist in order to adopt a 5-membered hydrogen bonding ring. In addition, an automated microwave synthetic methodology has been optimised for the *N*-alkylated scaffold, whilst molecular dynamics simulations on protein-bound oligoamides have been performed. In other developments, we have shown that mass spectrometry can be

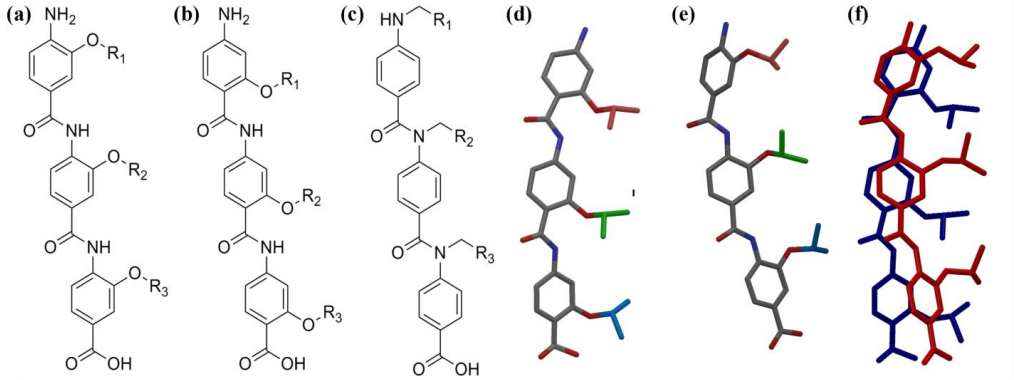


**Figure 1:** structures of **(a)** 3-*O*-alykylated helix mimetic scaffold; **(b)** 2-*O*-alkylated scaffold and **(c)** *N*-alkylated scaffold. Modes of **(d)** a 2-*O*-alkylated scaffold where $R_1 = R_2 = R_3$ = iPr and **(e)** a 3-*O*-alkylated scaffold where $R_1 = R_2 = R_3$ = iPr. **(f)** Overlay of both model structures demonstrating the difference in backbone curvature.

used to speciate dynamic combinatorial libraries based on iron (II) tris bipyridyl chelates**.**

**Covalent cross-linking within peptide nanostuctures**

Covalent cross-linking between the strands of a β-sheet using 3-aryl-3-(trifluoromethyl)diazirine (TFMD) to generate a highly reactive and non-selective carbene in combination with mass sepctrometry (MS) and tandem mass spectrometry (MS/MS) has been found to be a very effective method for the analysis of secondary structure in amyloid fibrils (Figure 2). Peptide $A\beta_{16\text{-}22}$ (Ac-KLVFFAE-$NH_2$) is known to form distinct morphologies at different pHs; at neutral pH it assembles into filaments and at pH 2.0 into nanotubes. Two TFMD modified peptides were synthesised with the cross-linker substituted for either F19 or F20 ($A\beta_{16\text{-}22}$-F19* and $A\beta_{16\text{-}22}$-F20*, respectively). It was therefore envisaged that the change in sequence or a change in pH would after supramolecular structure and lead to formation of distinctive cross-links. The resulting MS fragmentation patterns were only dependent on cross-linker position and not on the assembly pH. It is likely therefore that the filament and nanotube structures contain the same β-sheet unit with pH-dependent higher order packing.

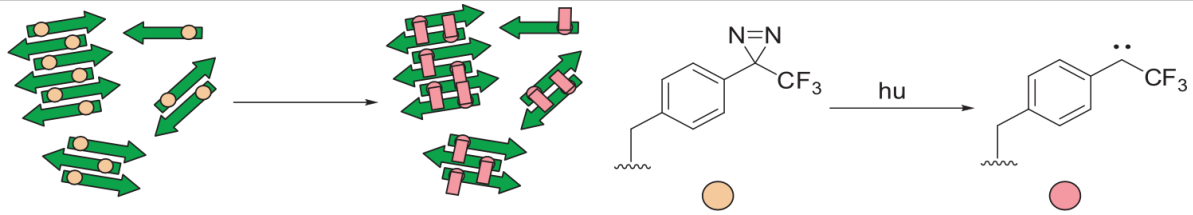


**Figure 2:** cartoon representation of the cross-linking strategy along with the structure of the 3-aryl-3-(trifluoromethyl)diazirine cross-linker and its corresponding reactive carbene.

**Publications**

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**Collaborators**

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