Chemoselective Fragment Condensation Of Peptidomimetic Oligomers

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Supporting Information

A. Synthesis of Peptoid Oligomers and General Ligation Reaction Conditions

The submonomer amines 2-methoxyethylamine, propargylamine, allylamine, propylamine, aniline, (S)-(-)-1-phenylethylamine, and benzylamine were purchased from either Aldrich or TCI America. Other reagents were obtained from commercial sources and used without additional purification.

HPLC analysis and purifications were performed on a Beckman Coulter System Gold instrument. For analytical analysis, a C_{18} reversed-phase HPLC column was used (Peeke Scientific). Samples were eluted with a 5-95% acetonitrile/water gradient (0.1% TFA) in 10 minutes with a flow rate of 0.7 mL/min and monitored at 214 nm. For purifications, semi-preparative C_{18} reversed-phase HPLC columns were used (Peeke Scientific). Samples were eluted with a 5-95% acetonitrile/water gradient (0.1% TFA) in 50 minutes with a flow rate of 2.5 mL/min, and monitored at 230 nm.

Mass spectrometry was performed on an Agilent 1100 Series Capillary LCMSD Trap XCT system mass spectrometer (electrospray-ion trap) or a Bruker OmniFLEX MALDI-TOF mass spectrometer.

Synthesis of Peptoid Fragments: Peptoid oligomers were synthesized on 2-chlorotrityl resin (Novabiochem). The resin (100 mg) was swollen in 3 mL of dichloromethane (DCM) for 5 min. and washed twice (3 mL DCM) for 1 min. Bromoacetic acid (90.3 mg), 107 μL N,N-Diisopropylethylamine (DIPEA) and 1 mL DCM were added for 40 min. Washing steps using DCM (3×2 mL) and DMF (4×2 mL) were performed and 1.0 mL corresponding amine in DMF (10 mL/g resin) was added and the reaction was shaken for 30 min (Step A). Following washing, Bromoacetic acid (0.167g/1mL DMF) and diisopropylcarbodiimide (DIC, 2 mL/g resin) in DMF were added to the resin and

allowed to shake at rt for 20 min (Step B). Steps A and B were then repeated until peptoid oligomers of desired length were obtained. Prior to cleavage, peptoid oligomers were taken up in 3 mL of a 1:2:3 (acetic anhydride/pyridine/DMF) solution for 2 hours to obtain N-terminal acetylated peptoid oligomers. The 2-chlorotrityl resin was then cleaved with 20% hexafluoroisopropanol in DCM and the peptoid was lyophilized overnight. An equimolar amount of salicylaldehyde was prepared in dry DCM (2-4 mL) and a slight excess of DIC (1.2 equiv.) and a catalytic amount of DMAP (0.1 equiv.) were added to the peptoid.[1] The reaction(s) were allowed to proceed for 6-24 hours. The resulting peptoid esters were purified by HPLC, and lyophilized overnight.

Synthesis of Peptides: Peptides were prepared as previously reported.[2]

General Procedure for Ligation: The peptide fragment (1.1 equiv.) and the peptoid phenyl ester fragment (1.0 equiv.) were dissolved in pyridine/acetic acid (1:1 mole/mole) to a final concentration of 0.05 M. The reaction was stirred at room temperature and monitored using LCMS and HPLC. Following completion of the reaction (consumption of salicylaldehyde to form the acteal intermediate), the solvent was removed by lyophilization and the intermediate product was treated with TFA/H_2O/*i*-Pr_3SiH (94/5/1, v/v/v) for 10 min to give the product containing a native amide bond at the ligation site.

B. MS Characterization of Peptoid and Peptide Oligomers

Entry	Compound	Calc. m/z	Obs. m/z
1	**1**	541.2	564.1[a]
2	**2**	587.3	609.9[a]
3	**3**	559.2	581.9[a]
4	**4**	322.2	344.9[a]
5	**5**	336.2	358.9[a]
6	**6**	846.2	868.9[a]
7	**7**	1,013.5	1,014.7[b]
8	**8**	1,680.9	1,680.6[b]

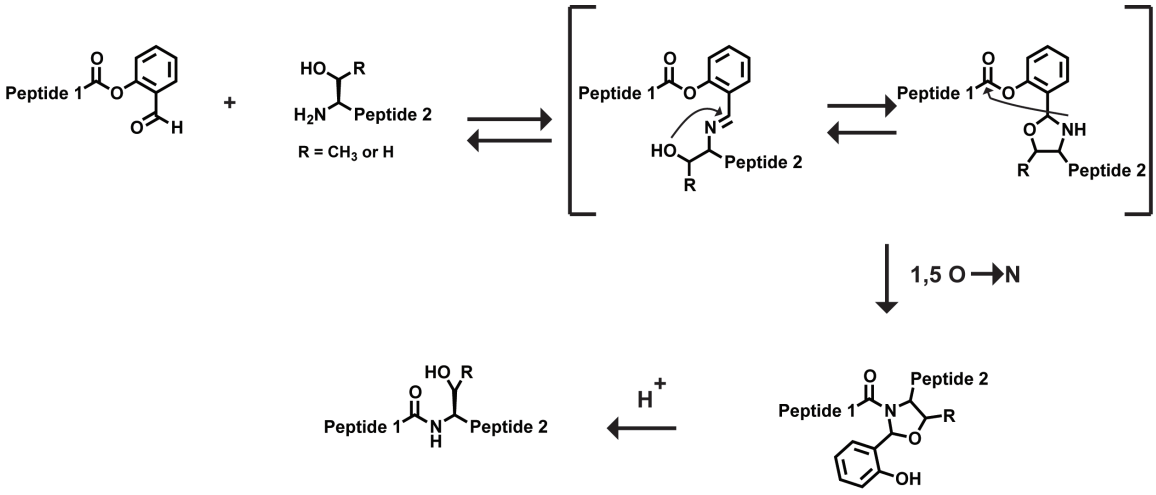
Supplementary Table 1**.** Mass spectrometry data for peptoid and peptide oligomers acquired using ESI techniques. [a] [M+Na]. [b] Mass spectrometry data for peptide oligomer acquired using MALDI-TOF techniques.

B. MS Characterization of Intermediate Products

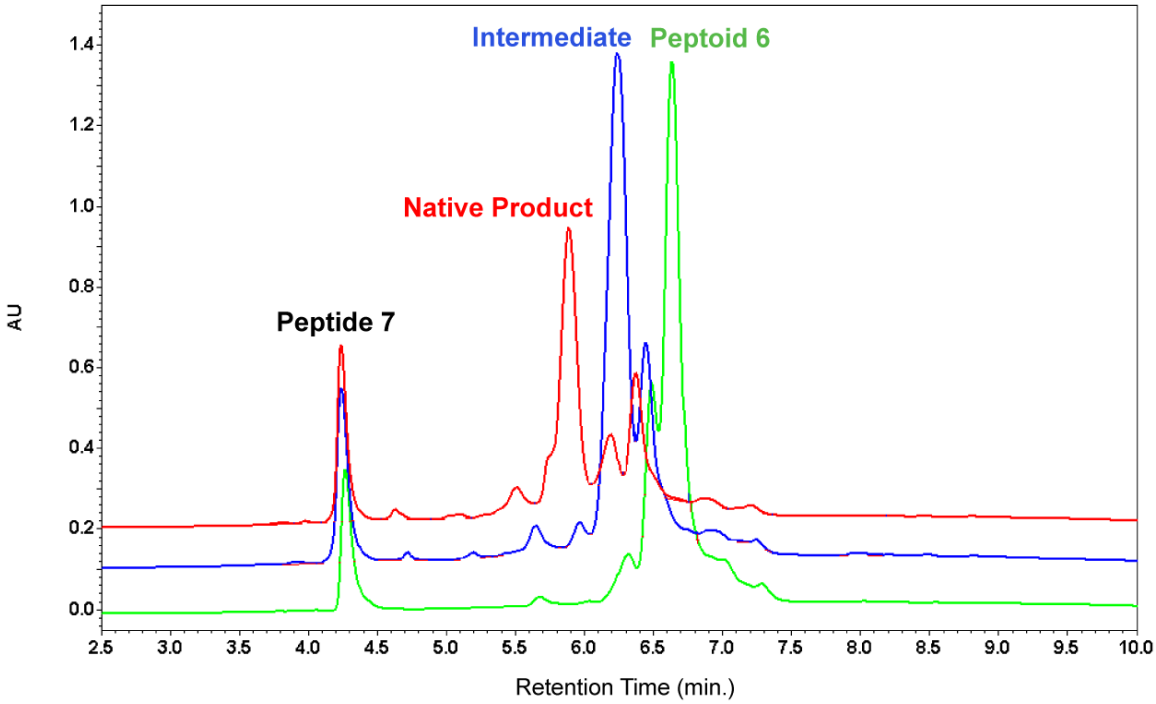
Entry	Compound	Calc. m/z	Obs. m/z
1	**1+4**	845.4	867.9[a]
2	**2+4**	891.4	913.8[a]
3	**2+5**	905.4	927.9[a]
4	**3+4**	863.4	885.9[a]
5	**3+5**	877.4	899.9[a]
6	**2+8**	2,249.0	2,250.7[b]

Supplementary Table 2**.** Mass spectrometry data for peptoid and peptide oligomers acquired using ESI techniques. The conversion is calculated based on the consumption of the salicylaldehyde. [a] [M+Na]. [b] Mass spectrometry data for oligomer acquired using MALDI-TOF techniques.

C. Supporting Information Figures



Supplementary Figure 1. Proposed ligation mechanism to form native Ser/Thr linkages. Figure adapted from Ref. 1.



Supplementary Figure 2. Fragment condensation between C-terminal peptoid salicylaldehyde ester **6** and peptide **7** as crude components. The reaction went to completion within 6 hours. Upper traces of intermediate (blue) and native product (red) are offset in *y*-direction for clarity. Reaction monitored by HPLC (214 nm) and LCMS.

E. References

1. X. Li, H. Y. Lam, Y. Zhang and C. K. Chan, *Org. Lett.*, 2010, **12**, 1724.
2. R. A. Turner, R. J. Weber and S. Lokey, *Org. Lett.*, 2010, **12**, 1852.