

Chemical Synthesis of Cyclotide from the Medicinal Plant *Clitoria ternatea*

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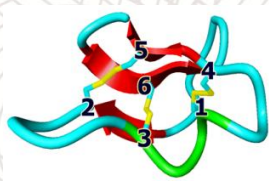
Introduction

Cyclotides are a special class of ultrastable peptides which possess a cyclized backbone and three disulfide bridges. They display many bioactivities and are attractive targets for drug discovery. The medicinal plant *Clitoria ternatea* contains a high abundance cyclotides, named as cliotides (cT, which have been shown to constitute the active ingredients of *C. ternatea*).

cT19 is one of the cliotides that showed strong antibacterial and immunomodulatory activities. Native cT19 yield from conventional extracting methods is relatively low and it is difficult to separate cT19 from its deaminated form cT19a. With the recent understanding of cT19's structure and amino acid sequence, chemical synthesis of cT19 is made possible.

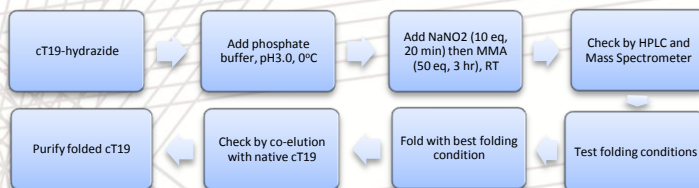


Clitoria ternatea



The Structure of cliotide

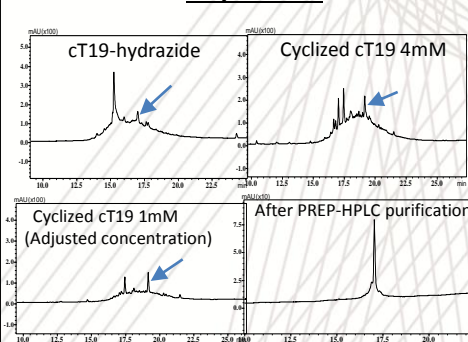
Methodology



Results

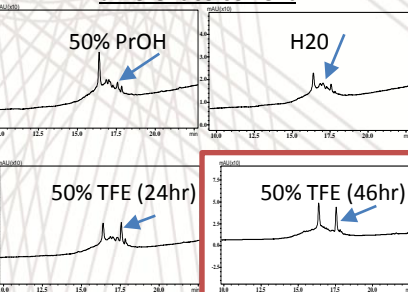
Images generated by High-performance liquid chromatography

I. Cyclization



Cyclized cT19 was obtained for further folding.

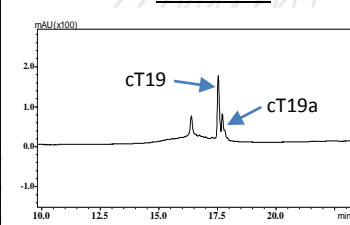
II. Folding: Test Conditions: 0.1mg/ml cT19, cT19:GSH:GSSG=1:150:5, RT with different co-solvent



Thus, 50% TFE was chosen as the co-solvent, the incubation time was 46hr.

III. Co-elution & Yield

0.1mg/ml cT19 Co-Elution with Native cT19

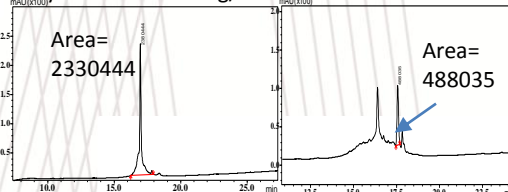


The sample co-eluted with native cT19.

Calculating Yield:

Starting material: cyclized cT19 0.1mg/ml

Folded cT19



$$\text{Yield} = \frac{488035}{2330444} = 20.9\%$$

Conclusion

The peptide cT19 was successfully synthesized. The folding condition provides information for further study on cyclotide folding. Further studies can be done on better folding conditions to achieve a higher yield.

Acknowledgement

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Reference

1. Nguyen GK, Zhang S, Nguyen NT, Nguyen PQ, Chiu MS, Hardjojo A, Tam JP. (2011). *Discovery and characterization of novel cyclotides originated from chimeric precursors consisting of albumin-1 Chain a and cyclotide domains in the Fabaceae family*. J Biol Chem. 286(27),24275-87
2. Zheng JS, Tang S, Ye G, Hangman HN, Liu L. (2011). *Synthesis of Cyclic Peptides and Cyclic Proteins via Ligation of Peptide*. ChemBioChem. DOI: 10.1002