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Peptidomimetic building blocks for drug discovery: An overview

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Abstract

We have developed new asymmetric and enantioselective routes for the preparation of a variety of conformationally constrained peptidomimetic building blocks. These molecules include α-methylcysteine, α-methylthreonine, *allo*-threonine, α,β-dimethylcysteine, α,β-dimethyltryptophan, twisted amide, and substituted lanthionine structures. It has been shown that these molecules are very useful building blocks for the peptidomimetic drug design.

In the past two decades, a wide variety of naturally occurring peptides have been discovered. These peptides play very important biological roles as hormones, enzyme inhibitors, substrates, neurotransmitters and immunomodulators among others. After binding to their corresponding receptors or enzymes, they can influence cell-cell communication and control a series of vital functions such as metabolism, immune defense, digestion, respiration, sensitivity to pain, reproduction, behavior, and electrolyte levels. Thus, extensive studies have been undertaken in an effort to understand the physiological effects of these peptidic molecules toward the design of new peptide-based therapeutic agents.

For peptide-based drug design, there are several major considerations that limit clinical applications such as: (1) rapid degradation by many specific or nonspecific peptidases under physiological conditions; (2) conformational flexibility which allows a peptide to bind to more than one receptor or receptor subtype leading to undesirable side effects; (3) poor absorption and transportation because of their high molecular mass or the lack of specific delivery systems, especially for some peptides which require the passage through the blood-brain-barrier (BBB) to act in the central nervous system (CNS). In an effort to counteract these problems, peptidomimetic drug design has emerged as an important tool for both peptide chemists and medicinal chemists. This approach has evolved as an interdisciplinary scientific endeavor combining organic chemistry, biochemistry and pharmacology.[1]

As one of the major efforts in organic chemistry, a variety of molecules have been designed to mimic the secondary structures of peptides, such as α-helices, β-turns, and β-sheets. In order to explore the structure-activity relationships (SAR) of bioactive peptides, a number of strategies have been developed by incorporation of conformationally constrained amino acids, modification of the peptide backbone by amide bond isosteres, cyclizations, attachment of pharmacophores to a template or scaffold, and the synthesis of nonpeptide analogs. As a result of such endeavors, the advantage of peptidomimetics over the native peptides has been demonstrated by increasing the potency and selectivity, decreasing the side effects, improving oral bioavailability, and the half-life of the activity through minimizing enzymatic degradation.[1] In this paper, we describe our recent efforts on design of peptidomimetic building blocks.

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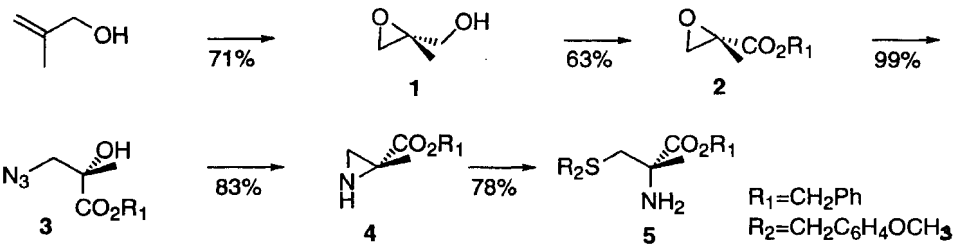
1. Asymmetric synthesis of α-methylcysteines via chiral aziridines

The use of chiral α,α-disubstituted amino acids in a peptide can severely restrict the conformations of the peptide backbone. Among them, α-methylcysteine and derivatives are attractive target molecules because they can form further constrained cyclic structures via disulfide bridge formation. In addition, Heathcock and Pattenden showed that α-methyl cysteine is an important building block for a new family of natural products, thiangazole, tantazoles and mirabazoles, which exhibit antitumor and anti-HIV-1 activities. [2]

Because of the labile nature of the sulfhydryl group, very few routes have been reported for the asymmetric synthesis of α-methyl cysteine.[3] Our desire to prepare protected α-substituted amino acids suitable for peptide syntheses led us to explore a general synthetic strategy. We have recently developed a new asymmetric synthesis of protected α-methyl cysteine derivatives based on chiral aziridine synthons.[3]

Scheme 1



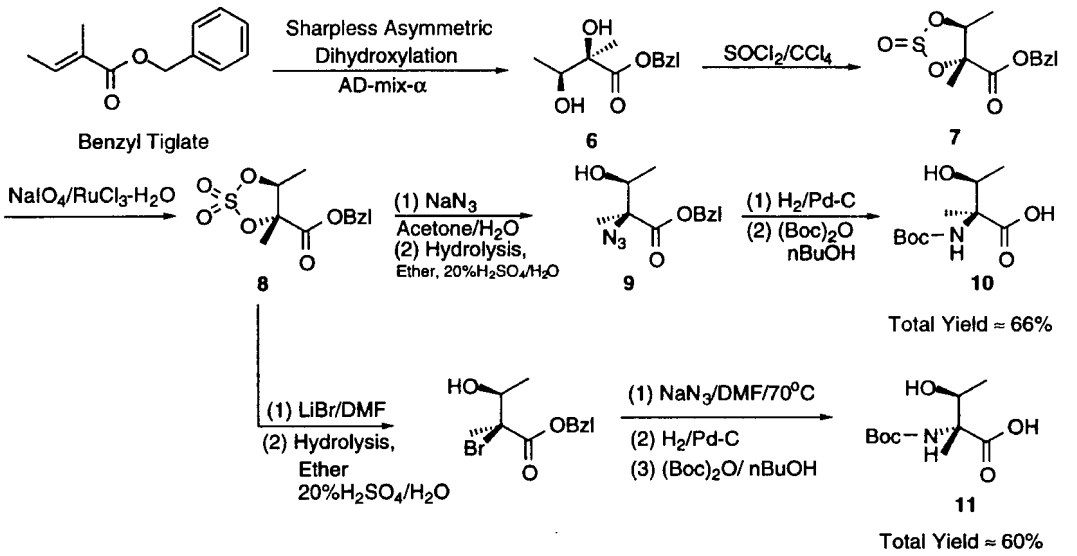
Starting with the allylic alcohol, 2-methyl-2-propen-1-ol (**Scheme 1**), the (R)-2-methylglycidol (**1**) was readily obtained in high enantiomeric purity by Sharpless asymmetric epoxidation.[4] Oxidation of (**1**) with ruthenium(VIII) oxide provided the corresponding carboxylic acid (**2**, R_1=H). The carboxylic acid was converted without further purification to the benzyl ester (**2**) by the standard carbodiimide esterification method. Treatment of benzyl (S)-2-methyl-2-oxirane carboxylate (**2**) with sodium azide resulted in regioselective ring opening to form the azido alcohol (**3**). Refluxing the azido alcohol (**3**) with triphenylphosphine in acetonitrile generated the benzyl aziridinecarboxylate (**4**). Overall, the transformation of the epoxide to the aziridine occurs with no loss of enantiomeric purity and in good to excellent overall yields.

With the aid of a Lewis acid, the ring opening of benzyl aziridine-2-carboxylate with a thiol predominantly occurs at C-3 in an S_N2 fashion. Regioselective ring opening of the chiral aziridine (**4**) with 4-methoxy-α-toluenethiol and boron trifluoride etherate resulted in the desired (S) α-methyl cysteine(**5**). The enantiomers of (**1**) through (**5**) were also synthesized based on Scheme **1** using diethyl L-tartrate in the Sharpless asymmetric epoxidation. Both enantiomers were obtained in excellent optical purities. These protected α-methyl cysteine derivatives are useful building blocks for peptide synthesis.

2. Stereoisomeric synthesis of α-methylthreonines

Among the naturally occurring amino acids, serine and threonine are unique residues because the hydroxyl groups in their side chain are crucial linkages in glycopeptides and phosphopeptides.[5] It has been shown that O-glycosylation of serine and threonine in peptide or protein structures can not only influence the physical properties as well as the conformations, but also protect these conjugates against proteolytic attack. Moreover, the carbohydrate portions of the glycoproteins have been found to be important recognition labels, e.g., in infectious processes, cell differentiation, and the regulation of cell growth.[5a] On the other hand, the phosphopeptides and proteins formed through O-phosphonation on serine and threonine represent a important family of molecules such as enzymes, growth factors receptors, cytosketetal contractile, and oncogenic proteins.[5b] Our goal is to design four stereoisomeric α-methyl threonines as a new set of conformationally constrained linkages in the drug design of glycopeptides and phosphopeptides.

Scheme 2

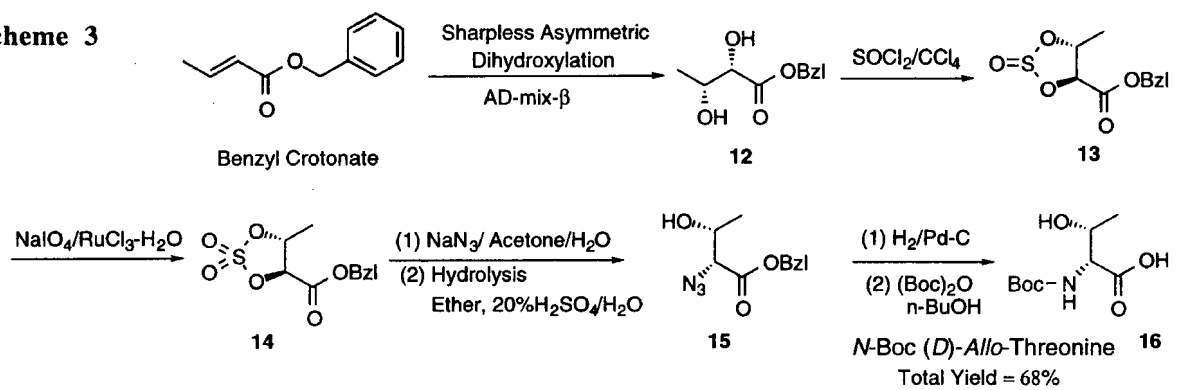


Starting with benzyl tiglate (60mmol), the Sharpless AD reaction[6] with AD-mix-α in the presence of methanesulfonamide proceeds smoothly to give diol **6** (**Scheme 2**) with excellent optical purity (>98% ee). The diol **6** is converted to its 2,3-cyclic sulfite **7** with $SOCl_2$ and oxidized to cyclic sulfate **8** in a one-pot fashion. Nucleophilic substitution then occurs at the α-C of **8** with clean inversion of chirality. Acidic hydrolysis provides the desired α-azido ester **9**. Compound **9** readily undergoes catalytic hydrogenation to produce the enantioisomerically pure (2S, 3S) 2-methylthreonine **10**. The X-ray diffraction derived structure of **10** proves the correct stereochemistry of the final product. In addition, the cyclic sulfate **8** can be converted to its diastereoisomer, (2R, 3S) 2-methylthreonine **11** by two sequential inversion steps with LiBr and NaN_3. This method provides a new route to prepare four stereoisomers of α-methylthreonine derivatives using the corresponding azido alcohols.

3. Asymmetric synthesis of *allo*-threonines

In addition to the importance of serine and threonine addressed above, it has been shown that the nonproteinogenic L-*allo*-threonine and D-*allo*-threonine are useful building blocks for a wide variety of bioactive peptides, such as the phytotoxic syringopeptins, the antiviral agent viscosin, the antibiotic cyclodepsipeptide complex CDPC 3510, and glycopeptidolipid antigens from infectious diseases particularly in AIDS patients.[5c, 5d] The use of these amino acids has been hampered by synthetic difficulties. As an extension of the above strategy, we have synthesized the *allo*-threonine analogs starting with benzyl crotonate (**Scheme 3**). Starting with benzyl crotonate, the Sharpless AD reaction[6] with AD-mix-β in the presence of methanesulfonamide proceeds smoothly to give diol **12** with excellent optical purity (>99% ee). The diol **12** is converted to its 2,3-cyclic sulfite **13** with $SOCl_2$ and oxidized to cyclic

Scheme 3

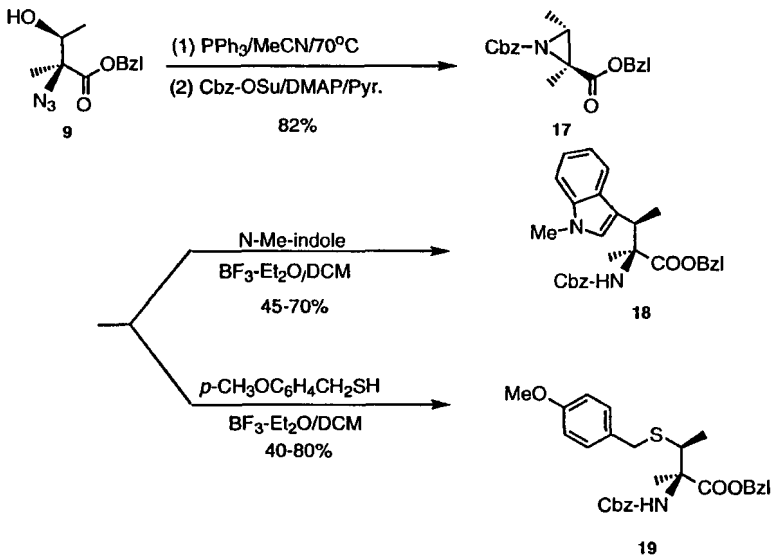


sulfate **14** in a one-pot fashion. Nucleophilic substitution then occurs at the α-C of **14** with clean inversion of chirality. Acidic hydrolysis provides the desired α-azido ester **15**. Compound **15** readily undergoes catalytic hydrogenation to produce the enantioisomerically pure (2R, 3R) *allo*-threonine **16**. This efficient method can provide the *allo*-threonine in 30 mmol scale and high optical purity which we anticipate will provide large amounts of L- and D-*allo*-threonines in a facile manner.

4. Asymmetric synthesis of α,β-dimethylcysteines and α,β-dimethyltryptophans

The α,β-dimethylcysteine and tryptophan peptidomimetics represent another family of novel building blocks. Because of their labile side chains and chirality requirement, they cannot be prepared by conventional routes. Using the chemistry we developed in our laboratory (**Scheme 4**), these molecules can be synthesized efficiently and stereospecifically. The α-azido ester **9** can be stereospecifically transformed to the actived benzyloxylcarbonyl protected aziridine-carboxylate **17** Compound **17** is a very important synthon for the synthesis of α,β-dimethylatedtryptophan **18** and cysteine **19**. Based on this method, four stereoisomers of cysteine and tryptophan analogs can be prepared using the corresponding azido alcohols.

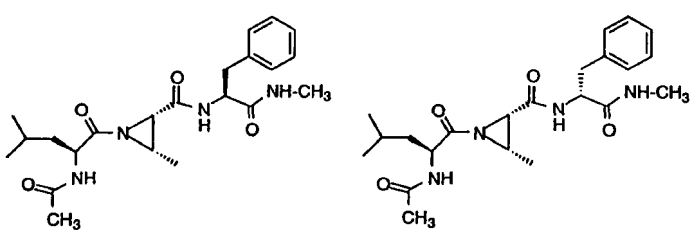
Scheme 4



5. Synthesis of twisted amide containing building blocks

Recently, we developed a new model to study the conversion of *cis-trans* amide conformation in peptide structures.[7] This approach involves the incorporation of aziridine building blocks into peptide sequence. It has been shown that a tilted amide exists in the ground state of the structure by X-ray diffraction and ^{13}C-NMR investigation. Recently, several groups reported that aziridine-containing peptides were effective cysteine protease inhibitors.[8] The following compounds (**Scheme 5**) have been prepared and are currently being tested for bioactivity:

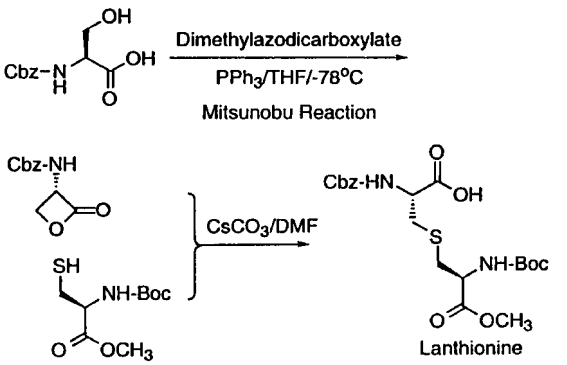
Scheme 5



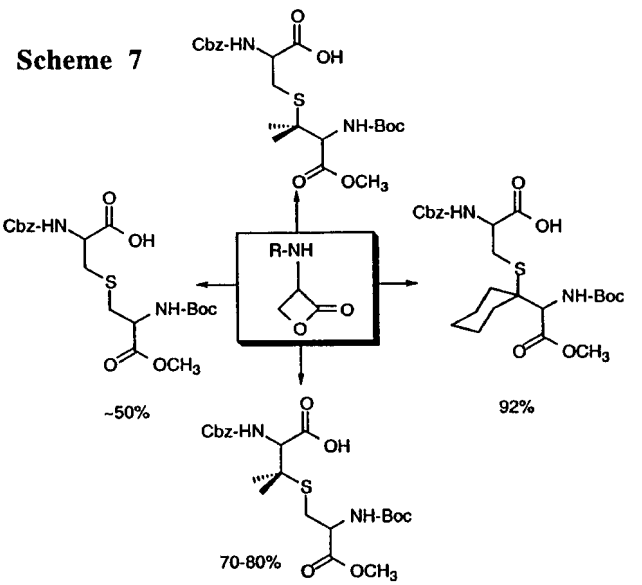
6. Stereoisomeric synthesis of orthogonally protected lanthionine building blocks

Lanthionine is an unusual amino acid composed of two alanine-like residues bridged by a sulfide bond. This analog of cystine with a monosulfide structure is an important building block in natural biologically active peptides called "lantibiotics".[9] It can also be used as a novel conformationally constrained peptidomimetic structure in drug design. To date, only few routes leading to orthogonally protected lanthionines have been reported . We have developed a most efficient strategy to generate lanthionines by regioselective ring opening of *N*-benzyloxycarbonyl(Cbz)-serine β-lactone with cysteine derivatives in the presence of Cs_2CO_3 in DMF (**Scheme 6**).[10]

Scheme 6

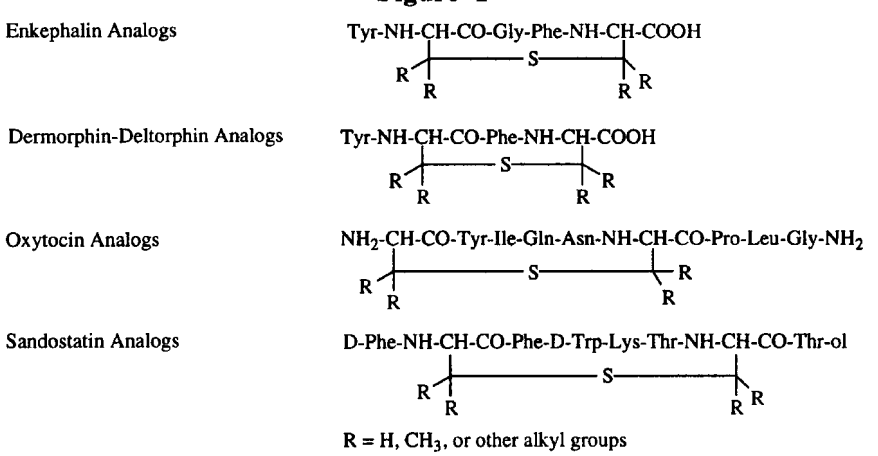


Scheme 7



In order to constrain the lanthionine bridge in these analogs, a variety of substituents have been incorporated into the structures (**Scheme 7**). These building blocks are very useful in cyclic peptide molecules. Incorporation of the lanthionine building blocks into several bioactive peptides have generated very potent and selective ligands (**Figure 1**) which are very promising candidates as new drug agents.

Figure 1



In conclusion, we have demonstrated new asymmetric and enantioselective routes for the preparation of novel peptidomimetic building blocks. These building blocks can be incorporated into peptidic structures to yield molecules with excellent biological activities. In our continuing research effort, we are actively working on new pharmaceutical agents using the peptidomimetic building blocks obtained by the stereospecific reactions described in this presentation.

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References:

[1] (a) M. Goodman, S. Ro, *Burger's Medicinal Chemistry and Drug Discovery, Vol. 1* (Eds.: M. E. Wollf), John Wiley & Sons, Inc., **1995**, pp. 803- 861; (b) R. Hirschmann, K. C. Nicolaou, S. Pietranico, J. Salvino, E. M. Leahy, P. A. Sprengeler, G. Furst, A. B. Smith, C. D. Strader, M. A. Cascieri, M. R. Candelore, C. Donaldson, W. Vale, L. Maechler, *J. Am. Chem. Soc.* **1992**, *114*, 9217-9218; (c) A. B. Smith, M. C. Guzman, P. A. Sprengeler, T. P. Keenan, R. C. Holcomb, J. L. Wood, P. J. Carroll, R. Hirschmann, *J. Am. Chem. Soc.* **1994**, *116*, 9947-9962; (d) F. Cornille, U. Slomczynska, M. L. Smythe, D.D. Beusen, K. D. Moeller, G. R. Marshall, *J. Am. Chem. Soc.* **1995**, *117*, 909-917; (e) A. Giannis, T. Kolter, *Angew. Chem.* **1993**, *105*, 1303-1326; *Angew. Chem. Int. Ed. Engl.* **1993**, *32*, 1244-1267; (f) J. Gante, *Angew. Chem.* **1994**, *106*, 1780; *Angew. Chem. Int. Ed. Engl.* **1994**, *33*, 1699-1720; (g) V. J. Hruby, A. Gehrig, *Med. Res. Rev.* **1989**, *9*, 343-401.

[2] (a) Walker, M.A.; Heathcock, C.H. *J. Org. Chem.* **1992**, *57*, 5566-5568. (b) Pattenden, G.; Thom, S.M. *Synlett* **1992**, 533-534.

[3] Shao, H.; Zhu, Q.; Goodman, M. *J. Org. Chem.* **1995**, *60*, 790-791.

[4] Gao, Y.; Hanson, R.M.; Klunder, J.M.; Ko, S.Y.; Masamune, H.; Sharpless, K.B. *J. Am. Chem. Soc.* **1987**, *109*, 5765-5780.

[5] (a) *The Biology of Glycoproteins*, Ivatt, R. J. (Ed.), Plenum Press, **1984**, New York, U.S.A. (b) *Peptides and Protein Phosphorylation*, Kemp, B. E. (Ed.), CRC Press, **1990**, 289. (c) Wipf, P. ; Miller, C. P. *J. Org. Chem.* **1993**, *58*, 1575-1578. (d) Riviere, M.; Puzo, G. *Biochemistry* **1992**, *31*, 3575.

[6] For reviews of the AD reactions, see: (a) Kolb, H. C.; VanNieuwenhze, M. S.; Sharpless, K. B. *Chem. Rev.* **1994**, 2483-2547. (b) Berrisford, D. J.; Bolm, C.; Sharpless, K. B. *Angew. Chem. Int. Ed. Engl.* **1995**, *34*, 1059-1070. (c) Johnson, R. A.; Sharpless, K. B. In *Catalytic Asymmetric Synthesis*; Ojima, I., Ed.; VCH Publishers: New York, **1993**, 227-272.

[7] Shao, H.; Jiang, X.; Gantzel, P.; Goodman, M. *Chemistry & Biology*, **1994**, *1*, 231-234.

[8] (a) Korn, A.; Rudolph-Bohner, S.; Moroder, L. *Tetrahedron Lett.* **1994**, *50*, 1717-1730. (b) Martichonok, V.; Plouffe, C.; Storer, A. C.; Menard, R.; Jones, J. B. *J. Med. Chem.* **1995**, *38*, 3078-3085.

[9] (a) *Nisin and Novel Lantibiotics*; Jung, G., Sahl, H.G., Eds.; ESCON Science Publishers B. V.: Leiden, 1988. (b) Gross, E.; Morell, J.L. *J. Am. Chem. Soc.* **1971**, *93*, 4634-4635. (c) Fukase, K.; Wakamiya, T.; Shiba, T. *Bull. Chem. Soc. Jpn.* **1986**, *59*, 2505-2508. (d) Fukase, K.; Kitazawa, M.; Sano, A.; Shimbo, K.; Fijita, H.; Horimoto, S.; Wakamiya, T.; Shiba, T. *Tetrahedron Lett.* **1988**, *29*, 795-798. (e) Gross, E.; Kiltz, H.H.; Nebelin, E. *Hoppe Seyer's Z. Physiol. Chem.* **1973**, *354*, 810-812. (f) Allgaier, H.; Jung, G.; Werner, R.G.; Schneider, U.; Zahner, H. *Angew. Chem. Int. Ed. Engl.* **1985**, *24*, 1051-1053. (g) Allgaier, H.; Jung, G.; Werner, R.G.; Schneider, U.; Zahner, H. *Eur. J. Biochem.* **1986**, *160*, 9-22. (h) Wakamiya, T.; Ueki, Y.; Shiba, T.; Motoki, Y. *Tetrahedron Lett.* **1985**, *26*, 665-668. (i) Shiba, T.; Wakamiya, T.; Fukase, K.; Sano, A.; Shimbo, K.; Ueki, Y. *Biopolymers* **1986**, *25*, 11-19.

[10] (a) Shao, H.; Wang, S. H.; Lee, C.; Ösapay, G.; Goodman, M. *J. Org. Chem.* **1995**, *60*, 2956-2957. (b) Polinsky, A.; Cooney, M.G.; Toy-Palmer, A.; Ösapay, G.; Goodman, M. *J. Med. Chem.* **1992**, *35*, 4185-4194. (c) Ösapay, G.; Goodman, M. *J. Chem. Soc. Chem. Commun.* **1993**, 1599-1600.