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PYROGLUTAMIC ACID DERIVATIVES: BUILDING BLOCKS FOR DRUG DISCOVERY

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Abstract – Substituted prolines namely amino acid proline chimeras, have received a great synthetic efforts with the aim to develop a full library of compounds to be used in bioactive peptides drug discovery. Conversely pyroglutamic acid derivatives, strictly related to proline chimeras have been used only marginally in peptide modifications, probably due the lack of handled asymmetric syntheses. In this work we reviewed the "state of the art" on the synthetic approaches to pyroglutamic acid derivatives bearing a side chain related to that of natural amino acids.

INTRODUCTION

Proline (**1**) is the only natural amino acid with a cyclic structure and a secondary amine. Its peculiar structure gave rise to a series of studies with the aim to understand its importance in the protein folding and structural ligand-receptor recognition. In the past few decades a series of substituted prolines, namely proline-chimeras, have been synthesized as useful tools in drug discovery (Figure 1).

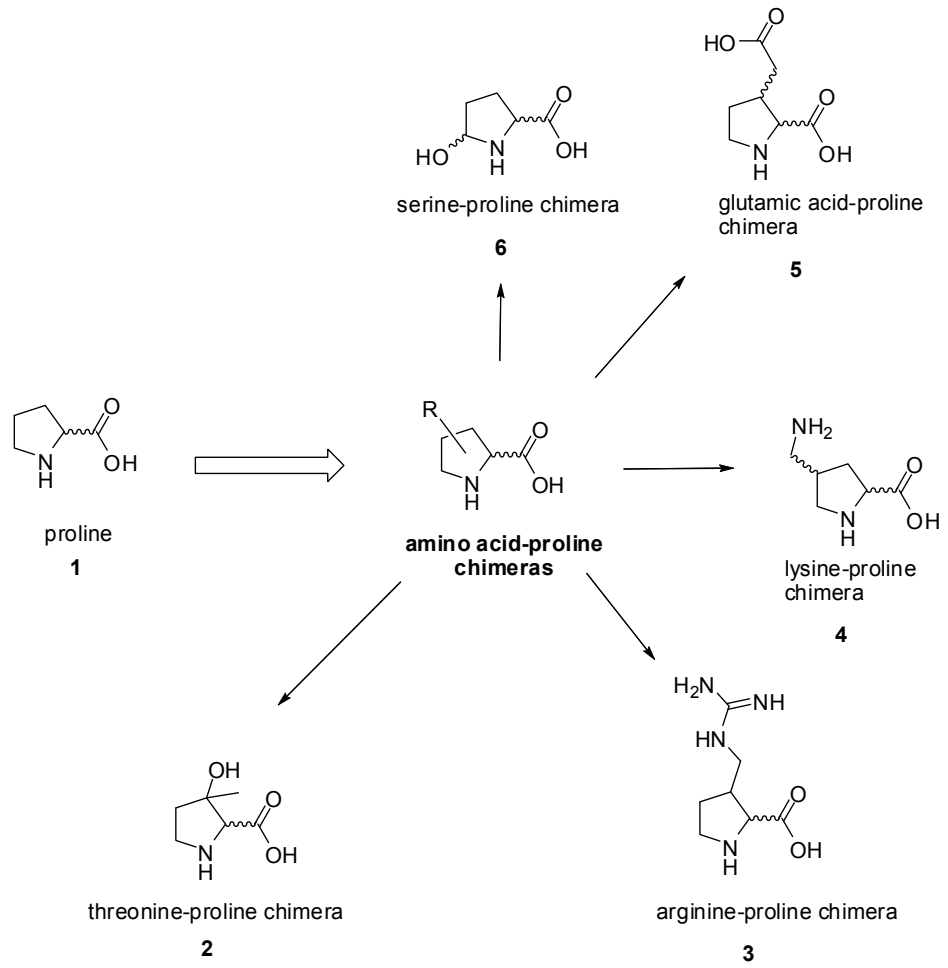


Figure 1. Representative amino acid-proline chimeras (**2-6)**

Proline chimeras (**2-6**) are usually mono-substituted prolines bearing on the pyrrolidine ring a substituent, deeply related to the side chain of the natural occurring amino acids (Figure 1). Once inserted, in place of the native amino acid, into a bio-active peptide, a proline chimera can modify the folding of the peptide, still maintaining the natural side chain important for recognition. It has been demonstrated that a proline residue can induce a β-turn folding in the peptide backbone which is an important secondary structure often presents in bioactive peptides.[1] Another important features of proline-chimeras is that the substituent on the pyrrolidine group is a mimetic of the side chain of other amino acids, but the rotation on the χ^1 and χ^2 angles results heavily restricted.[1] This restriction in the conformational freedom could deeply change the bio-activity of the peptide, ranging from super-potent agonists to antagonists or other relevant changes in bio-activity. Pyroglutamic acid (**8**) is structurally linked to proline and substituted pyroglutamic acids can be easily inserted into a bio-active peptides backbone, similarly to proline-chimeras, in order to modify their secondary structure and bias the interaction with the biological target (Figure 2).[1]

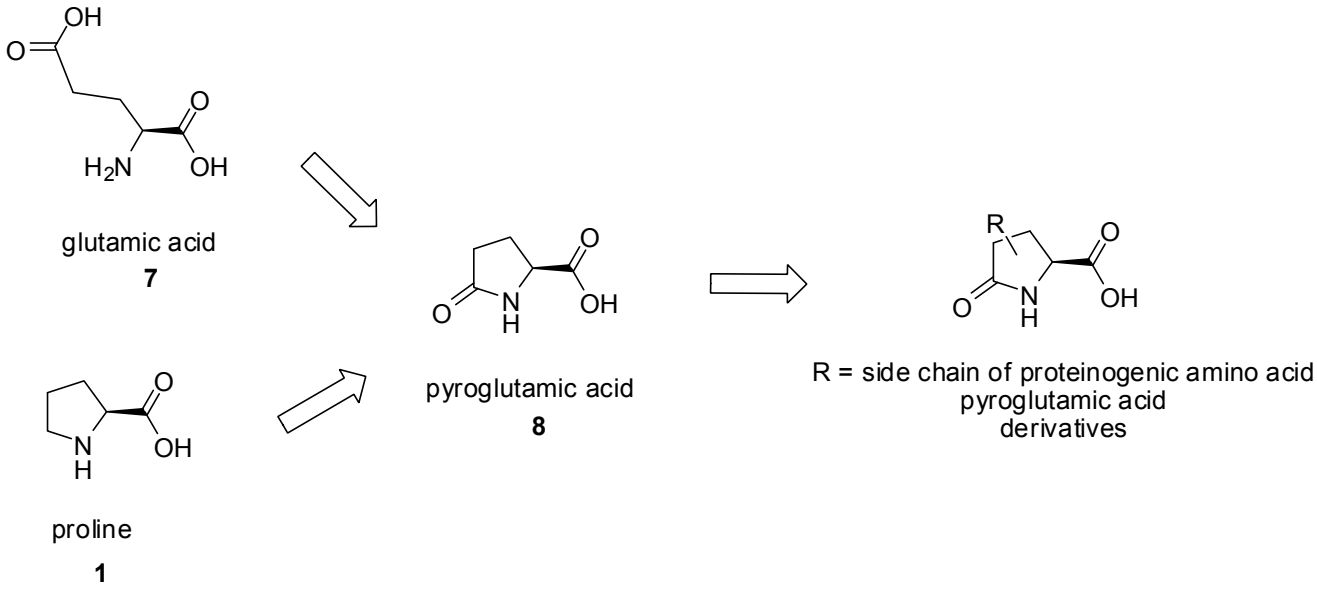


Figure 2. Schematic representation of pyroglutamic acid structure (**8**) and its derivatives

The use of substituted pyroglutamic acids as surrogate of natural occurring amino acids is less explored. In a recent paper Tsai *et al.* [1f] reported the use of substituted pyroglutamate derivatives in the design and SAR studies of Fibloblast activation protein (FAP) inhibitors. FAP is a serine protease that cleaves bioactive peptides preferentially after proline residues being expressed in near the 90% of epithelial cancers[1g] it has been suggested that FAP promotes tumor genesis, and its inhibition may attenuate the tumor growth. The design of novel petidomimetics, introduces a constraint in the P2 portion of lead IG-409, the conversion of the P2 side secondary amine to amide and a series of modification at the pyrrolidine *C* terminal, as show in Figure 3. Modifications on the amine substituents in position 4 and at the carbonylmethyl group lead to the discover of potent pyroglutamic acid-based FAP inhibitors with high selectivity for FAP over DPP-IV, DPP-II, DPP8 and DPP9.

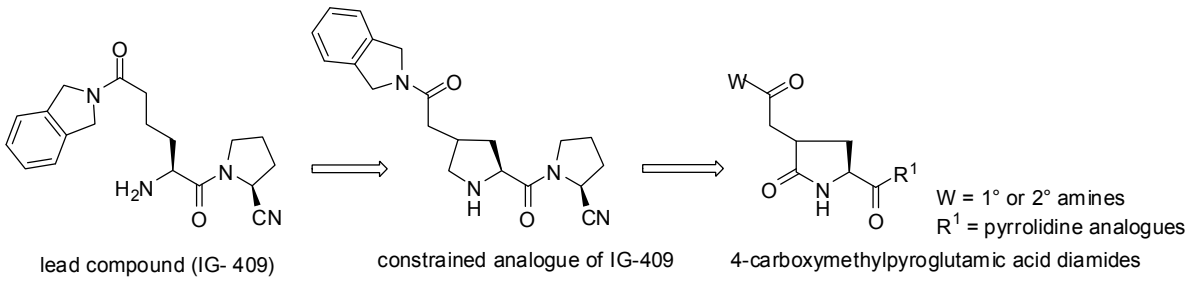


Figure 3. Design of novel 4-substituted pyroglutamic acids as FAP inhibitors

Pyroglutamic acid **(8)** has been also widely used as chiral synthon for the preparation of a series of chiral bio-active compounds, as chiral auxiliary in asymmetric synthesis,[2] and as precursor for other amino acids,[3] such as α-amino acids and conformationally constrained glutamic acid, alanine, lysine and ornithine derivatives.[4,5] A recent review by Panday *et al.*[4] reported a series of synthesis where pyroglutamic (8) acid was used as chiral synthon. Some derivatives have shown biological activity, as inhibitors of the vascular cell adhesion molecule-1 (VCAM-1) and integrin interactions, which makes them particularly useful for treating rheumatoid arthritis and asthma.[6]

Both *S* and *R*-pyroglutamic acids are commercially available in optically pure form: *S*-pyroglutamic derivatives have been used to resolve racemic mixtures by forming diastereomeric species,[7] or as substrate of enantioselective enzymatic reactions.[8]

Parrish *et al.*[9] reported a series of pyroglutamic acid derivatives, easily prepared from the pyroglutamic diketopiperazines. Pyroglutamic containing DKPs undergo to a ring opening by reaction with diamines, in such way substituted pyroglutamic acids have also been synthesized.[10]

Numerous approaches to symmetric and asymmetric functionalized pyroglutamic acid derivatives have been reported, although each presents some limitations.[11,12] One common method to obtain these functionalized pyroglutamates is the alkylation of the lithium enolate derived from *N*-protected pyroglutamic esters to give C4 substituted derivatives.[13-17] This method works well for reactive electrophiles such as allylic halides and aryl aldehydes, with moderate yields and stereospecificity. Ohta *et al.*[18] reported a hydroxylation of the lithium enolate derived from benzyl *N*-Boc-(*L*)-pyroglutamate with 3-phenyl-2-toluenesulfonyl-1,2-oxaziridine to give *trans*-4-hydroxypyroglutamate.[19] Recently, Merino *et al.* reported an asymmetric synthesis of 4-hydroxypyroglutamic acid, involving a dipolar-cycloaddition of a nitrone with acrylamide derived from Oppolzer's sultam.[20]

Functionalized pyroglutamic acid derivatives have been also prepared using the Michael reaction of amide-tethered di-acids with alkynones.[21] One practical synthetic pathway is the homologation of nucleophilic glycine-equivalents for the synthesis of sterically constrained 3-substituted pyroglutamic acids, thus increasing the synthetic efficiency of the Michael addition reactions.[22]

In this review, various methods both asymmetric and symmetric, for the synthesis of pyroglutamic amino acid-chimeras are described (Figure 4).

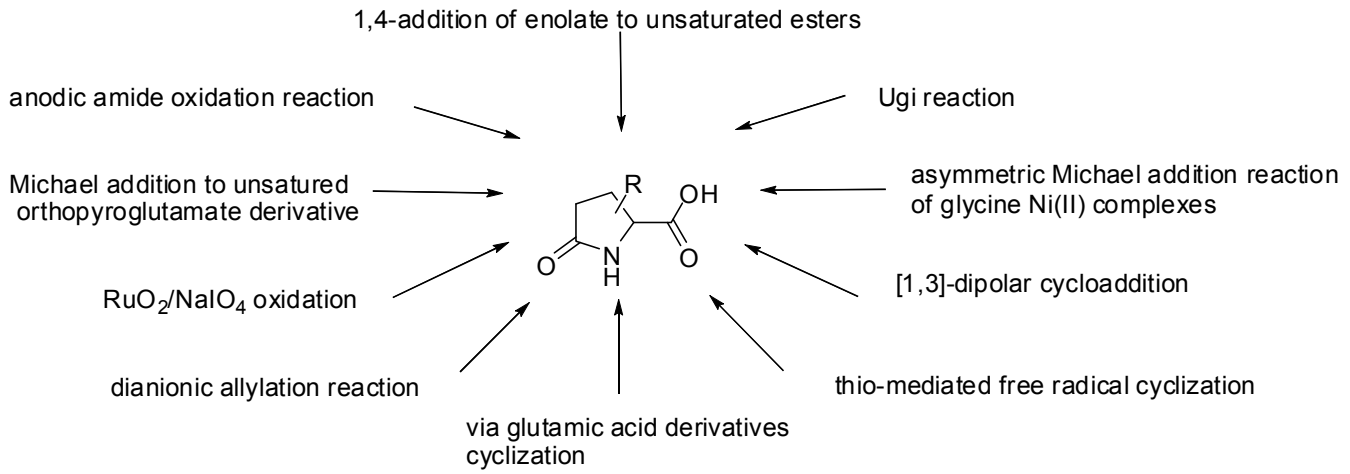


Figure 4. Schematic representation of synthetic approaches for pyroglutamic acid chimeras

Each paragraph has been named on the basis of the position on the pyrrolidine ring reporting side chain fragments of proteinogenic amino acids when present. Taking into account the presence of a review on pyroglutamic acid derivatives,[23] we have focused our attention on the chemical strategies contributions of the literature from 2000 to 2014.

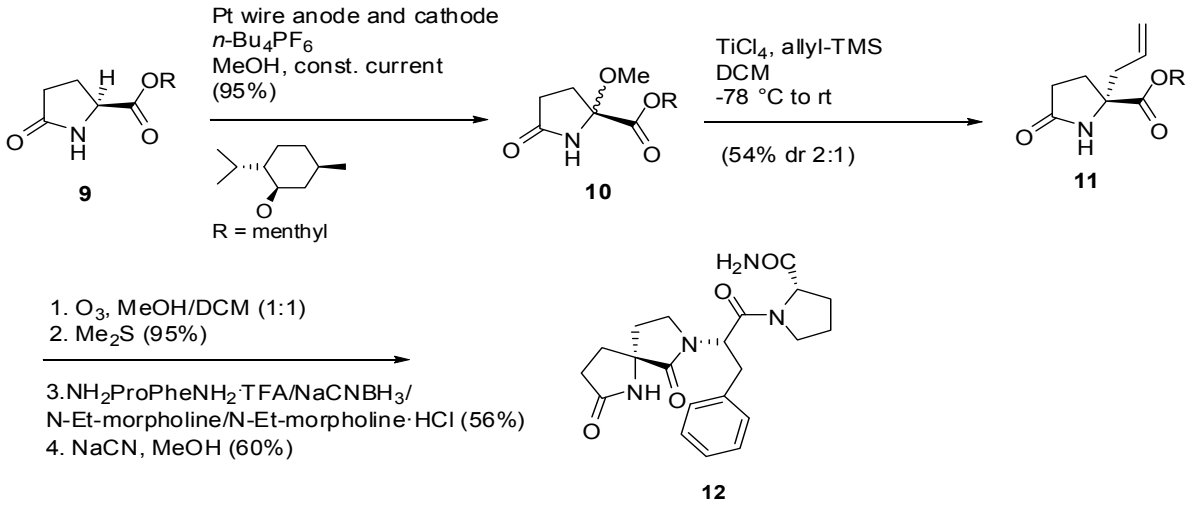
2. SYNTHESES OF PYROGLUTAMIC ACID DERIVATIVES

2.1 α-SUBSTITUTED PYROGLUTAMIC ACID DERIVATIVES

2.1.1 ANODIC AMIDE OXIDATION REACTION

Simpson *et al.*[24] realized the synthesis of a series of TRH analogues. Initially, the strategy utilized allowed to reach the synthesis in six steps from pyroglutamic acid. A revised strategy for synthesizing the spirocyclic lactam ring skeletons was then proposed (Scheme 1).

The key step was the conversion of the 5-allylpyroglutamate derivative **11** into an advanced intermediate that could be used to build the desired analogue **12**. This would be accomplished by using the olefin of the allyl group to set up either an intramolecular reductive amination reaction or an intramolecular Mitsunobu reaction. Thus, both the intermolecular reductive amination and the diketopiperazide formation would be avoided. The allyl-substituted pyroglutamate intermediate **11** was made by first functionalizing menthyl pyroglutamate **9** with the use of an anodic amide oxidation reaction,[25,26] and then treating the resulting product **10** with allylsilane and $TiCl_4$ to afford a mixture of stereoisomers allylpyroglutamate derivatives. Crystallization led to **11** in a 54% isolated yield.

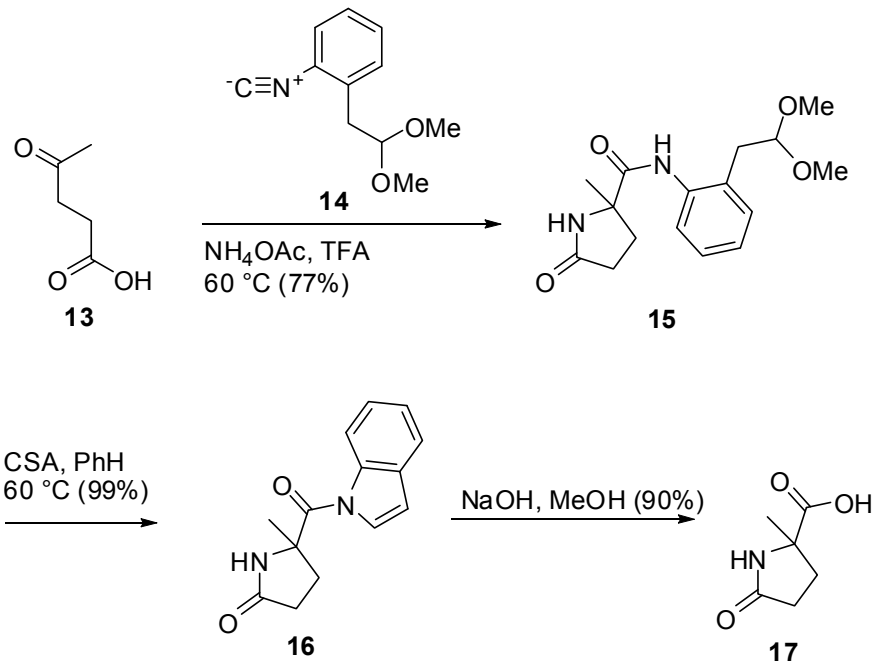


Scheme 1. 2-Substituted-allylpyroglutamic acid synthesized by Simpson *et al.*[24]

2.1.2 UGI REACTION

For the synthesis of quaternary (α-substituted) pyroglutamic acid, the Ugi multicomponent reaction (U4C-3CR),[27,28] was conducted on β-keto acid to give an amide derivative. The hydrolysis of the amide derivative is a limiting step.

Recently Isaacson *et al.*[27] modified the procedure using a convertible isonitrile, 1-isocyano-2-(2,2-dimethoxyethyl)benzene (**14**) in the Ugi reaction, which consents a mild selective cleavage of the resulting *C*-terminal amide. The unprotected α-methylpyroglutamic acid **17**, was thus obtained starting from levulinic acid (**13**) (Scheme 2).[28-30]



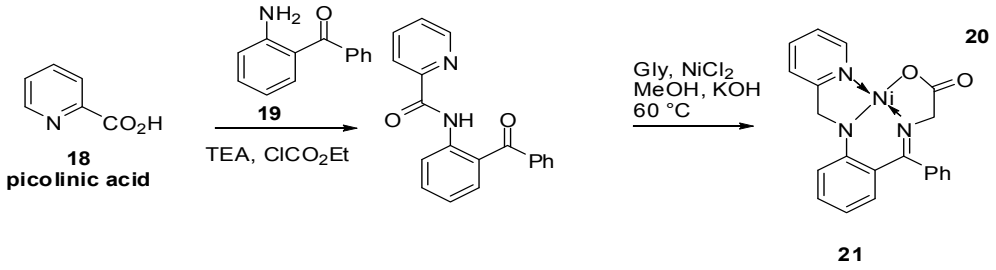
Scheme 2. Synthesis of 2-methyl-pyroglutamic acid derivative by Isaacson *et al.*[27]

2.2 3-SUBSTITUTED PYROGLUTAMIC ACID DERIVATIVES SYNTHESES

2.2.1 ASYMMETRIC MICHAEL ADDITION REACTION OF GLYCINE NI(II) COMPLEXES

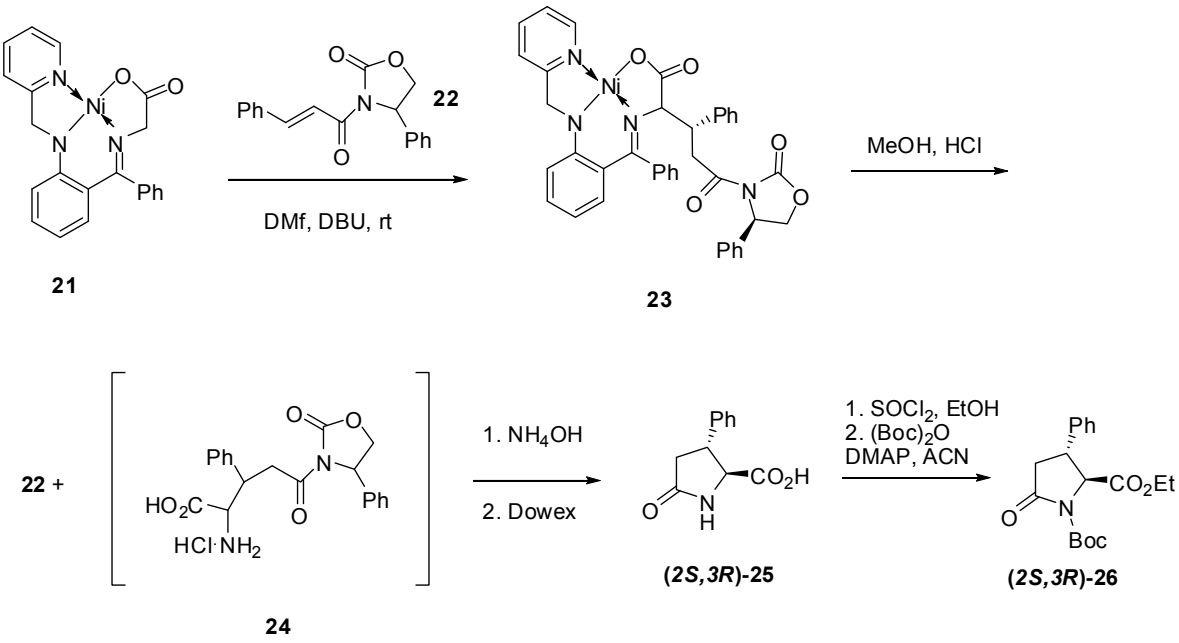
Ellis *et al.*[31,32] recently reported the first practical synthesis of symmetrically (*R,R*)-dialkyl-substituted amino acids, including 2-aminoindane-2-carboxylic acid, using complex **20** as a stable, but still reactive glycine equivalent. Belokon's group described the catalytic asymmetric alkylation of the glycine equivalent **20** under phase-transfer conditions.[33] The Ni(II)-complex, was readily prepared on a multigrams scale under inexpensive conditions (Scheme 3).[34]

First was studied the addition between the complex **21** and oxazolidin-2-one carried out under standard reaction conditions.



Scheme 3. Generation of the Ni complex[34]

The reaction occurred at a substantially high rate and the sole diastereomeric product obtained was decomposed without any purification, to afford the corresponding enantiomerically pure pyroglutamic acid (2*S*,3*R*)-25 (Scheme 4).[35]

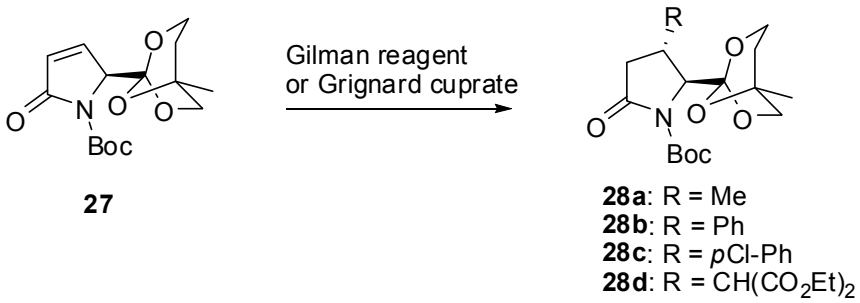


Scheme 4. Synthesis of 3-substituted Phe-pyroglutamic acid-chimera described by Soloshonok *et al.*[35]

Transformation of **25** to ethyl ester by thionyl chloride in ethanol, followed by protection with Boc groups produced the *N*-Boc-3-arylpyroglutamate **26** in good yields (75-96%).

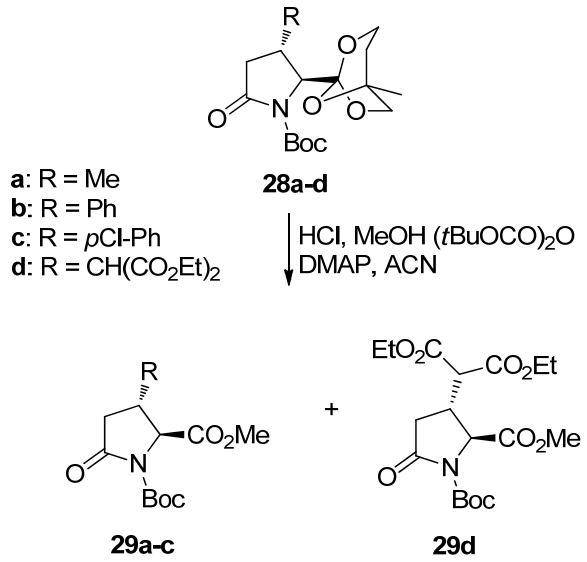
2.2.2. MICHAEL ADDITION TO UNSATURATED ORTHOPYROGLUTAMATE DERIVATIVES

Oba *et al.*[36] described a chiral synthesis of 3-substitued pyroglutamate derivatives via Michael addition using ABO (2,7,8-trioxabicyclo[3.2.1]octane) as *C*-terminal protecting group. Treatment of Michael acceptor **27** with Gilman reagent, (prepared from MeLi and CuI), gave derivative **28a** in 90% yield (Scheme 5).



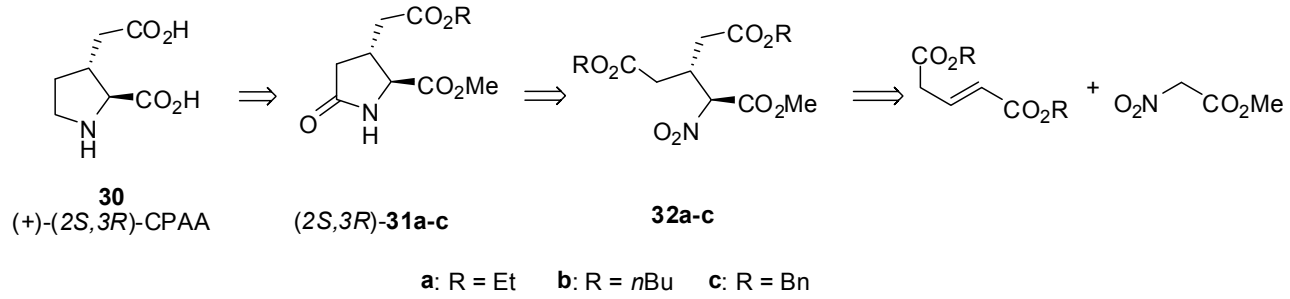
Scheme 5. Michael addition to unsaturated orthopyroglutamate proposed by Oba *et al.*[36]

Similar reactions with Grignard-cuprates readily produced the derivatives **28b** and **28c** (71% and 68% respectively). The addition of sodium diethyl malonate afforded the Michael adduct **28d** (quantitative). The ABO ester was converted to methyl ester by acid methanolysis followed by reprotection of NH group by Boc group furnishing substituted pyroglutamate derivatives **29a-d** in good yields (Scheme 6).



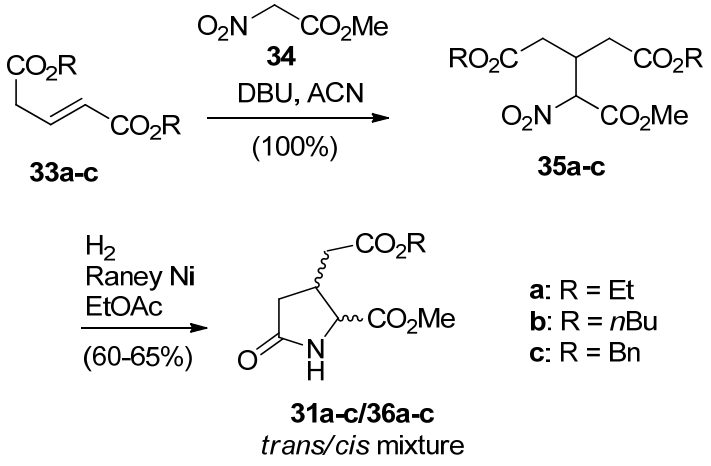
Scheme 6. Synthesis of 3-substituted pyroglutamic acid-proline chimeras[36]

Felluga *et al.*[37] synthesized the nucleus of kainoid amino acids by a chemoenzymatic process, exploiting the diastereomeric *cis/trans* methyl pyroglutamate derivatives as key intermediates. Compounds **31a-c** are accessible by cyclization of intermediates **32a-c** obtained from conjugate addition of methyl nitroacetates to the appropriate glutaconic diesters and are ready to enzymatic kinetic resolution (Scheme 7).



Scheme 7. Retrosynthetic approach for (+)-CPAA reported by Felluga *et al.*[37]

The *trans* pyroglutamic acid derivatives **31a-c** were prepared in their racemic form by DBU mediated addition of methyl nitroacetate **34** to the appropriate glutaconic diesters **33a-c**. The reaction furnished the corresponding nitrotriesters **35a-c**. The nitro group of **35a-c** was reduced with Raney nickel following by a spontaneous cyclization to form the lactams **31a-c** and **36a-c**. Unfortunately, this step is not completely diastereoselective and led to *cis/trans* mixtures of **31a-c** and **36a-c** in 9:1 and 4:1 ratio respectively (Scheme 8).

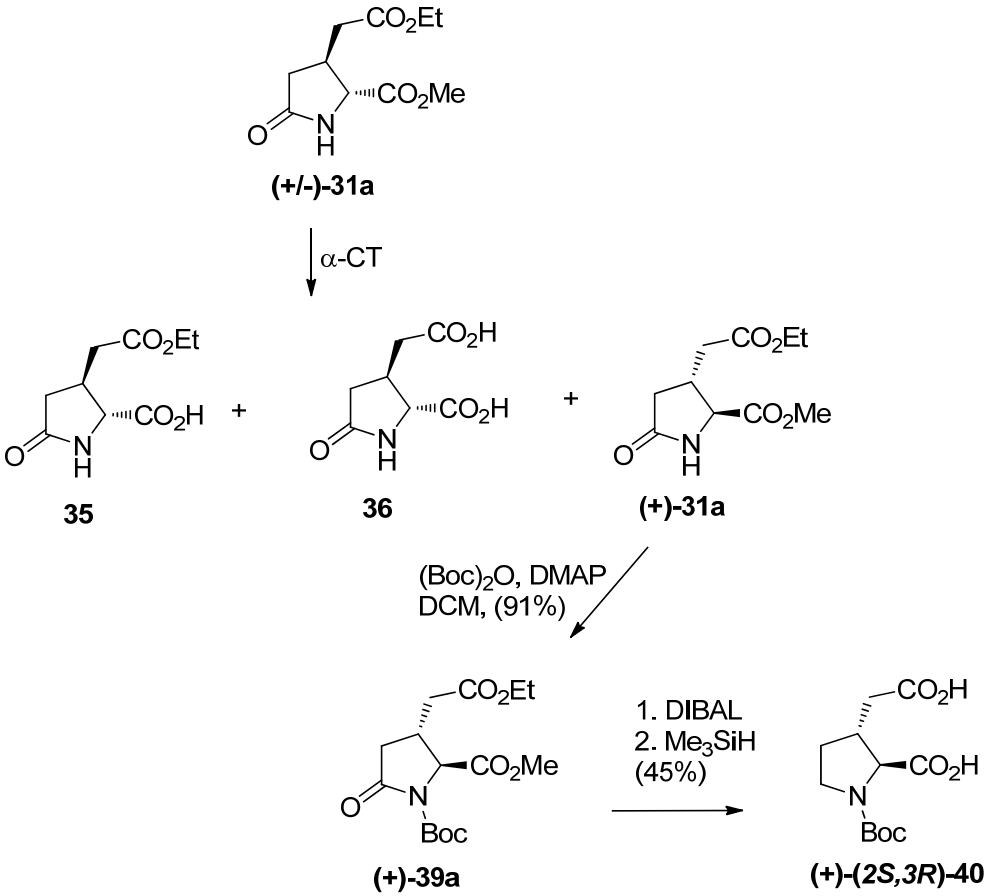


Scheme 8. Synthesis of the racemic lactams as diastereomeric mixture[37]

The *trans* and *cis* diastereoisomers were unseparable and each mixture was subjected to enzymatic resolution with alpha-chymotrypsin *(*α-CT). When the hydrolysis reaction was allowed to proceed to 80%

conversion, the unreacted diesters (+)-**31a-c** were isolated with 99% ee in the case of (+)-**31a,b** and 95% for (+)-**31c**.

Protection of the amidic nitrogen with Boc group gave compound (+)-**39**, which after selective and exhaustive lactam carbonyl reduction, afforded the proline derivative (+)-(2S,3R)-**40**, having a positive optical rotation, in accordance with the data reported for the natural NMDA agonist (Scheme 9).



Scheme 9. Enzymatic hydrolysis and transformation to pyroglutamate derivative[37]

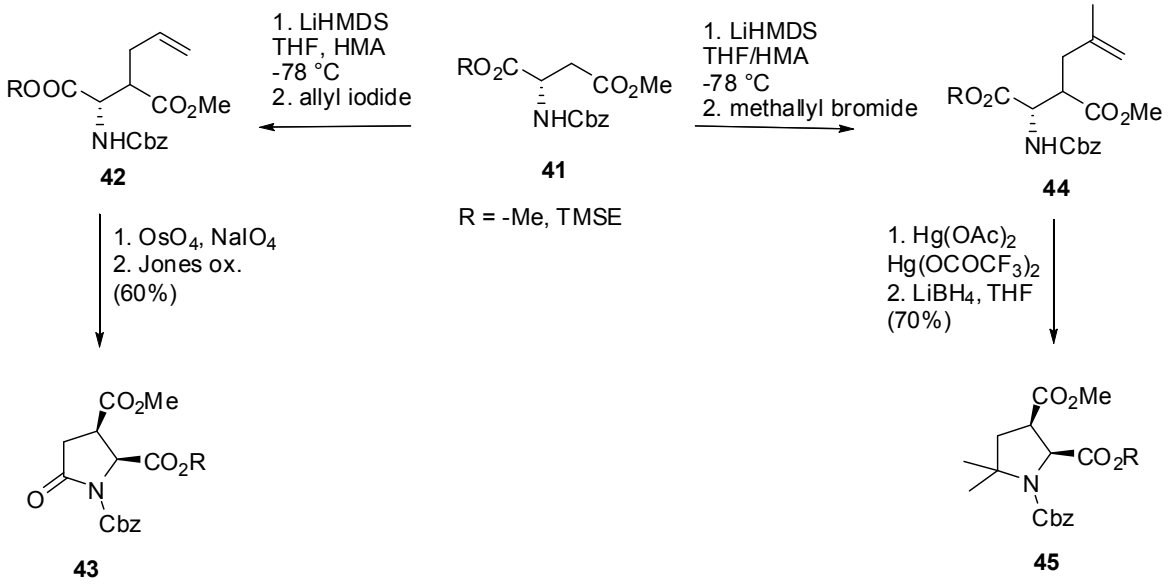
2.2.3 DIANIONIC ALLYLATION REACTION

A useful synthesis of 3-substituted pyrrolidines and pyrrolidinones, was achieved by the modification of the γ-unsaturated side-chain in allylated derivatives.[38,39]

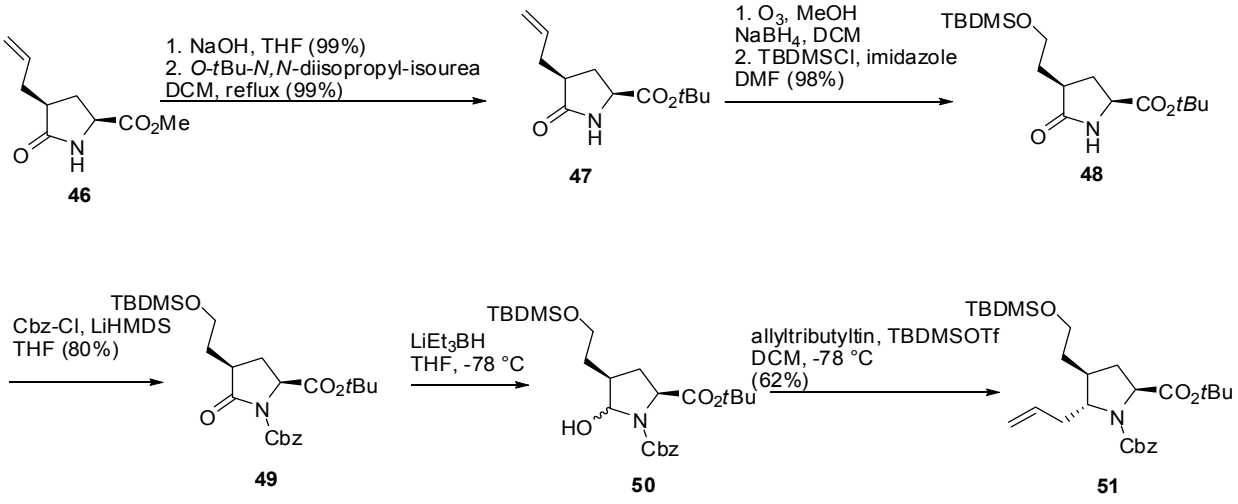
Dihydroxylation and oxidative cleavage of the ester **41** gave the pyrrolidinone **43** (60% overall yield) (Scheme 10).

Cyclization of **44** in the presence of $Hg(OAc)_2$ followed by demercuration led to the 5,5-dimethyl-pyrrolidine diester **45** in 70% overall yield.[41] An alternative route presents the ozonolytic cleavage of the allyl group in **44**, followed by hydrogenation, leading to the *des*-Cbz analogue of **45**.[42,43]

This synthetic pathway was also used by Artale *et al.*[44] for the synthesis of 4-allylpyroglutamic acid **46** promptly achieveable from glutamic acid (Scheme 11).



Scheme 10. Hanessian's procedure for the synthesis of 2,3-*cis*-substituted pyrrolidines and pyrrolidinones[40]



Scheme 11. Polysubstituted proline chimera from pyroglutamic acid derivative reported by Artale *et al.*[44]

Methyl ester **46** was converted into *tert*-butyl ester **47**,[45] ozonolysis of **47** and treatment with $NaBH_4$ gave the corresponding alcohol, which was protected as a *tert*-butyldimethylsilyl ether **48** in 78% yield. The resulting lactam was *N*-protected affording **49** in 80% yield suitable for peptide synthesis.[46]

2.3 4-SUBSTITUTED PYROGLUTAMIC ACID DERIVATIVES SYNTHESES

2.3.1 $RuO_2/NaIO_4$ OXIDATION

The 4-hydroxypyroglutamic acid is strictly related to 4-OH-proline and it has been widely used as chiral starting material or chiral auxiliary for asymmetric syntheses of γ-substituted glutamic acid (Figure 5).[16]

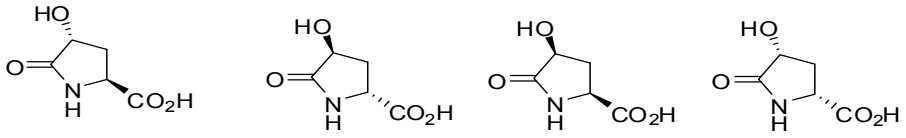
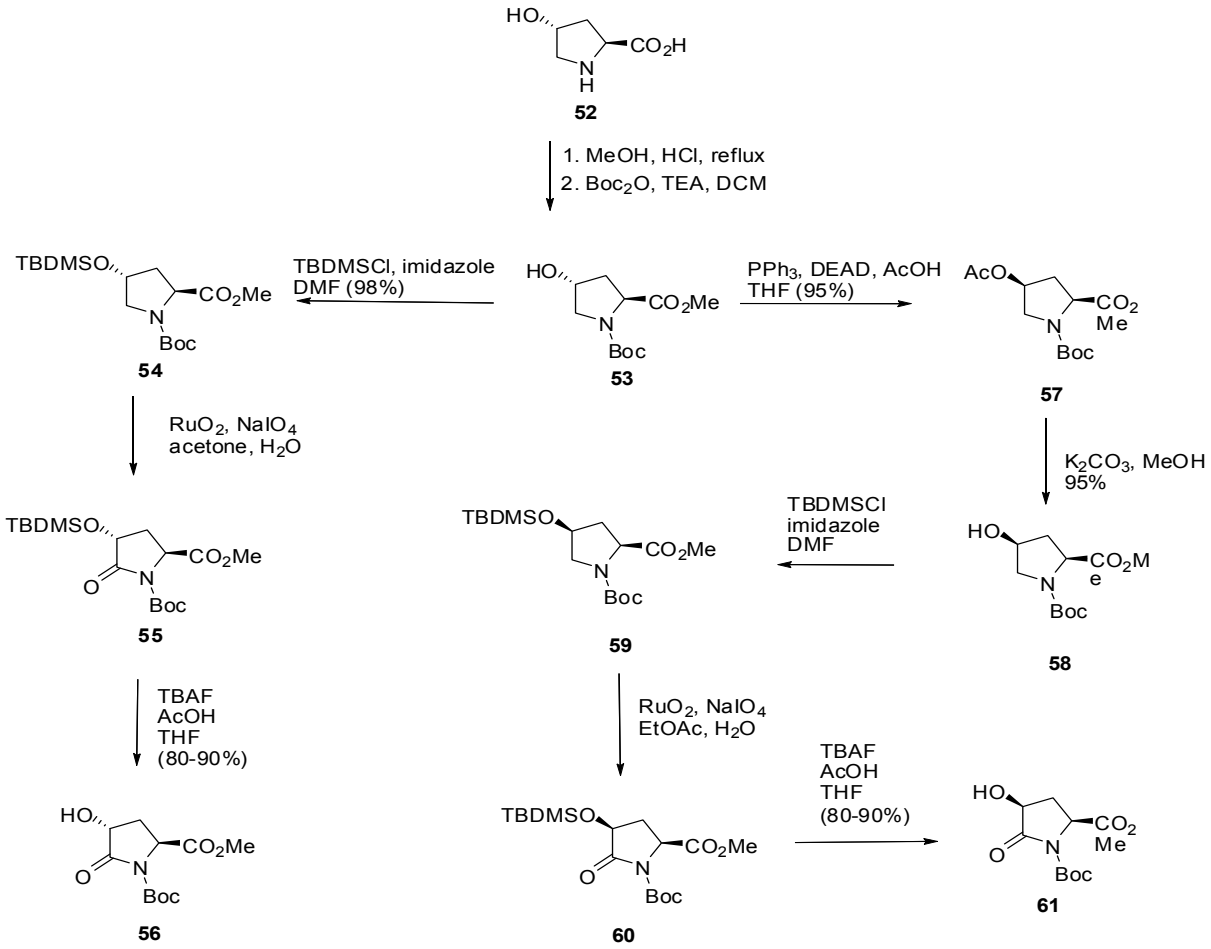


Figure 5. Different types of 4-hydroxypyroglutamic acids

Zhang *et al.*[47] reported an efficient route to chiral pure methyl *N*-Boc-4-hydroxypyroglutamates and their derivatives, starting from the commercially available (4*R*)-hydroxyproline 37 (Scheme 12).



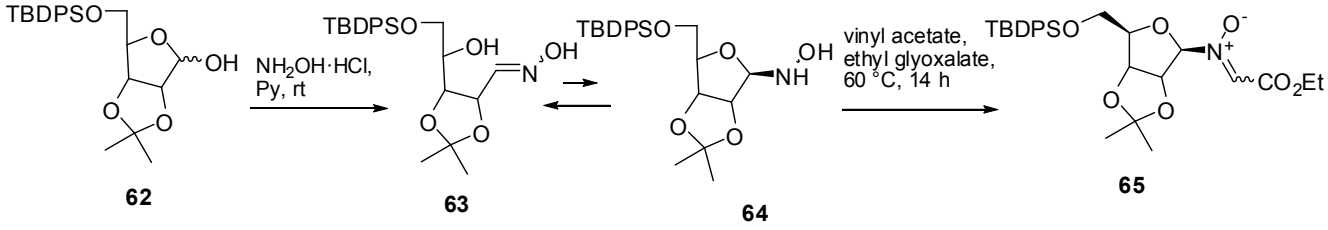
Scheme 12. Preparation of *cis/trans*-Boc-4-hydroxypyroglutamates[47]

The key step is a $RuO_2/NaIO_4$ oxidation of the opportunely protected 4-hydroxyproline.[48,49] This oxidation has been previously reported by Yoshifuji *et al.*[50,51] for the conversion of cyclic α–aminoacids to α-aminodicarboxylic acids. Application of this protocol to **54** and **57**, respectively *N*-Boc-4-silyloxy and 4-acetoxyprotected proline derivatives led to the two diasteromers **55** and **60**. At the beginning, (4*R*)-Hydroxyproline (**52**) was esterificated and *N*-Boc protected to give **53** in quantitative yield.[52]

At this point, in order to obtain the two diasteromeric pyroglutamic acid derivatives, two different pathways have been used. For 4-(*R*)-*trans*-*N*-Boc-4-hydroxylpyroglutamate, OH group of **53** was protected by TBDMS (**54**). Compound **54** was oxidized by $RuO_2/NaIO_4$ in $EtOAc/H_2O$ at rt to gave **55**. The TBDMS protecting group was removed with TBAF to give methyl (4*R*)-*trans*-*N*-Boc-4-hydroxyl- pyroglutamate (**56**) in 80% yield. For (4*S*)-*cis*-*N*-Boc-4-hydroxylpyroglutamate **61**, a two-step Mitsunobu inversion on C4 was conducted. The protection of **58** as silyl ether **59**, oxidation of **59** with $RuO_2/NaIO_4$ and deprotection of **60** with TBAF, gave rise to methyl (4*S*)-*cis*-*N*-Boc-4-hydroxyl- pyroglutamate (**61**) in very good yield.

2.3.2 [1,3]-DIPOLAR CYCLOADDITION

Particularly appealing is the approach based on nitrones reactivity.[53]

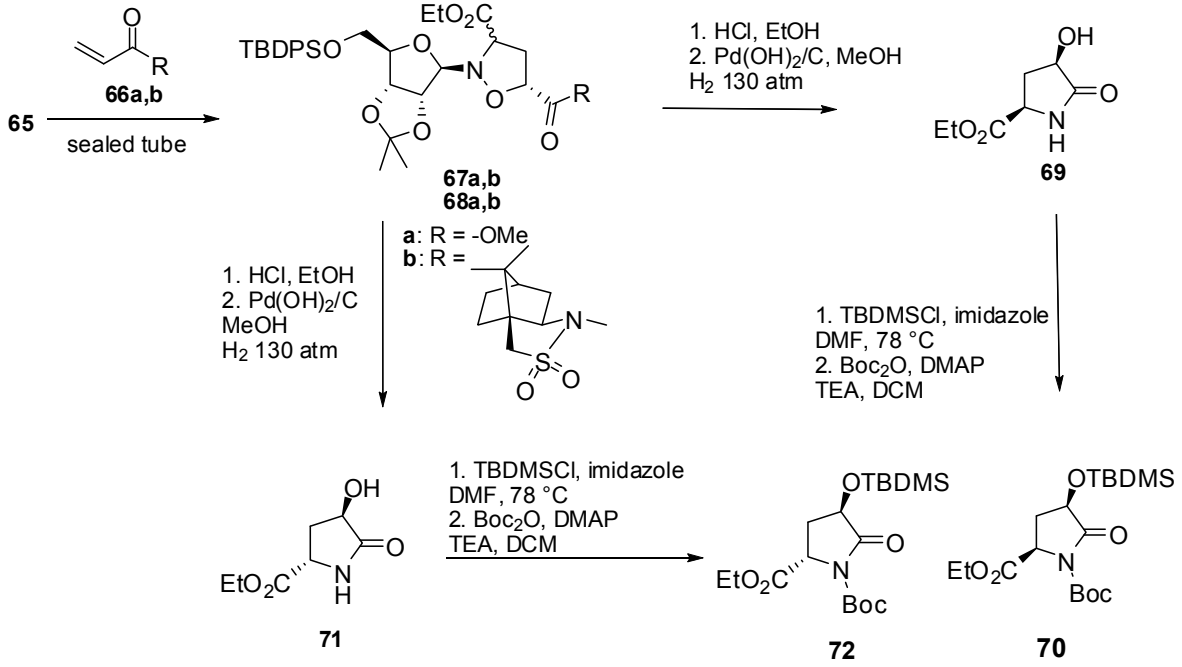


Scheme 13. Synthesis of the substituted nitrone **65** as intermediate for the preparation of *cis* and *trans* 4-hydroxypyroglutamic acid derivatives

In situ prepared nitrone **65**,[55] (Scheme 13) was reacted with methyl acrylate **66a** in a sealed tube overnight. Two intermediates **67a** and **68a** (d.r. 2:1), were obtained. A reaction carried out with Oppolzer's sultam derived acrylamide **66b**,[56] gave the **67b**:**67b** ratio increased to 20:1 with *trans* adducts observed preferentially.

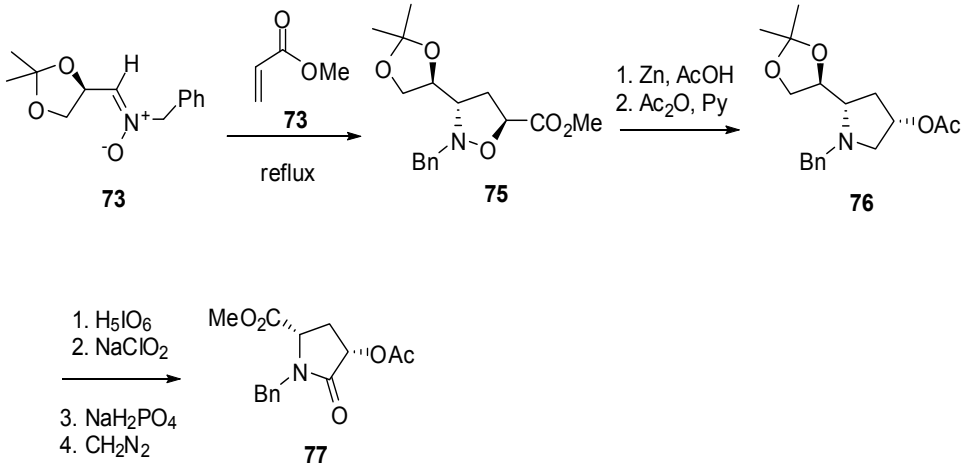
The Conversion of intermediates **67a,b**, **68a,b** to 4-hydroxy-*D*-pyroglutamic acid derivatives consisted in four sequential step: (*i*) The sugar was eliminated by acid treatment; (*ii*) *N–O* cleavage by hydrogenolysis gave unprotected ethyl 4-hydroxy-*D*-pyroglutamates **69** and **71**; (*iii*) i*n situ* protection of OH group with

TBDMS; (*iv*) NH protection with Boc group to give the desired compounds **70** (D-pyroglutamate derivative) and **72** (in 1:20 ratio). (Scheme 14)



Scheme 14. Preparation of *cis* and *trans* 4-hydroxypyroglutamic acid derivatives via nitrone chemistry[54]

On the basis of these results, Merino *et al.*[57] reported the first enantioselective synthesis of all the isomers of 4-hydroxypyroglutamic acid, using Oppolzer's sultam as a chiral auxiliary (Scheme 15).



Scheme 15. Preparation of *cis*-4-hydroxypyroglutamic acid derivatives via nitrone[57]

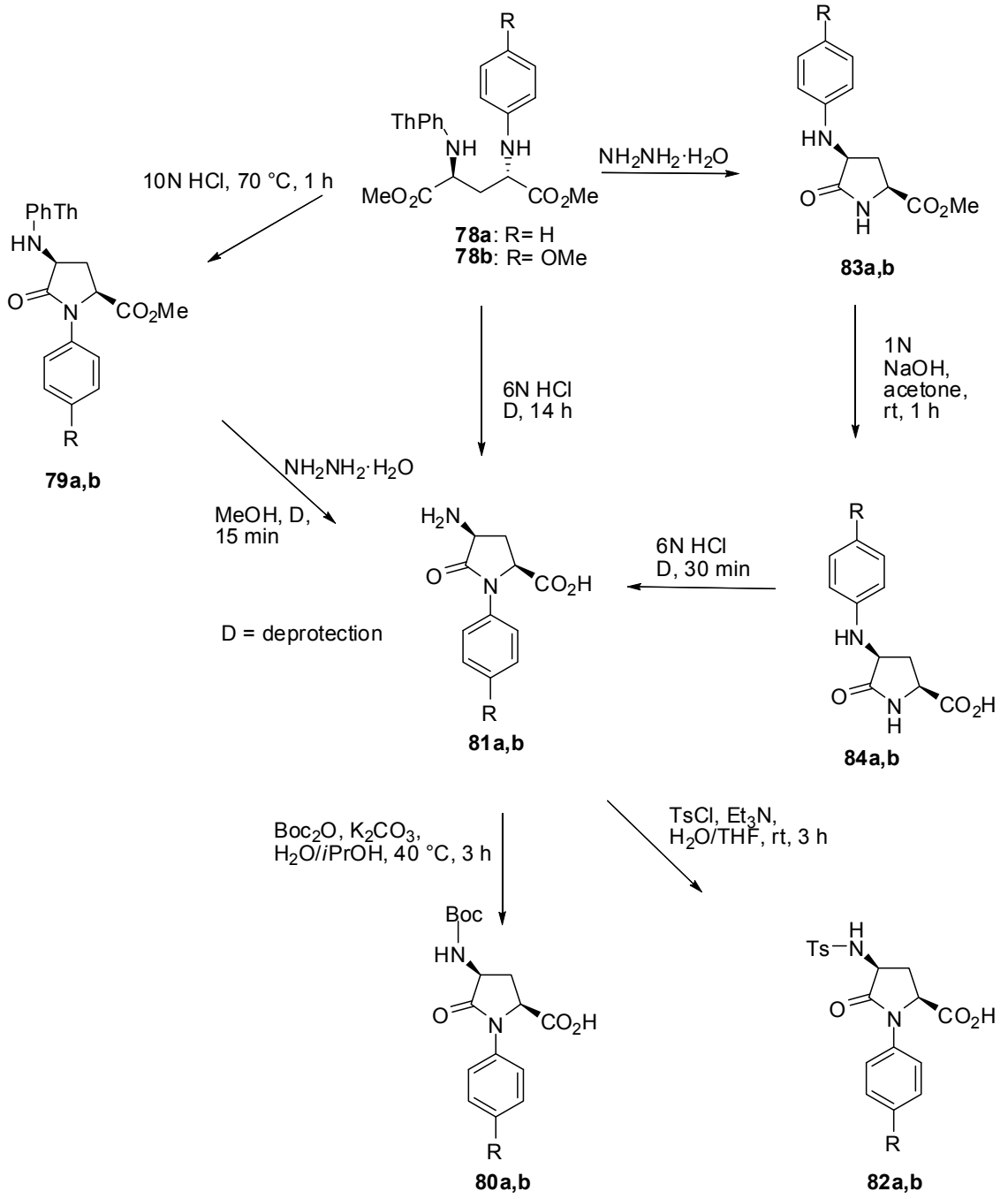
Nitrone **73** and acrylate **74** were used as starting materials. Reduction and acetylation of **75** gave the intermediate **76**. Periodic acid and sodium ipochloride converted the dioxolane ring into the carboxylic functionality (**77**).[57] All of the possible isomers of 4-hydroxypyroglutamic acid can be prepared by this method. Thus, this also served to confirm previous configurational assignments. The orthogonal protection of compound **77** allows its use as building blocks in peptide chemistry.

2.3.3 VIA 4-PROTECTED AMINO-GLUTAMIC ACID DERIVATIVES CYCLIZATION

Recently, Krasnov *et al.*[58] explored the reactivity of γ-protected amino-glutamic acid dimethyl esters (**78a,b**) (Scheme 16). This study revealed an efficient synthetic approach for producing various substituted pyroglutamate derivatives and, after reduction, of proline derivatives. Derivatives of 4-amino-5-oxophenylproline, such as 4-tosylamino- (**82a,b**), 4-phthalimido- (**79a,b**) and 4-Boc-amino-(2S,4S)-5oxophenylprolines (**79a,b**) were prepared starting from dimethyl (2S,4S)-4-phenylamino- (**78a**) and (2S,4S)-4-(4-methoxyphenyl)amino-*N*-phthaloylglutamates [59] (**78b**), by hydrolysis in different conditions. Treatment with 10N HCl for 1 h at 70 °C resulted in the selective hydrolysis of ester group with the formation of compounds **79a** and **79b** respectively. Acids **79a** and **79b** were isolated and crystallized. The hydrolysis of compounds **78a, b** in prolonged (12-14 h) refluxing 6N HCl leads to compounds **81a** and **81b** which posses the free 4-amino group and the protected N^{α} amino group. Removal of the phtaloyl group of compounds **79a** and **79b** by hydrazinolysis, under mild condition resulted in compounds **81a** and **81b**. Synthesis of 4-substituted pyroglutamic acids **84a** and **84b** was achieved by hydrazinolysis of compounds **78a** and **78b** and heating in vacuo to have the full conversion into the 4-aminosubstituted pyroglutamates **83a**, **83b**. Hydrolysis of the ester group by 1N NaOH in acetone yielded pyroglutamic acids **84a** and **84b**.

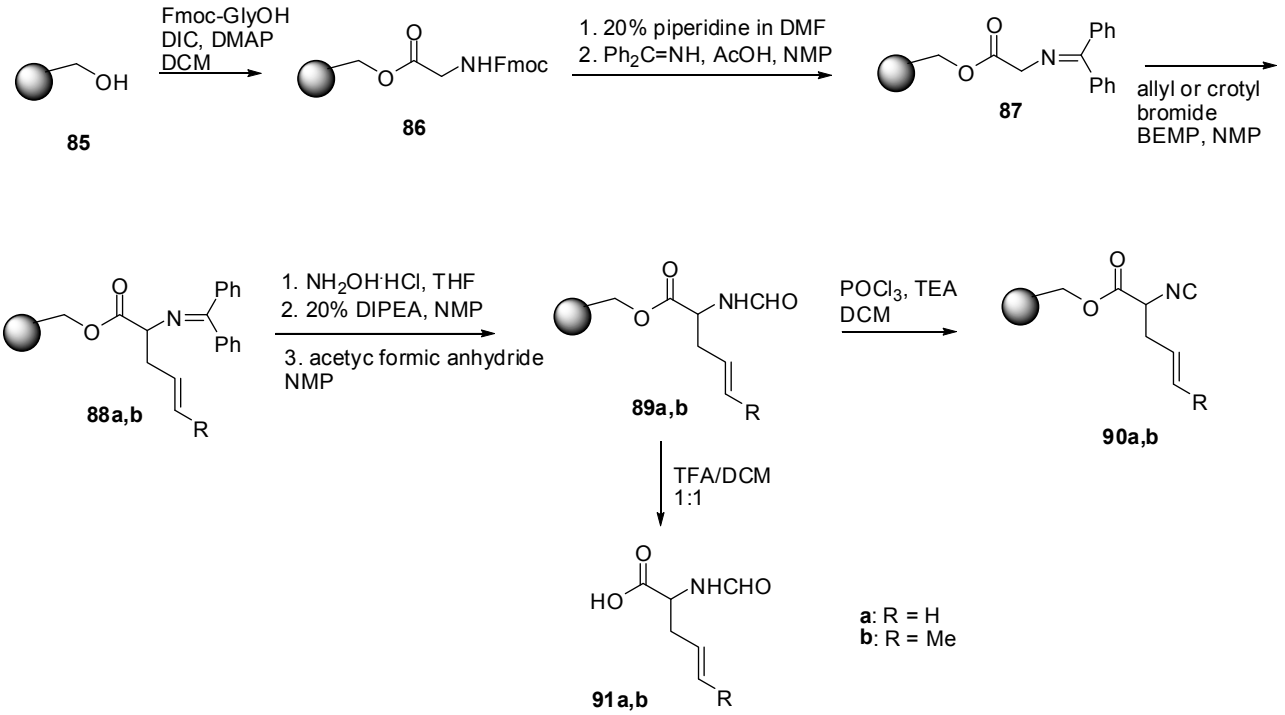
2.3.4 THIO-MEDIATED FREE RADICAL CYCLIZATION

Lamberto *et al.*[60] reported the synthesis of three novel alkenyl isocyanides in solid phase and their use in thiol-mediated radical cyclizations. To develop a general synthetic route to isocyanide resins with variation of the alkenyl side chain, the O'Donnell-Solid Phase methods was performed (Scheme 17).[61] Glycine coupled Wang resin **86** was reacted with benzophenone imine to activate the resin bound glycine for the next step.[62] Deprotonation and alkylation of the benzophenone imine resin **87** with allyl bromide or crotyl bromide was accomplished using iminophosphorane Schweisinger base, BEMP.[63]

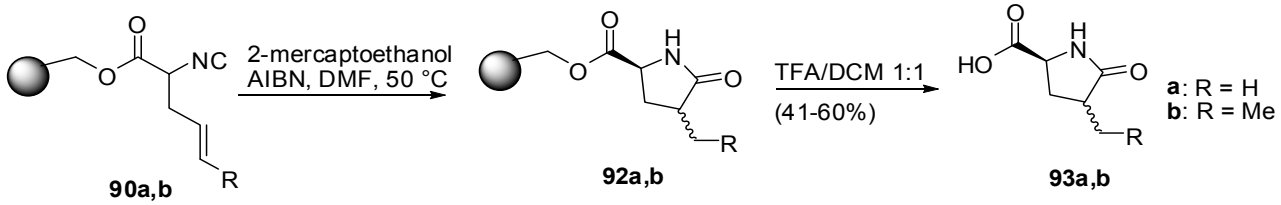


Scheme 16. Cyclization of protected glutamic acid derivatives in diverse acid conditions[58]

The residues on the resins **88a,b** were then hydrolyzed to afford the free amino group and formylated **89a,b**.[64] The desired alkenyl formyl-amino acids **91a,b** were obtained in excellent yields and purity (>90%). Dehydration of the resins **89a,b** afforded the desired isocyanides **90a,b**. The isocyanides **90a,b** were treated with mercaptoethanol and the cyclized products **92a,b** cleaved from the solid support to give **93a,b** (Scheme 18).



Scheme 17. Solid phase synthesis of polymer-supported isocyanides from glycine[60]

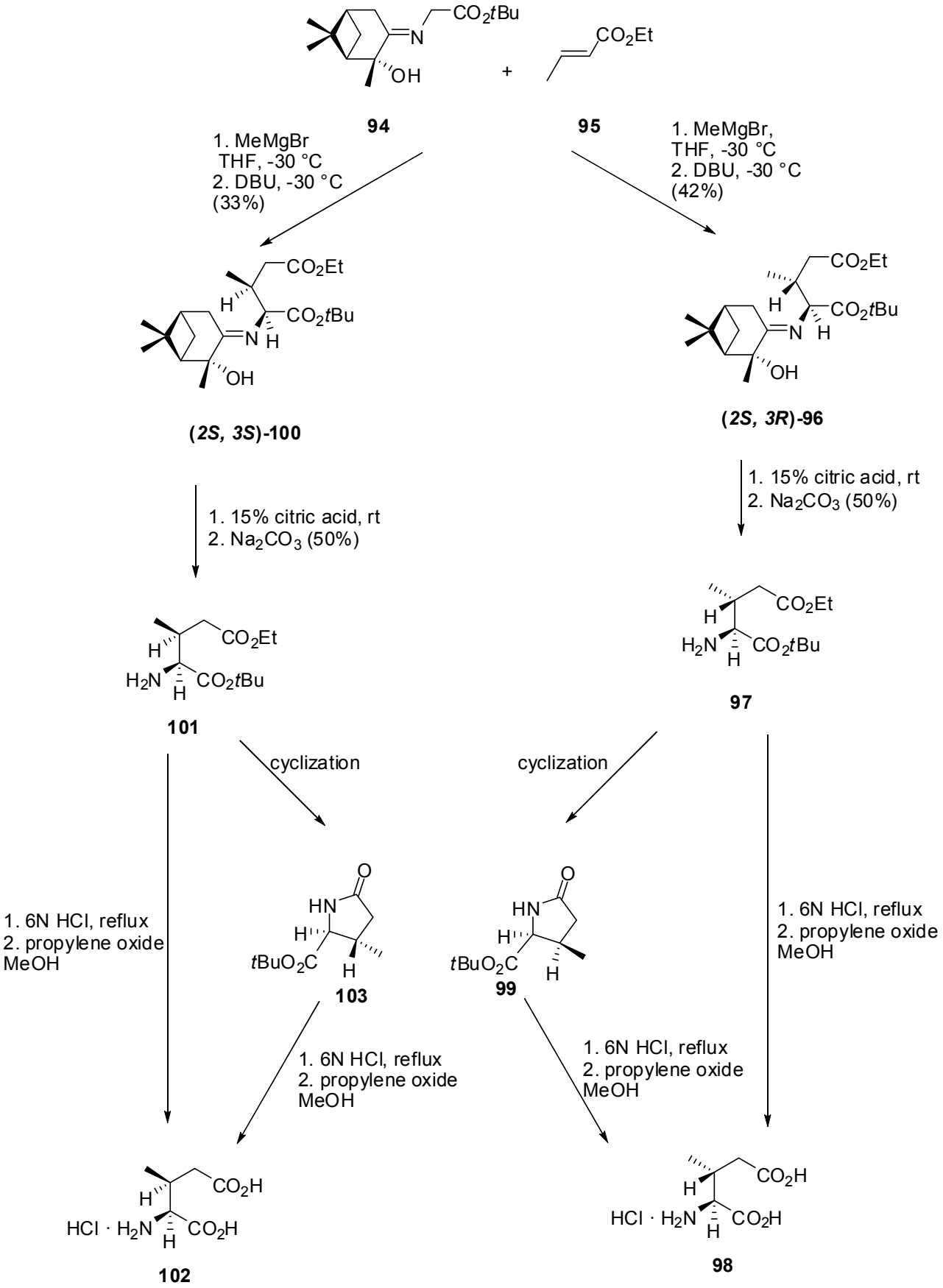


Scheme 18. Cleavage of the desired products from the resin[60]

2.3.5 1,4-ADDITION OF ENOLATE TO UNSATURATED ESTERS

Wehbe *et al.*[65] studied the preparation of several substituted glutamate analogues by Diels–Alder, 1,4-ionic and radical reactions starting from (2*S*)-4-methyleneglutamic acid. The synthesis of the analogues of (*S*)-Glu applying an intermolecular Heck reaction for soluble polymer (PEG) supported synthesis, was previously described. More recently, was demonstrated that (2*S*,3*S*)-3-methyl-glutamic acids act as blocker of the glutamate transport by EAAT2 (Scheme 19).[66]

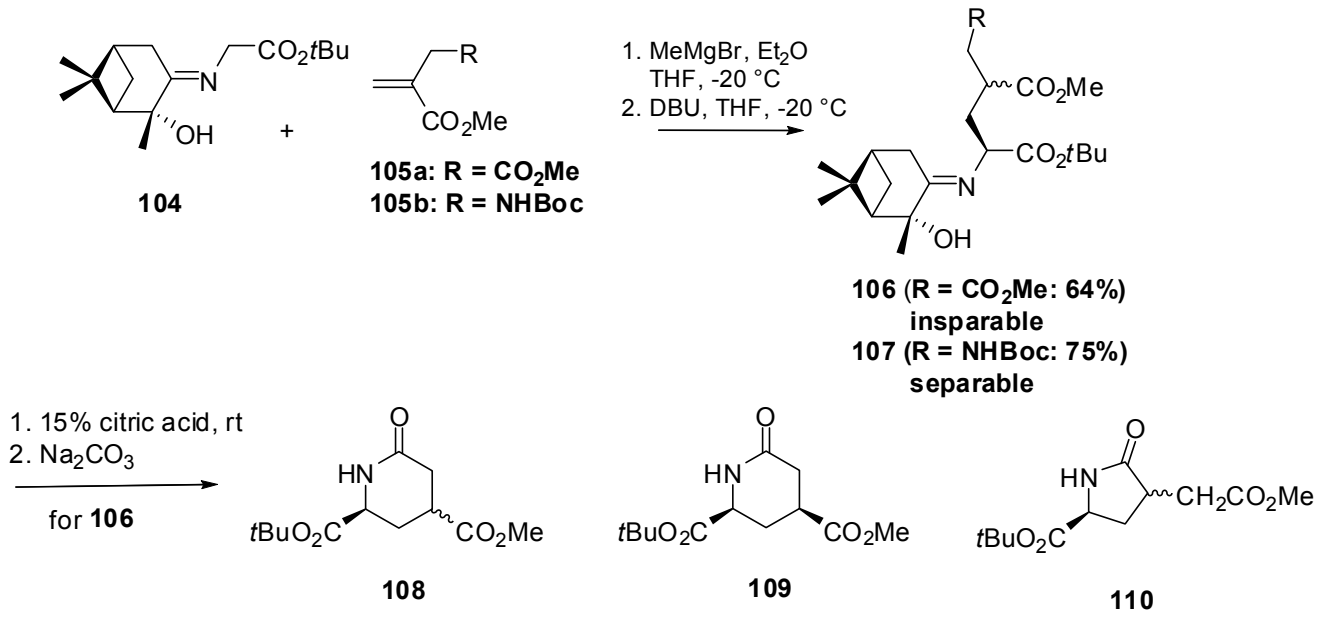
The Schiff base **94** was reacted with DBU to give two diastereomers **100** and **96** (75% yield, d.r. 56:44). After separation of the two diasteromers, the cleavage of the chiral auxiliary using citric acid at rt afforded the two amino esters **97** and **101** (82% and 73% yields respectively) which, after hydrolysis provided the corresponding hydrochloride salts (80% yield).



Scheme 19. Pyroglutamic acid derivatives via 1,4-addition[67]

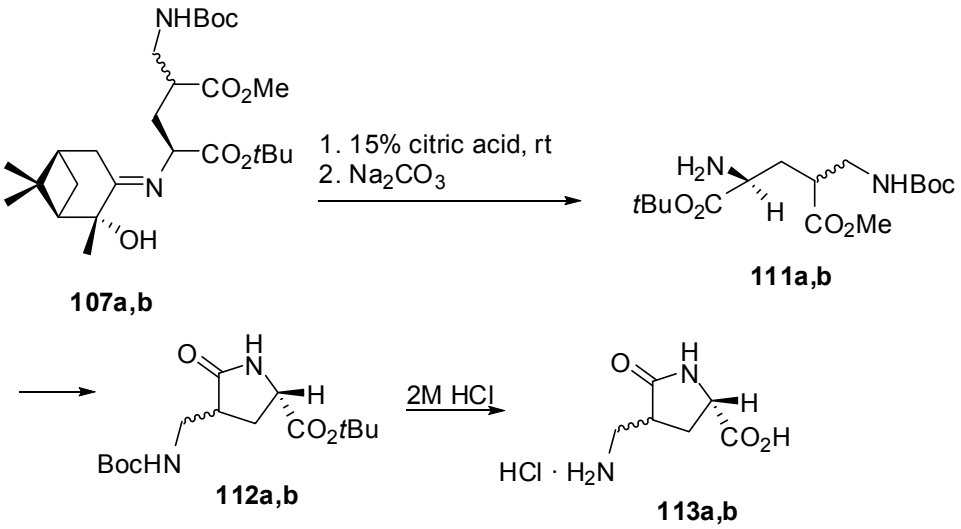
The hydrochloride salts were then treated with propylene oxide in MeOH to give the amino acids **98** and **101** in 90% yield. The cyclic *tert*-butyl-3-methylpyroglutamates **99** and **103** were obtained quantitatively at rt for 3 h. NMR analysis revealed that the stereochemistry of **99** was *cis* and for **103** was *trans*. These results permitted the assignment of (2*S*,3*R*) configuration to the major compound **98** and (2*S*,3*S*) configuration to the minor compound **102**.

Subsequently, Vandenberg *et al*.[67] studied the influence of the chemical group on the C4. The 4-substituted glutamic acid analogues with acid or basic function was chosen.[68] These were prepared using an asymmetric 1,4-addition of the enolate obtained from Schiff base, to unsaturated esters (Scheme 20).



Scheme 20. Synthesis of cyclic compounds reported by Wehbe *et al.*[66]

For the 4-carboxymethyl derivatives **106** and **107** *in situ* preparation of using DBU was chosen because it did not give polycondensation products as other chelating bases. At 20 °C in THF, **106** was obtained in 64% yield as a mixture of two inseparable diastereomers.[69,70] Acid treatment of **106** with citric acid followed by neutralization with carbonate yielded a diastereomeric mixture of the cyclic product **108** (74% yield) from which 1,3-*cis*-diastereomer **109** was obtained(35%) after recrystallization. The cyclization could produce a five membered ring **110**. The 1,4-addition reaction afforded **107** in 75% yield as a mixture of two diastereomers, which were separated by silica gel chromatography: **107a** (35%) and **107b** (30%). As shown in Scheme 19, cleavage of the chiral auxiliary from **107a** and **107b** by citric acid yielded the aminoesters **111a** and **111b** (85% yield), which were transformed slowly and quantitatively into cyclized products **112a** and **112b**; after removal of *t*Bu and Boc protecting groups by hydrolysis with 2M HCl, **113a** and **113b** were obtained (95% yield) (Scheme 21).



Scheme 21. Enantioselective synthesis of pyroglutamic acid derivatives[66]

3. CONCLUSIONS

In this work we have reviewed the most representative synthetic approaches to the development of pyroglutamate derivatives. Pyroglutamic acid and its derivatives are strictly related to prolines but conversely to the proline-chimeras that have been extensively studied and widely accepted as modification of peptide sequences, pyroglutamate derivatives have been only a marginal role in peptide drug discovery.[71] Their asymmetric syntheses have not been properly addressed yet, thus, there is a strong lack of pyroglutamic derivatives related to natural amino acids. A full pyroglutamic acid library derivatives, would expand the possibility to use these structures for drug discovery and certainly their insertion in place of native amino acids into bioactive peptides will achieve novel three dimensional arrangements and consequently peptides with different pharmacological and pharmacokinetic profiles.

REFERENCES

1. (a) D. K. Chalmers and G. R. Marshall, *J. Am. Chem. Soc.,* 1995, **117**, 5927; (b) A. Mollica, F. Pinnen, A. Stefanucci, L. Mannina, A. P. Sobolev, G. Lucente, P. Davis, J. Lai, S.-W. Ma, F. Porreca, and V. J. Hruby, *J. Med. Chem.,* 2012, **55**, 8477; (c) M. P. Paradisi, A. Mollica, I. Cacciatore, A. Di Stefano, F. Pinnen, A. M. Caccuri, G. Ricci, S. Duprè, A. Spirito, and G. Lucente, *Bioorg. Med. Chem.*, 2003, **11**, 1677; (d) A. Mollica, M. P. Paradisi, K. Varani, S. Spisani, and G. Lucente, *Bioorg. Med. Chem.,* 2006, **14**, 2253; (e) D. Torino, A. Mollica, F. Pinnen, F. Feliciani, S. Spisani, and G. Lucente, *Bioorg. Med. Chem.,* 2009, **17**, 251; (f) T.-Y. Tsai, T.-K. Yeh, X. Chen, T. Hsu, Y.-C. Jao, C.-H. Huang, J.-S. Song, Y.-C. Huang, C.-H. Chien, J.-H. Chiu, S.-C. Yen, H.-K. Tang, Y.-S. Chao, and W.-T. Jiaang, *J. Med. Chem.,* 2010, **53**, 6572; (g) P. Garin-Chesa, L. J. Old, and W. J. Rettig, *Proc. Natl. Acad. Sci. USA*,

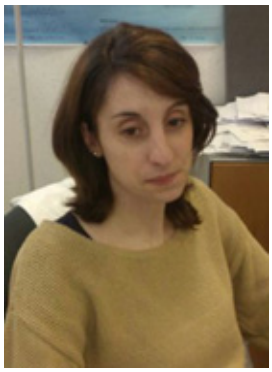
1990, **87**, 7235.

2. T. Itoh, M. Miyazaki, S. Ikeda, K. Nagata, M. Yokoya, Y. Matsuya, Y. Enomoto, and A. Ohsawa, *Tetrahedron,* 2003, **59**, 3527.
3. F. Effenberger, W. Muller, R. Keller, W. Wild, and T. Ziegler, *J. Org. Chem.,* 1990, **55**, 3064.
4. S. K. Panday, J. Prasad, and D. K. Dikshit, *Tetrahedron: Asymmetry,* 2009, **20**, 1581.
5. J. Dyer, S. Keeling, and M. G. Moloney, *Chem. Commun.,* 1998, 461.
6. L. Chen, J. W. Tilley, R. W. Guthrie, F. Mennona, T. Huang, G. Kaplan, R. Trilles, D. Miklowski, N. Huby, V. Schwinge, B. Wolitzky, and K. Rowan, *Bioorg. Med. Chem. Lett.,* 2000, **10**, 729.
7. D. W. Konas and J. K. Coward, *J. Org. Chem.,* 2001, **66**, 8831.
8. S. Conde, P. L. Serrano, and A. Martínez, *J. Mol. Cat. B,* 1999, **7**, 299.
9. D. A. Parrish and L. J. Mathias, *J. Org. Chem.,* 2002, **67**, 1820.
10. D. A. Parrish, L. J. Mathias, and K. M. Moore, *Macromolecules,* 2003, **36**, 4250.
11. S. El Ghammarti and B. Rigo, *J. Heterocycl. Chem.,* 1998, **35**, 555.
12. W. Huang and M. J. Miller, *Tetrahedron: Asymmetry,* 2008, **19**, 2835.
13. J. Ezquerra, C. Pedregal, A. Rubio, B. Yruretagoyena, A. Escribano, and S. S. Ferrando, *Tetrahedron,* 1993, **49**, 8665.
14. J. Ezquerra, C. Pedregal, A. Rubio, J. J. Vaquero, M. P. Matia, J. Martin, A. Diaz, J. L. G. Navio, and J. B. Deeter, *J. Org. Chem.,* 1994, **59**, 4327.
15. A. N. Bowler, P. M. Doyle, P. B. Hitchcock, and D. W. Young, *Tetrahedron Lett.,* 1991, **32**, 2679.
16. J. E. Baldwin, T. Miranda, M. Moloney, and T. Hokelek, *Tetrahedron,* 1989, **45**, 7459.
17. D. K. Dikshit and S. K. Panday *J. Org. Chem.,* 1992, **57**, 1920.
18. T. Ohta, A. Hosoi, and S. Nozoe, *Tetrahedron Lett.,* 1988, **29**, 329.
19. A. G. Avent, A. N. Bowler, P. M. Doyle, C. M. Marchand, and D. W. Young, *Tetrahedron Lett.,* 1992, **33**, 1509.
20. P. Merino, S. Anoro, S. Franco, F. L. Merchan, T. Tejero, and V. Tunon, *J. Org. Chem.,* 2000, **65**, 1590.
21. M. Scansetti, X. Hu, B. P. McDermott, and H. W. Lam, *Org. Lett.,* 2007, **9**, 2159.
22. T. K. Ellis, H. Ueki, T. Yamada, Y. Ohfune, and V. A. Soloshonok, *J. Org. Chem.,* 2006, **71**, 8572.
23. C. Nájera and M. Yus, *Tetrahedron: Asymmetry,* 1999, **10**, 2245.
24. J. C. Simpson, C. Ho, E. F. B. Shands, M. C. Gershengorn, G. R. Marshall, and K. D. Moeller, *Bioorg. Med. Chem.,* 2002, **10**, 291.
25. W. Li and K. D. Moeller, *J. Am. Chem. Soc.,* 1996, **118**, 10106.
26. T. Shono, Y. Matsumura, and K. Tsubata, In Organic Synthesis, ed. by G. Saucy, Organic Synthesis, 1984.

27. J. Isaacson, C. Gilley, and Y. Kobayashi, *J. Org. Chem.,* 2007, **72**, 3913.
28. (a) K. M. Short and A. M. M. Mjalli, *Tetrahedron Lett.,* 1997, **38**, 359; (b) G. C. B. Harriman, *Tetrahedron Lett.,* 1997, **38**, 5591; (c) C. Hanusch-Kompa and I. Ugi, *Tetrahedron Lett.,* 1998, **39**, 2725; (d) H. Tye and M. Whittaker, *Org. Biomol. Chem.,* 2004, **2**, 813.
29. C. B. Gilley, M. J. Buller, and Y. Kobayashi, *Org. Lett.,* 2007, **9**, 3631.
30. (a) T. Takenishi and O. Simamura, *Bull. Chem. Soc. Jpn.,* 1954, **27**, 207; (b) K. Pfister, W. J. Leanza, J. O. Conbere, H. J. Becker, A. R. Matzuk, and E. F. Rogers, *J. Am. Chem. Soc.,* 1955, **77**, 697.
31. (a) T. K. Ellis, V. M. Hochla, and V. A. Soloshonok, *J. Org. Chem.,* 2003, **68**, 4973; (b) V. A. Soloshonok, C. Cai, and V. J. Hruby, *Tetrahedron Lett.,* 2000, **41**, 135.
32. T. K. Ellis, C. H. Martin, H. Ueki, and V. A. Soloshonok, *Tetrahedron Lett.,* 2003, **44**, 1063.
33. Y. N. Belokon, N. B. Bespalova, T. D. Churkina, I. Cisarova, M. G. Ezernitskaya, S. R. Harutyunyan, R. Hrdina, H. B. Kagan, P. Kocovsky, K. A. Kochetkov, O. V. Larionov, K. A. Lyssenko, M. North, M. Polasek, A. S. Peregudov, V. V. Prisyazhnyuk, and S. Vyskocil, *J. Am. Chem. Soc.,* 2003, **125**, 12860.
34. T. K. Ellis, C. H. Martin, H. Ueki, and V. A. Soloshonok, *Eur. J. Org. Chem.,* 2003, **10**, 1954.
35. V. A. Soloshonok, H. Ueki, R. Tiwari, C. Cai, and V. J. Hruby, *J. Org. Chem.,* 2004, **69**, 4984.
36. M. Oba, T. Saegusa, N. Nishiyama, and K. Nishiyama, *Tetrahedron,* 2009, **65**, 128.
37. F. Felluga, C. Forzato, P. Nitti, G. Pitacco, F. Ghelfi, and E. Valentin, *Chirality,* 2012, **24**, 112.
38. S. Hanessian and R. Margarita, *Tetrahedron Lett.,* 1998, **39**, 5887.
39. (a) S. J. Wittenberger, S. A. Boyd, and W. R. Baker, *Synlett,* 1993, 795; (b) K. Narasaka and Y. Ukaji, *Bull. Chem. Soc. Jpn.,* 1988, **61**, 571; (c) K. Narasaka and Y. Ukaji, *Chem. Lett.,* 1986, **1**, 81.
40. (a) S. Hanessian and R. Schaum, *Tetrahedron Lett.,* 1997, **38**, 163; (b) A. Mollica, A. Stefanucci, F. Feliciani, D. Torino, I. Cacciatore, F. Pinnen, and G. Lucente *Tetrahedron Lett.,* 2010, **51**, 1333.
41. G. Cardillo and M. Orena, *Tetrahedron*,1990, **46**, 3321.
42. J. A. Marshall, A. W. Garofalo, and R. C. Sedreni, *Synlett,* 1992, 643.
43. (a) I. B. Parr, S. K. Boehlein, A. B. Dribben, S. M. Schuster, and N. G. J. Richards, *J. Med. Chem.,* 1996, **39**, 2367; (b) J. M. Humphrey, R. J. Bridges, J. A. Hart, and A. R. Chamberlin, *J. Org. Chem.,* 1994, **59**, 2467.
44. E. Artale, G. Banfi, L. Belvisi, L. Colombo, M. Colombo, L. Manzoni, and C. Scolastico, *Tetrahedron,* 2003, **59**, 6241.
45. (a) C. Alvarez-Ibarra, A. G. Csaky, M. Maroto, and M. L. Quiroga, *J. Org. Chem.,* 1995, **60**, 6700; (b) N. A. Sasaki, R. Pauly, C. Fontaine, A. Chiaroni, C. Riche, and P. Potier, *Tetrahedron Lett.,* 1994, **35**, 241.
46. (a) S. Hanessien, G. McNaughton-Smith, H. G. Lombart, and D. W. Lubell, *Tetrahedron,* 1997, **53**, 12789; (b) J. Gante, *Angew. Chem., Int. Ed. Engl.,* 1994, **33**, 1699.

47. X. Zhang, A. C. Schmitt, and W. Jiang, *Tetrahedron Lett.,* 2001, **42**, 5335.
48. M. Kaname and S. Yoshifuji, *Tetrahedron Lett.,* 1992, **33**, 8103.
49. K. Tanaka and H. Sawanishi, *Tetrahedron: Asymmetry,* 1998, **9**, 71.
50. S. Yoshifuji, K. Tanaka, T. Kawai, and Y. Nitta, *Chem. Pharm. Bull.,* 1985, **33**, 5515.
51. S. Yoshifuji and M. Kaname, *Chem. Pham. Bull.,* 1995, **43**, 1617.
52. T. Rosen, D. T. W. Chu, I. M. Lico, P. B. Fernades, K. Marsh, L. Shen, V. G. Cepa, and A. G. Pernet, *J. Med. Chem.,* 1988, **31**, 1598.
53. P. Merino, J. Revuelta, T. Tejero, U. Chiacchio, A. Rescifina A. Piperno, and G. Romeo, *Tetrahedron: Asymmetry,* 2002, **13**, 167.
54. (a) M. Shimazaki, F. Okazaki, F. Nakashima, T. Ishikawa, and A. Ohta, *Heterocycles,* 1993, **36**, 1823; (b) J. D. White, R. A. Badger, H. J. Kezar, A. J. Pallenberg, and G. A. Schieser, *Tetrahedron,* 1989, **45**, 6631; (c) M. Burdisso, A. Gamba, R. Gandolfi, and R. Oberti, *Tetrahedron,* 1988, **44**, 3735; (d) P. Herczegh, I. Kovacs, L. Szilagyi, T. Varga, Z. Dinya, and F. Sztaricskai, *Tetrahedron Lett.,* 1993, **34**, 1211; (e) E. Coutouli-Argyropoulou, E. Malamidou-Xenikaki, X. N. Stampelos, and I. N. Alexopoulou, *Tetrahedron,* 1997, **53**, 707.
55. P. Merino, J. Revuelta, T. Tejero, U. Chiacchio, A. Rescifina, A. Piperno, and G. Romeo, *Tetrahedron: Asymmetry,* 2002, **13**, 173.
56. P. Merino, E. Castillo, S. Franco, F. L. Merchan, and T. Tejero, *Tetrahedron,* 1998, **54**, 12301.
57. P. Merino, E. Castillo, S. Franco, F. L. Merchan, and T. Tejero, *J. Org. Chem.,* 1998, **63**, 2371.
58. V. P. Krasnov, I. A. Nizova, A. Y. Vigorov, T. V. Matveeva, G. L. Levit, P. A. Slepukhin, M. A. Ezhikova, and M. I. Kodess, *Eur. J. Org. Chem.,* 2008, 1802.
59. I. A. Nizova, V. P. Krasnov, O. V. Korotovskikh, and L. V. Alekseeva, *Izv. Akad. Nauk SSSR, Ser. Khim.,* 1989, 2781.
60. M. Lamberto, D. F. Corbett, and J. D. Kilburn, *Tetrahedron Lett.,* 2004, **45**, 8541.
61. M. J. O'Donnell, C. Zhou, and W. L. Scott, *J. Am. Chem. Soc.,* 1996, **118**, 6070.
62. M. J. O'Donnell and R. L. Polt, *J. Org. Chem.,* 1982, **47**, 2663.
63. (a) R. Schweisinger, J. Willaredt, H. Schlemper, M. Keller, D. L. Schmidt, and H. Fritz, *Chem. Ber.,* 1994, **127**, 2435; (b) D. L. Griffith, M. J. O'Donnell, R. S. Pottorf, W. L. Scott, and J. A. Porco, *Tetrahedron Lett.,* 1997, **38**, 8821.
64. (a) J. C. Sheehan and D.-D. H. Yang, *J. Am. Chem. Soc.*, 1958, **80**, 1154; (b) L. I. Krimen, *Org. Synth.,* 1988, **50**, 8.
65. J. Wehbe, V. Rolland, M.-L. Roumestant, and J. Martinez, *Tetrahedron: Asymmetry,* 2003, **14**, 1123.
66. J. Wehbe, V. Rolland, A. Fruchier, M.-L. Roumestant, and J. Martinez, *Tetrahedron: Asymmetry,* 2004, **15**, 851.

67. R. J. Vandenberg, A. D. Mitrovic, M. Chebib, V. J. Balcar, and G. A. R. Johnston, *Mol. Pharmacol.,* 1997, **51**, 809.
68. Z.-Q. Gu, D. P. Hesson, J. C. Pelletier, M.-L. Maccecchini, L.-M. Zhou, and P. Skolnick, *J. Med. Chem.,* 1995, **38**, 2158.
69. A. El Achqar, M. Boumzebra, M. L. Roumestant, and P. Viallefont, *Tetrahedron,* 1988, **44**, 5319.
70. T. Oguri, N. Kawai, T. Shioiri, and S. Yamada, *Chem. Pharm. Bull.,* 1978, **26**, 803.
71. (a) S. K. Panday, M. Dikshit, and D. K. Dikshit, *Med. Chem. Res.,* 2009, **18**, 566; (b) R. J. Nachman, K. Kaczmarek, H. J. Williams, G. M. Coast, and J. Zabrocki, *Biopol.*, 2004, **75**, 412; (c) A. Y. Vigorov, I. A. Nizova, K. E. Shalunova, A. N. Grishakov, L. S. Sadretdinova, I. N. Ganebnykh, M. A. Ezhikova, M. I. Kodess, and V. P. Krasnov, *Russ. Chem. Bull., Int. Ed*., 2011, **60**, 873.



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