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Cyclopropane Amino Acids

(2,3- and 3,4-Methanoamino Acids)

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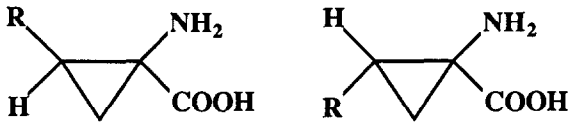
1. Introduction

For purposes of this review, the title compounds will be named and treated as modified amino acids. The carboxyl carbon atom is given the number one (1) rather than the quaternary ring carbon atom which is C_α in the common nomenclature system. The individual amino acids will be referred to as 2,3-methano- or 3,4-methanoamino acids, depending on the position of the cyclopropane ring on the amino acid carbon chain, instead of using the more colloquial (and ambiguous) "cyclopropyl"

prefix[1]. The only exception to the above will be the simplest cyclopropane amino acid, 1-aminocyclopropanecarboxylic acid (Acc), which might be called 2,3-methanoalanine, but is well established in the literature as Acc.

Except for the 2,3-methano analogs of proline, valine and pyroglutamic acid, each protein amino acid cyclopropane analog exists in diastereomeric E- and Z-forms in which the characteristic functionality at the β-carbon atom of the specific amino acid is *cis* to the carboxyl or to the amino function, respectively, (**Scheme 1**). Of course, each of these diastereomers consists of an enantiomeric pair as does 2,3-methanoproline, -valine and - pyroglutamic acid.

Scheme 1



Z-Isomer **E-Isomer**

The title amino acids are particularly interesting because they constitute a unique form of "conformationally constrained" amino acid which has been found in nature, generally in the unbound form or, in some cases, as simple dipeptides[2]. Several research groups, including that of the author, have been interested in the synthesis of these compounds for purposes of investigating their biological properties and those of peptides containing them. Since the cyclopropane ring introduces steric constraints into the amino acid residue, changes in the chemical reactivity of the pendant functional groups; e. g., reduced rates of hydrolysis of peptide or ester groups, result. Besides this effect, the "unsaturated" character of the cyclopropane ring favors certain conformations of the residue itself, i. e.; restriction of torsion angles about the C_α-C=0 bond (ψ) to small values, due to conjugation of the carbonyl group with the ring. When introduced into a peptide chain, the peculiar nature of this kind of amino acid residue is expected to cause profound changes in the proximal peptide conformation, which may affect the ability of the peptide to fit an enzyme active site and/or its intended bioreceptor. Also, as a consequence of the severe carbon-carbon angle deformation demanded by the cyclopropane ring, a latent instability is incorporated into the peptide which, if unmasked *in vivo*, will form reactive entities capable of capturing nucleophiles or electrophiles present in a receptor or enzyme active site.

2. Naturally Occurring Cyclopropane Amino Acids

A. 2,3-Methanoamino Acids

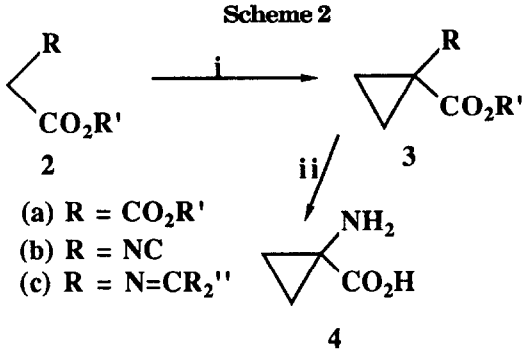
i. 1-Aminocyclopropanecarboxylic Acid (Acc).

Since the first report of its isolation from cider apples and perry pears by Burroughs[3] and its identification as an intermediate in the biosynthesis of ethylene,[4] 1-aminocyclopropane carboxylic

acid, Acc, has been of considerable interest to plant biologists and chemists alike. Previous reviews[5,6] have described some of its chemistry and biochemistry and summarized the investigations[7,8] that led to the discovery of its role as a biosynthetic intermediate in the conversion of methionine to ethylene in plants. This review will endeavor to cover recent synthetic chemical and biologically important research since 1980.

Synthesis. Many different approaches to the chemical synthesis of Acc and some of its simple ring-alkylated analogs are worth reviewing for their chemical content and bearing on important biological investigations. One of the earliest and most straightforward synthetic methods entails the alkylation of a glycine derivative or congener with ethylene dibromide or its equivalent (**Scheme 2**). The first synthesis[9] of Acc started with the diester **2a, R'=C_2H_5**, followed by conversion of one ester

Scheme 2

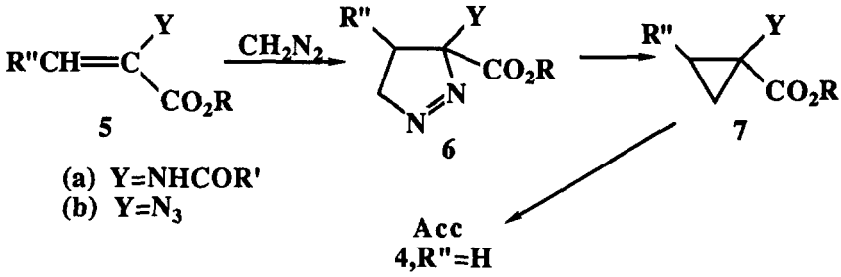


Reagents: i, $BrCH_2CH_2Br$/Base; ii, HCl/H_2O

function into an amino group by alkaline rearrangement of the *bis*-N-bromoamide into a spirohydantoin, which was hydrolyzed to give the free amino acid **4**. This method has been repeated successfully by others.[10] A recent report of the use of **2a** describes the synthesis of the N-Boc derivative of **4** and its deblocking in boiling water.[11] The more elegant use of the isonitrile **2b**, prepared from N-formylglycine ester, obviates the need for the amide rearrangement since simple hydrolysis of the isonitrile affords the amino group directly.[12] This approach was also used to prepare specific isomers of ring-deuterated Acc for use in biological studies.[13] Use of the benzophenone Schiff base[14] (**2c, R"= Ph**) simplifies the process even further and its conversion into **4** is accomplished by phase-transfer catalyzed cycloalkylation to give **3c** followed by hydrolysis.[15] Using **2a** (**R'= t-butyl**), this method was used successfully to synthesize 2-methyl-Acc and various deuterated stereoisomers for enzyme studies.[16]

A second synthetic approach to **4** is the "diazo addition" method in which diazomethane is added to an α-substituted acrylic acid derivative (**5a, R"=H**) forming a pyrazoline (**6a, R"=H**) convertible into the desired cyclopropane (**Scheme 3**). This method[17] was first carried out with **R=R'**=Me followed by thermolysis to convert the pyrazoline (**6a, R"=H**) into cyclopropane (**7a, R"=H**). Alkyl and aryl substituted diazomethanes give 5-substituted pyrazolines and, subsequently, 2-substituted-Acc derivatives.[18] In the author's laboratories, the final removal of the N-acyl group by vigorous acidic hydrolysis, or hydrogenolysis in the case of the benzyloxycarbonyl blocking group,

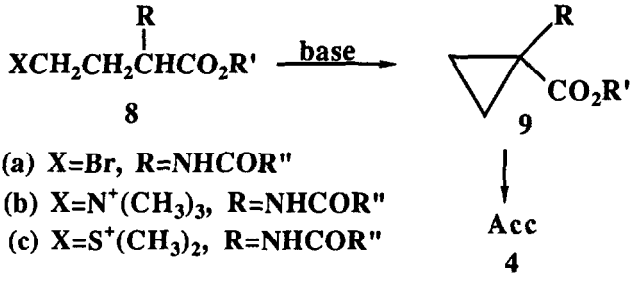
Scheme 3



has been successful only when the 2-substituent is hydrogen or alkyl.[19] However, when **Y** is an azido group (**5b**), the final step requires reduction of the azido function[20] and is reported to be successful when **R"= H, Me, Et** and **Ph.**

A third type of Acc synthesis proceeds through the intramolecular cyclization of a γ-substituted α-aminobutyric acid derivative or precursor, **8** (**Scheme 4**). Use of strong base on **8a, R"= OBzl**, gave the cyclopropane **9a, R"=OBzl**, which was converted to **4** by hydrolysis.[21] Since the γ-carboxyl group of glutamic acid can be converted into a leaving group, glutamic acid can serve as the ultimate starting material for Acc.[22] The quaternary ammonium function[23] of **8b** and the dimethylsulfonium group[24] of **8c** can also replace bromide as leaving group in this cyclization giving good yields of various cyclopropane intermediates (**9**). Acc can also be prepared by the nitration of the α-carbanion formed from cyclopropane carboxylic acid esters with t-butyllithium

Scheme 4



followed by reduction of the intermediate nitro compound.[25] Interestingly, the intermediate nitro ester, having geminal electron accepting groups on the ring, is quite susceptible to ring opening by various nucleophiles[26] with the formation of γ-substituted α-nitro acids convertible by reduction into special amino acids.

Alkylation of the chiral intermediate, (R)-(-)-2,5-dimethoxy-3-benzyl-3-methyl-3,6-dihydropyrazine, used previously in an asymmetric synthesis of amino acids[27], allowed the preparation of optically active 2,2-dideutero-Acc and the determination of the absolute configurations of its enantiomers by NMR.[28]

Crystallographic determination of the conformation of the Acc residue in simple derivatives and peptides has been carried out and has just appeared.[29]

Biology. The stereochemistry of the enzymatic decomposition of Acc to ethylene and its synthesis from S-adenosylmethionine (SAM) in plants has been studied in considerable detail. An early paper[30] showed that postclimacteric apple slices and mungbean hypocotyls converted the (1R,2S)-2-ethyl-Acc into 1-butene 50-200 times faster than they decomposed the other three stereoisomers of 2-ethyl-Acc. Further work with [1-^{14}C]Acc led to the conclusion that, in mung bean,[31] the carboxyl group yielded carbon dioxide and the quaternary carbon (C_α) was released as hydrogen cyanide. Work with 1-azidocyclopropanecarboxylic acid and ^{13}C and ^{2}H labelled Acc led to the same conclusion.[32] A detailed mechanism[33] has been proposed and the reaction pathway has been subjected to semiempirical molecular orbital calculations,[34] which support the formation of radical intermediates. Surprisingly, it was found that *enzymatic* decomposition of stereospecifically deuterated Acc derivatives occurred *non*-stereospecifically while *chemical* decomposition, using sodium hypochlorite, took place with reproducible retention of configuration[16a,35]. These stereochemical results can be explained by invoking cation radical and nitrene intermediates in the first and second reactions, respectively.[36] Further studies of the chemical decomposition of Acc using various reagents has recently been reported.[37] The required deutero compounds were prepared by alkylation of a glycine Schiff base with *meso*- and *racemic* dideuterodibromoethylenes. For these studies, 2-methyl- and 2-ethyl-Acc were also prepared[16b] by condensing *meso*- and *racemic*-1,2-dibromopropanes with di-t-butyl malonate, as described in **Scheme 2**, and the racemic amino acid esters were resolved and the absolute configurations determined.[38] It is noteworthy that among the four racemic diastereomers prepared, only the (Z)-2-ethyl-Acc ester was resolvable enzymatically (Acylase I hydrolysis of the N-chloroacetyl derivative), while the others required chemical resolution. Similarly, in the author's laboratory, attempts at the enzymatic resolution of several different derivatives of 2,3-methanophenylalanine also failed.

Shortly after it was reported[39] that microorganisms of *Pseudomonas sp.* deaminated Acc to form α-ketobutyrate and ammonia, mechanistic studies of this conversion appeared.[40] Separately, the optically active 2,2-dideutero-Acc used in these studies[28] was prepared by two unique syntheses, one using a benzene ring as precursor of the required carboxyl function and the other proceeding through a chiral epoxide.[41]

Studies on the stereochemistry of the enzymatic formation of Acc from S-adenosylmethionine in plants have also appeared[42]. The results indicate that inversion occurs at the γ-carbon atom of the methionine residue with direct intramolecular nucleophilic displacement of adenosyl methyl sulfide by an α-carbanion to form the cyclopropane ring. Attempts to find alternate substrates and inhibitors of Acc synthetase from tomato plants showed that the enzyme accepts only very specific substrates.[43]

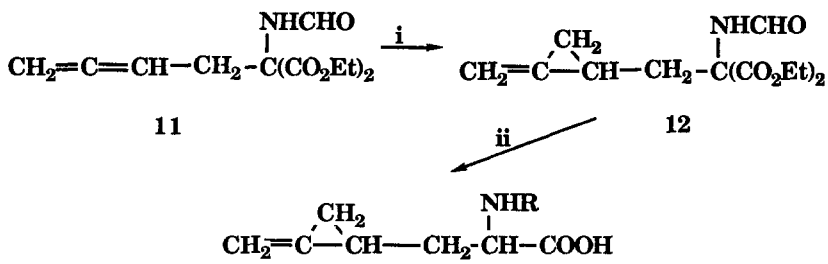
Recently, new *in vivo* inhibitors of ethylene biosynthesis have been reported.[44] 1-Aminocyclopropene and 2-cyclopropyl-1-aminocyclopropanecarboxylic acid are both k_{cat} inhibitors of ethylene forming enzyme and inhibit senescence in carnations. It appears that during processing, the unsaturated analog of Acc forms an especially stable cyclopropenone imine intermediate which can react with nucleophiles at the enzyme active site. The phosphorus analog of Acc, 1-aminocyclopropanephosphonic acid, is reported to be a time-dependent inactivator of Acc

deaminase from a *Pseudomonas* species and an alanine racemase from *B. stearothermophilus.*[45] In the rat brain, Acc has been found to mimic the effects of glycine on the NMDA receptor.[46]

ii. Hypoglycine (10).

Hypoglycine A (**10a**), a substance having marked hypoglycemic effects in animals, and hypoglycine B (**10b**), its N-γ-glutamyl derivative, were first isolated from the fruit of *Blighia sapida* ("Ackee") in 1954[47] and later by others[48] from other sources. Later, a homolog of hypoglycine A, α-(methylenecyclopropyl)-glycine, was isolated from *Lichti chinensis*[49] and also caused hypoglycemia in animals. Early synthetic work afforded **10a** in poor yields[50], but a later synthesis (**Scheme 5**) in which the allene, 1-bromo-2,3-butadiene, is alkylated with diethyl formylamino-malonate to form **11** followed by cyclopropanation (Simmons-Smith procedure) and decarboxylation gave **10a** in somewhat higher yield[51]. These workers also showed that the absolute configuration of the natural (+)-isomer was (2S,3S).

Scheme 5



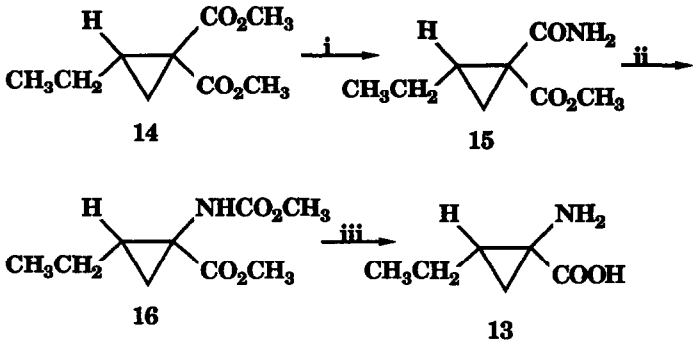
10 a, R = H
b, R = γ-glutaminyl

Reagents: i, CH_2I_2, Zn-Cu; ii, NaOH then HCl

iii. Coronamic Acid (13).

Coronatine, a toxin produced by *Pseudomonas corona-facience* which induces chlorosis in Italian ryegrass, consists of coronamic acid, (+)-(E)-2-ethyl-1-aminocyclopropane-carboxylic acid, which is N-acylated by coronafacic acid[52], a hydrindanonecarboxylic acid. The first synthesis[53] of **13** (**Scheme 6**) began with the diester, **14**, prepared by dialkylation of dimethyl malonate with 1,4-dibromo-2-butene followed by hydrogenation of the remaining vinyl group. Aminolysis of the less hindered ester function gave the amide, **15**, which was converted into the urethane **16** by Hofmann degradation. Hydrolysis of **16** gave racemic **13** which was resolved as the brucine salt of the N-formyl derivative and by L-acylase hydrolysis of the N-acetyl derivative. Surprisingly, application of the sector rule[54] to this cyclopropane amino acid led to the conclusion that the (+)-form obtained directly from the enzymatic hydrolysis had the unlikely[55] 1R,2R-configuration. This assignment was later revised[56] to the 1S,2S-configuration based on enzymatic oxidation experiments and x-ray crystallography, indicating that the sector rule may not

Scheme 6



Reagents: i, NH_3/MeOH; ii, Br_2/NaOH; iii, H_2O

be applicable in the case of cyclopropane amino acids. A synthesis of the optically active forms of *allo*coronamic acid was also reported by the same research group[57] in which dimethyloxosulfoxonium methylide was used to cyclopropanate propylidenecyanoacetic ester followed by resolution of the free amino acid *via* the quinine salt. A chiral synthesis of *allo*coronamic acid in poor yield has also been reported.[58] A later synthesis of racemic **13** in the author's laboratory[17] by the addition of diazoethane to a dehydroalanine derivative (see **Scheme 3**) followed by pyrolysis and deblocking was quite efficient. Coronafacic acid has also been synthesized.[59]

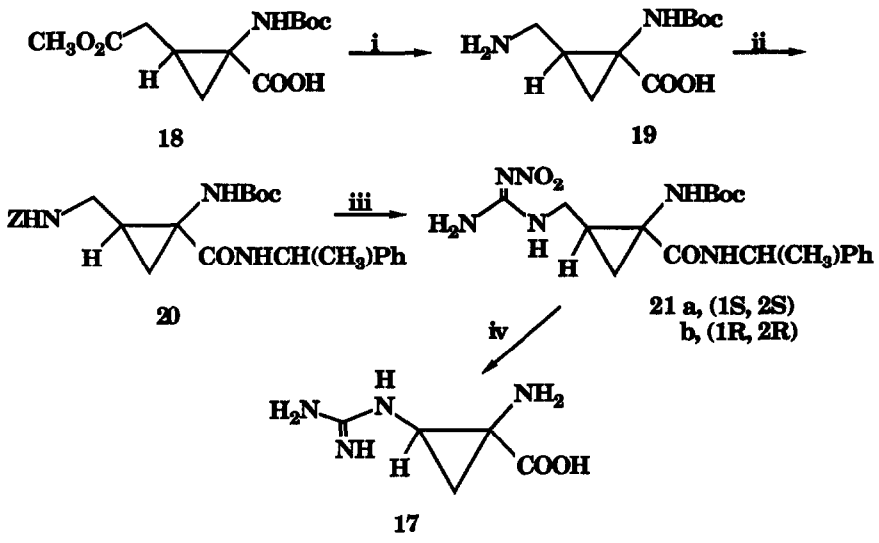
Norcoronatine, a minor component of the phytotoxic fraction of *Pseudomonas syringae* pv. *glycinea* has been shown to contain norcoronamic acid, E-(1S,2S)-2-methyl-1-aminocyclopropanecarboxylic acid.[60]

Studies of the biosynthesis[61], enzymatic deamination[62] and the bioactivity of a series of coronatine analogs[63] have also been published.

iv. Carnosadine (17).

In 1984, the isolation of a new 2,3-methanoamino acid, carnosadine, from a red alga, *Grateloupia carmosa*, was reported.[64] This was followed by a report of the synthesis[65] of **17** (**Scheme 7**) which proceeded by way of the 2,3-methanoglutamic acid derivative, **18**, also synthesized for the first time. Conversion of the γ-ester function of **18** into an amino group by Hofmann degradation of the corresponding amide gave **19** which was coupled with (R)-(+)-α-methyl-benzylamine to give the amides (**20**) separable into the expected diastereomers. The γ-amino group was then deprotected, guanidinated (3,5-dimethyl-1-nitroguanylpyrazole) and deblocked by hydrogenolysis followed by hydrolysis to give **17**. The natural compound was shown to have the (1S,2S) configuration.

Scheme 7



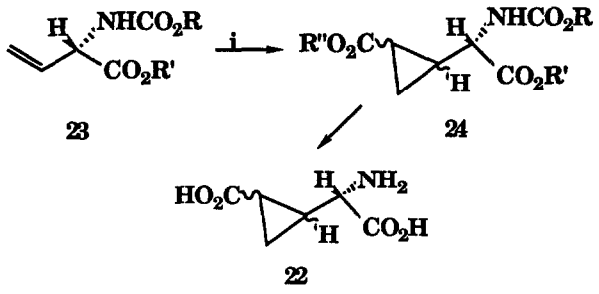
Reagents: i, (a) NH_3/MeOH, (b) Br_2/NaOH; ii, (a) ZCl/NaOH, (b) (+)-$PhCH(CH_3)NH_2$/DCC/HOBt; iii, (a) H_2/Pd, (b) DNG; iv, (a) H_2/Pd, (b) 6M HCl.

B. 3,4-Methanoamino Acids

i. 3,4-Methanoglutamic Acid (22).

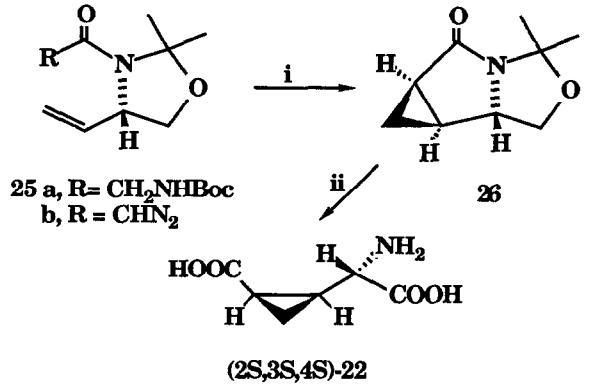
The isolation and structural assignment of both cis-(2S,3S,4R)- and trans-(2S,3S,4S)-3,4-methanoglutamic acid (**22**) (along with 3,4-methanoproline, *vide infra*), the former from *Aesculus parviflora* and the latter from *Blighia sapida*, were reported[66] in 1969. An isomer, trans-α-(2-carboxymethylcyclopropyl)-glycine has been isolated from *Blighia unijugata*.[67] The first reported synthesis[68] of **22** was not enantioselective and used cis- and trans-cyclopropane-1,2-dicarboxylic acids as intermediates. Three partially enantioselective syntheses using the diazo addition method have also been reported. The first[69] proceeded by rhodium acetate catalyzed addition of diazoacetic ester to (2S)-vinylglycine (**23**) to form all four of the chromatographically separable diastereomers of **24** (**Scheme 8**).[70] A second diastereoselective synthesis[71] of **22** used a stereoselective oxidation of allylglycine, giving *threo*-3-hydroxyallylglycine, which was then converted through several steps into the natural product. The third synthesis[72], being enantioselective, gave the natural (2S,3S,4S) isomer of **22** (**Scheme 9**). The starting alkene, **25a**, prepared from (2S)-2-amino-3-butenol, was deblocked, diazotized and ring-closed to give **26**, the desired isomer, in 6:1 ratio. Removal of the ketal grouping and hydrolysis of the amide followed by the necessary N-blocking, oxidation and N-deblocking gave enantioselectively the natural isomer of **22**. Direct addition of diazoacetic ester to **25a** gave all four cyclopropanes, with the natural isomer being formed in only 15% yield.

Scheme 8



Reagents: i, $N_2CHCO_2R'/Rh(OAc)_4$.

Scheme 9



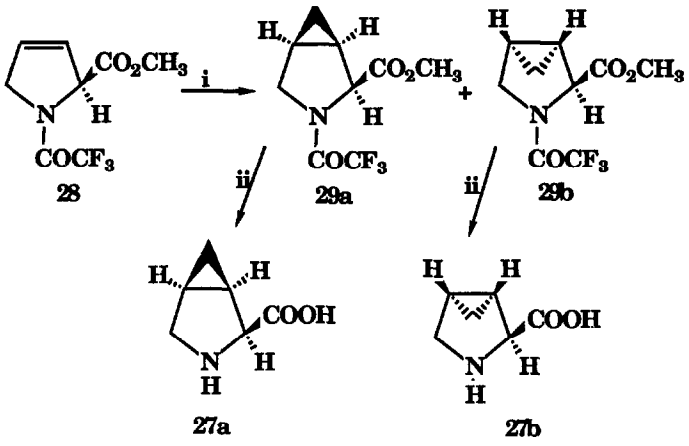
Reagents: i, (a) TMSOTf, (b) $NaNO_2$/pH 3, (c) $Pd(Ac)_2$; ii, (a) $AcOH/H_2O$, (b) NaOH, (c) $(Boc)_2O$/pH 9, (d) Jones oxid., (e) TFA.

ii. 3,4-Methanoproline (27).

The isolation of this cyclopropane amino acid was reported[66] in 1969 from seeds of *Aesculus parviflora* along with its probable biogenetic percursor, 3,4-methanoglutamic acid **22** (*vide supra*). It was shown by hydrogenation studies and NMR spectroscopy to have the cis configuration. In 1971, the synthesis (**Scheme 10**) of both cis- and trans-3,4-methanoproline (**27a** and **27b**) from (2S)-3,4-dehydroproline (**28**) was published[73] confirming the cis-(2S,3R,4S)-configuration (**27a**). A crystallographic study of **27a** showed that the boat conformation was preferred.

iii. Cleonine. Cleonine replaces the threonine residue of bleomycin to form cleomycin.[74] This novel amino acid, 3,4-methanothreonine, is the only natural 3,4-methanoamino acid having an hydroxyl group directly on the cyclopropane ring. Racemic cleonine has been synthesized.[75]

Scheme 10



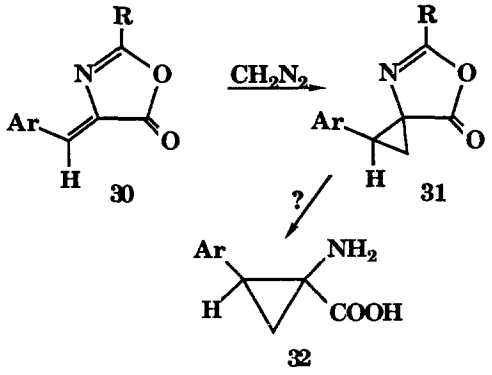
Reagents: i, CH_2N_2/CuCl; ii, 2N NaOH/r.t.

3. Synthetic Cyclopropane Amino Acids

A. Aromatic 2,3-Methano Amino Acids.

Several methods have been used to prepare cyclopropane-containing aromatic amino acids. The addition of diazomethane to 4-arylidene-5(4H)-oxazolones was one of the earliest methods reported[76] for preparation of cyclopropanes that might be converted into aryl-2,3-methanoamino acids (**Scheme 11**). The configurations of the arylideneoxazolones **30**, generally prepared by the

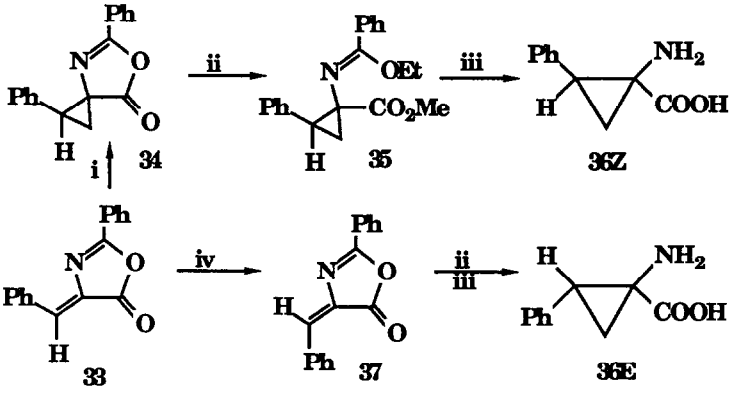
Scheme 11



Erlenmeyer-Plöchl reaction[77], were studied chemically[78] and finally settled by NMR studies of the oxazolones[79] and the derived N-benzoyl dehydroamino acid esters[80] as well as by an x-ray structure of a hydrolysis product, 2-benzamidocinnamic acid.[81] The stable isomer in all cases had the Z-configuration in which the aryl group and the carboxyl function have the trans orientation. In the

author's laboratories, attempts to hydrolyze a spirooxazolone (**31, R=Ph, Ar=Ph**) to the free 2,3-methanoamino acid confirmed that these compounds might not be prepared this way.[82] The formation of styrylglycine from attempted hydrolysis of **31** (**R=Ph, Ar=Ph**) was explained mechanistically by assuming protonation of the carbonyl group leading to formation of a benzyl carbocation with release of strain in the cyclopropane ring. It has been shown in other work that aliphatic groups at the 3-position do not destabilize the ring in this way, thus allowing acid catalyzed hydrolysis to be performed on derivatives of 2,3-methano aliphatic amino acids. Other workers[83] had isolated the corresponding lactone, 3-amino-5-phenylbutyrolactone, from attempted hydrolysis of **31**. The first synthesis[84] of the (E)- and (Z)-isomers of racemic **2,3-methanophenylalanine** from **33** (**Scheme 12**) was carried out in the author's laboratory using mild hydrolysis of an imino ester

Scheme 12



Reagents: i, CH_2N_2; ii, (a) MeOH/DMAP, (b) $Et_3O^+BF^-$; iii, (a) 1N HCl/r.t., (b) NaOH, (c) HOAc; iv, HBr.

intermediate (**35**) formed from the N-benzoyl group by treatment with Meerwein's reagent. Hydrolysis of a remaining methyl ester or hydrogenolysis of a benzyl ester afforded the free amino acid **36Z** in good yield. Isomerization of **33** with hydrogen bromide[85] gave the E-oxazolone **37** convertible into the E-amino acid by the same procedure used for the Z-isomer. An x-ray crystallographic, ^1H and ^{13}C NMR studies of these compounds have been reported,[82] confirming the structure. A recent chiral synthesis[86] of (Z)-2,3-methanophenylalanine was reported in which **30** (**R=Me, Ar=Ph**) was converted into an optically active 2,5-piperazinedione with L-proline, or to a chiral ester with (-)-N-methylephedrine, *before* diazomethane cyclopropanation. Both the (+)- and (-)-enantiomers of the amino acid were reportedly obtained in good yields by acid-catalyzed hydrolysis of their N-benzoyl derivatives.

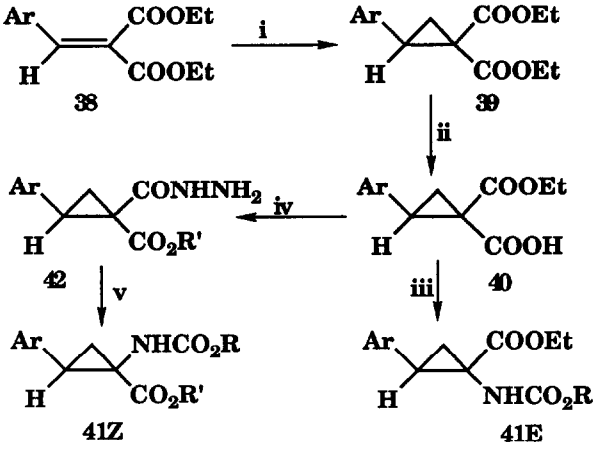
The instability of the cyclopropane ring to strong acid hydrolysis[82] and to hydrogenolysis[82], shown in earlier work, was circumvented during the synthesis of some twenty-four aromatic ring-substituted 3-aryl-2,3-methanoamino acids by the use of the sulfur-containing heterocycles, 4-arylidene-2-benzylthio-5(4H)-thiazolones,[83] which are cleavable to the amino acids by strong aqueous

base. A number of the 2,3-methanophenylalanines prepared by this method were shown to be reversible time-dependent inhibitors of both 3',4'-dihydroxyphenylalanine (DOPA) decarboxylase[87] and tyrosine aminotransferase.[88] The kinetic results of these studies suggested that very complex mechanisms were operating. Very recent results from the author's laboratory confirm the inhibitory action of 3'-hydroxy-2,3-methanophenylalanine on DOPA decarboxylase. The reactions of diazomethane with oxazolones[89], thiazolones[90] and (E)- and (Z)-2-acylaminocinnamates[91] have also been studied. In the author's laboratories, **(Z)-2,3-methanotyrosine** was prepared by diazomethane cyclopropanation of a thiazolone intermediate which allowed removal of the resulting N-thiobenzoyl group by alkaline cleavage.[92] Surprisingly, 2,3-methano-DOPA has not been reported and published attempts[93] using the oxazolone method failed.

Cyclopropanation of **33** with dimethyloxosulfonium methylide failed due to reaction at the electrophilic carbonyl group of the oxazolone[94], but 5-arylidene Meldrum's acid derivatives gave superior yields of cyclopropanes using this reagent. The derived cyclopropane diesters were then converted into 2,3-methanoamino acids by conversion of one ester function into an amino group.[95]

A cyclopropane analog of **(Z)-2,3-methanothyronine** and its 3',5'-bromine derivative, having little thyroxine-like activity, have also been synthesized by the oxazolone method.[76c] **(Z)-2,3-Methanohistidine** has also been synthesized by diazomethane cyclopropanation of the appropriate oxazolone[76b], albeit in poor yield. Both of the spirooxazolones encountered in these syntheses were reportly converted into the desired 2,3-methanoamino acids by acid catalyzed hydrolysis without apparent decomposition. The histidine analog had a weak inhibitory effect on histidine decarboxylase.

Scheme 13

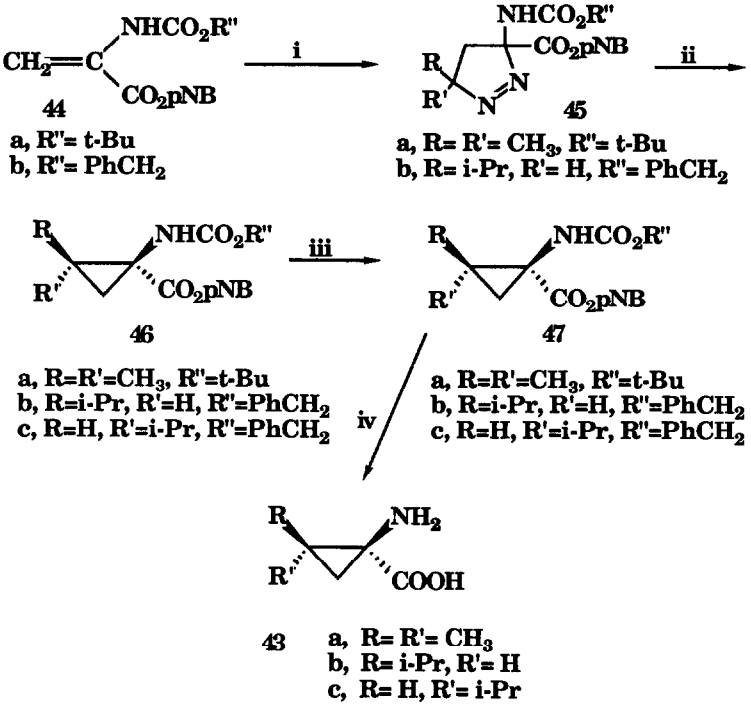


Reagents: i, $(CH_3)_2S^+(O)CH_2^-$; ii, $NaOH/H_2O$; iii, $(PhO)_2PON_3$, ROH; iv, NH_2NH_2/EtOH; v, HONO, ROH.

The malonate method, first reported in a patent[96], has recently been used for the synthesis of aromatic 2,3-methanoamino acids; i. e., the cyclopropanation of an arylidene malonic acid diester

with dimethyloxosulfonium methylide[97] followed by introduction of the amino group using an azide rearrangement. This method (**Scheme 13**) has two advantages: (1) either diastereomer of the target amino acid can be synthesized at will, (2) diazocompounds are not used. It is, however, longer than the oxazolone process since one of the ester functions must be converted into an amino group. Both diastereomers of **2,3-methanotyrosine** have recently been synthesized by this method.[98] The cyclopropane **39** is easily saponified to give the less sterically hindered acid (**40**) which is rearranged to the isocyanate with diphenylphosphoryl azide. Conversion to the urethane (**41E**) is done *in situ* by isocyanate alcoholysis and the final deblocking steps depend on the alcohol used. The (Z)-isomer (**41Z**) is obtained from **40** by hydrazinolysis of the ester function followed by nitrosation, rearrangement and alcoholysis as before. Resolution of the racemic amino acids obtained by this method is necessary if the optical isomers are desired.

Scheme 14



Reagents: i, $RR'CN_2$, ii, Δ or hν, iii, $NaOH/H_2O$; iv, HCl or H_2/5% Pd/C.

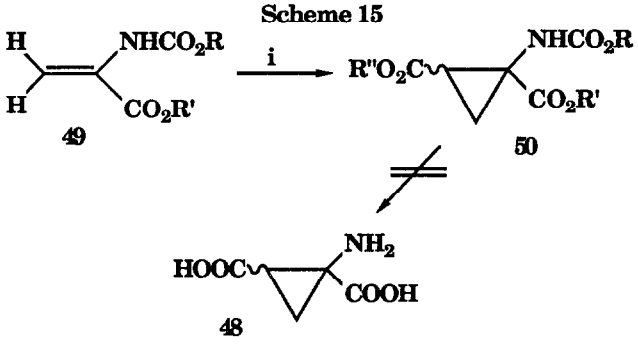
B. Aliphatic Amino Acids.

Several different methods have been used to prepare cyclopropane analogs of aliphatic amino acids. Racemic **2,3-methanovaline** (**43a**) and both (**Z**)- and (**E**)-**2,3-methanoleucines** (**43b** and **43c**) were prepared by the addition of a substituted diazomethane to a dehydroalanine derivative[99] (**44**) (**Scheme 14**). Photolytic conversion, which is superior to pyrolysis, of the pyrazolines (**45**) to the cyclopropanes

(**46**) followed by standard deblocking procedures allows final isolation of the amino acids. The aliphatic 2,3-methanoamino acids are stable to both hot aqueous acid and hydrogenolysis (5%Pd/C) at atmospheric pressure. Compound **43a** is formed in the hydrolysis mixture when an N-t-butyl group is removed by strong hydrolysis of 1-N-t-butyl-1-cyano-2,2-dimethylcyclopropane.[100]

The synthesis of **2,3-methanoaspartic acid** (**48**) has been attempted by the addition of various diazoacetic esters to a number of dehydroalanine derivatives[101] (**Scheme 15**). Both stereoisomers of **50**, the E-isomer predominating, were prepared, and, even though both carboxyl groups could be deblocked to form the diacid uneventfully, no cyclopropane was isolated when the amino group was liberated.[102] This confirms earlier reports[103] that 2-aminocyclopropanecarboxylic acids or esters rapidly decompose with release of ring strain to form the corresponding imino compounds which hydrolyze during workup giving the keto compounds finally isolated. Other reports[104] have confirmed the instability of this combination of functional groups.

Scheme 15

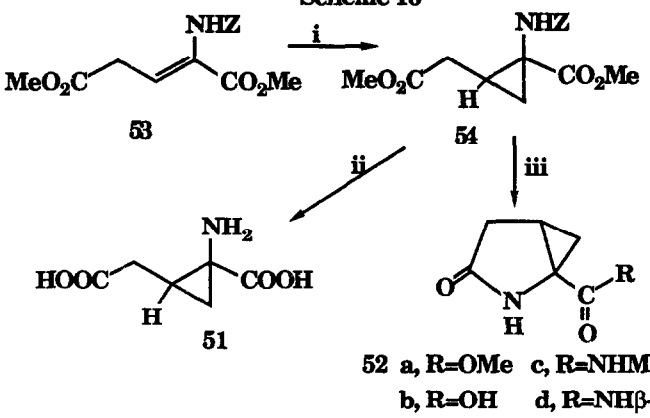


Reagents: i, N_2CHCO_2R''

The synthesis of racemic **(Z)-2,3-methanoglutamic acid** (**51**), reported recently[65,105], also allowed preparation of racemic 2,3-methanopyroglutamic acid[105b,c] (**52b, Scheme 16**). Cyclopropanation of a (Z)-dehydroglutamic acid derivative (**53**), prepared by the reaction of benzyloxycarbonylamide with 2-ketoglutaric ester, was accomplished by addition of diazomethane to **53** followed by photolysis of the resulting pyrazoline. Acidic hydrolysis of the cyclopropane (**54**) gave the free amino acid in good yield. Hydrogenolysis of the benzyloxycarbonyl group of **54** followed by ring closure in boiling *sec*-butyl alcohol gave the **2,3-methanopyroglutamic acid** ester (**52a**) which was readily hydrolyzed to the desired 2,3-methanopyroglutamic acid **52b**. An x-ray structure of the ester **52a** and an NMR study of the conformation of N-methylamide of **52c** have also appeared.[105b,c] Significantly, the β-naphthylamide (**52d**) was shown to be stable to enzymatic hydrolysis by pyroglutamate aminopeptidase *in vitro* .[105b]

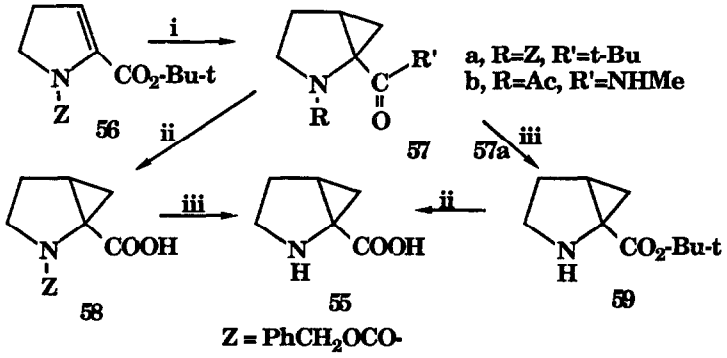
The synthesis of racemic **2,3-methanoproline** (**55**) has been accomplished recently[106] by diazomethane cyclopropanation of the 2,3-dehydroproline derivative **56** (**Scheme 17**) followed by deblocking. A careful NMR study of the N-acetyl N-methylamide of **57b** indicated that the 2,3-methano compound showed a slightly greater preference for the cis amide bond at the ring nitrogen

Scheme 16



Reagents: i, (a) CHN_2, (b) hν; ii, HCl/H_2O; iii, (a)H_2/Pd/C, (b) Δ; (c) $NaOH/H_2O$.

Scheme 17



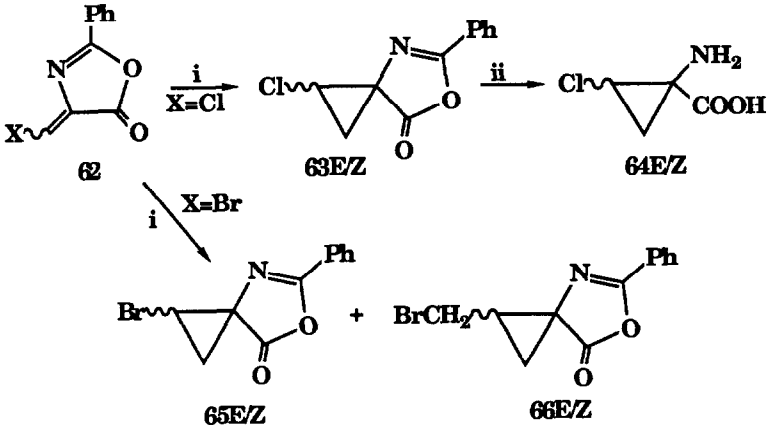
Reagents: i, (a) CH_2N_2, (b) hν; ii, CF_3CO_2H; iii, H_2/Pd

atom than does the same derivative of proline. An x-ray crystal structure of **57b** showed a very small(7°) ψ angle as compared to the same derivative of proline (16°), but an almost identical φ angle (76°). Racemic 2,3-methanoproline was found to be a weak inhibitor of ethylene forming enzyme in cucumber cotyledons and squash seeds.

Both **2,3-methanohomoserine** (**60**) and **2,3-methanomethionine** (**61**) were prepared in the process of investigating the cyclopropanation of heteromethylidene-oxazolones in the author's laboratories. Both (E)- and (Z)-diastereomers of **2-chloro-1-aminocyclopropanecarboxylic acid** (**64E/Z**) were obtained from **63** (**Scheme 18**) by acid catalyzed hydrolysis.[107] Even though only a 1:1 mixture of E- and Z-isomers of cyclopropanes (**63**) was obtained upon cyclopropanation of **62, X=Cl**, when **X=Br** five compounds, readily separated by chromatography, were obtained, with the insertion products, **66E/Z**, predominating. The (E)- and (Z)-isomers of **66** were separately converted into the methylthio

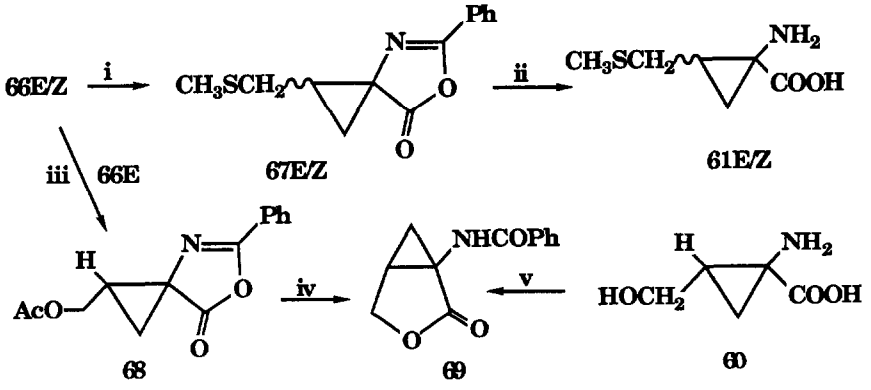
derivatives, **67E/Z**, which were hydrolyzed to form the diastereomers of 2,3-methanomethionine[108] (**61, Scheme 19**). In order to prove the configurations of these compounds, the presumed E-isomer (by NMR) of **66** was converted into the lactone **69**, prepared also from (E)-2,3-methanohomoserine, **60.** An independent synthesis of **60** ,in which alkylation of a glycine Schiff base with epibromohydrin gave a lactone convertible directly into **60**, showed conclusively that the 2,3-methanohomoserine obtained had the E-configuration.

Scheme 18



Reagents: i, CH_2N_2; ii, HCl/H_2O.

Scheme 19



Reagents: i, CH_3SNa/CH_3OH; ii, HCl/H_2O; iii, $KOAc/H_2O$; iv,HCl; v, (a) PhCOCl/NaOH, (b) HCl.

4. Peptides Containing 2,3-Methanoamino Acids

The first reported synthesis of a peptide containing a 2,3-methanoamino acid was that of **Tyr-Acc-Gly-Phe-Leu**, an analog of Leu^5-enkephalin[109], in which Acc replaced a glycine residue. In that investigation, the ease of coupling at the Acc amino and carboxyl functions was compared with that of 2-methylalanine (Aib). The Acc amino and carboxyl groups are more sterically hindered than those of normal α-amino acids, but less so than those of Aib. Acc underwent blocking, deblocking and coupling reactions uneventfully.

Another bioactive peptide containing Acc, **Acc^7-oxytocin**, was synthesized[110] with the finding that Na/NH_3(liq) deblocking of the cysteine sulfur atoms led to reductive opening of the cyclopropane ring, but that the ring was stable to anhydrous hydrogen fluoride. This peptide analog had lower bioactivity than oxytocin with dissociation of the uterotonic and galactogogic activities.

In 1984, it was reported that, during work on a series of aspartyl dipeptides containing 1-aminocycloalkanecarboxylic acids at the carboxyl terminus, **Asp-Acc-OMe** having a distinct sweet taste[111], was synthesized. Work on a series of esters, **Asp-Acc-OR**, carried out in the author's laboratories in 1984 showed[112] that the n-propyl ester, the sweetest of the series, had 250-300 times the sweetness potency of sucrose.

Significantly, two dipeptide derivatives, N-benzoyl-Acc-Phe-OH (**70**) and N-benzoyl-Acc-Pro-OH (**71**), showed time-dependent inhibition of carboxypeptidase A.[113] Thus incubation of the phenylalanine peptide **70** caused complete inactivation of the enzyme ($t_{1/2}$=4.5 min) following first order kinetics with an apparent $K_i=8.4 \times 10^{-4}$ M. The inhibition occurred about 2.3 times faster than hydrolysis of the peptide. The proline peptide, **71**, was more potent, having a $t_{1/2}$ for total inactivation of 3 min. and an apparent K_i of 5.5×10^{-4} M with no hydrolysis occurring, as expected. Using molecular graphics, it was postulated that the Zn^{++} atom, known to be present at the active site of carboxypeptidase A, coordinated with the peptide carbonyl group thus assisting the attack of the Glu^{270} carboxylate anion, an enzyme-situated nucleophile, on the cyclopropane ring leading to covalent bonding with the peptide and irreversible inactivation.

Substitution of conformationally restricted amino acids in the 2-position of the chemotactic peptide For-Met-Leu-Phe-OMe has recently been examined.[114] Ring containing amino acids, including Acc were placed in the 2-position of this peptide to determine their effects on bioactivity and conformation. Infrared spectroscopy indicated an intramolecular hydrogen bond in both the Acc^2 and Aib^2 peptides in contrast to the other conformationally constrained analogs. Both of these compounds were essentially inactive in the β-glucosaminidase assay.

Semi-empirical potential energy calculations performed on a simple Acc dipeptide and a 2-substituted Acc dipeptide indicated that different types of helices are energetically favored.[115] Potential energy maps showed that β-substituents may restrict either the ϕ or ψ values selectively.
Using both empirical and *ab initio* methods, the conformational effects of introducing Acc into a peptide chain have been studied.[116] The concensus of this work indicated that the Acc residue is quite different from the aminoisobutyric acid (Aib) residue since it favors folding of the peptide chain by formation of a C_7-helix or γ-turn with formation of a β-turn being a close second energetically. More recently, tri- and tetra-Acc peptides have been shown to fold into distorted type I β-bends and

3_{10} helices in contrast to the acyclic Aib, α,α-diethyl- and α,α-dipropylglycine residues which favor formation of regular type III β-bends and 3_{10}-helical structures.[117]

Both (-)-(2S,3R)- and (+)-(2R,3S)-(E)-2,3-methanophenylalanine have been incorporated into an enkephalin analog forming **[D-Ala^2, Leu^5, (-) and (+)-∇^{E}Phe^4]-enkephalins**[1]. These peptides were reported to show reduced activity in the mouse vas deferens (MVD) and guinea pig ileum (GPI) muscle assays[118], the peptide showing a strong preference for the δ–receptor of the GPI. It was later found that the peptide containing the 2R,3S-isomer showed a very high preference for the δ-receptor in rat brain, while the other peptide was inactive[119] indicating a possible differentiation between the peripheral and central nervous system receptors by these peptides. The synthesis of all four diastereomeric enkephalins containing the four stereoisomers of 2,3-methanophenylalanine at the four-position has been described[120]. Consistent with the high binding affinity of the Δ^{Z}Phe^4-enkephalin[1] investigated earlier[121], the **∇^{Z}Phe^4-enkephalins** showed stronger muscle receptor binding affinities than the E compounds. In this publication, it was assumed that the (-)-isomers of the methanoamino acids had the 2S-configurations while the (+)-isomers were configured (2R); these assignments are now known to be correct. Conjugation of the benzene ring with the carbonyl function is seen in the ultraviolet spectra of these amino acids; the (Z)-isomers show the greater effect probably due to the trans arrangement of the affected groups. All four of these peptides are stable to both chymotrypsin and carboxypeptidase Y hydrolysis *in vitro*. An x-ray structure[122] of the C-terminal dipeptide, **N-benzyloxycarbonyl-(+)-(E)-2,3-methanophenylalaninyl-L-leucine methyl ester**, proved the absolute configuration of the (+)-isomer to be 2R,3S and showed that the cyclopropane ring was twisted about 10° so that the phenyl and carbonyl groups avoid perfect eclipsing. A review[123] including a discussion of the above modified enkephalins has appeared.

With the expectation that at least one of the four compounds would be sweet and allow a close mapping of the "sweetness receptor", all four diastereoisomers of **Asp-∇Phe-OMe** were prepared[124] as analogs of **aspartame**, Asp-Phe-OMe. Surprisingly, all of these compounds were tasteless. NMR, CD and molecular modelling studies of these peptides led to the conclusion that the rigid positioning of the phenyl moiety by the cyclopropane ring prevented its necessary orthogonality[125] to the flat zwitterionic ring formed by the aspartic acid residue, thus prohibiting their binding to the taste receptor.

The incorporation[126] of (Z)-2,3-methanophenylalanine into the natural chlorosis-causing peptide, **tentoxin, *cyclo*[N(Me)Ala-Leu-(Z)-N(Me)–Δ^{Z}Phe-Gly]**, replacing the dehydrophenylalanine residue, led to a peptide with no bioactivity in the lettuce seedling assay. Tentoxin, a secondary metabolite of *Alternaria alternata*, is of agricultural interest because it selectively affects susceptible weeds while ignoring major crop plants.

A very recent report[127] indicates that among a series of cyclopropane peptides, ∇^{E}Phe-X-OR where X=Phe or Leu and R=H or Me, (2R,2S)-∇^{E}Phe-Phe-OMe was the best inhibitor of chymotrypsin with a K_i of 1.6 x 10^{-4} M. No hydrolysis of the ester functions was observed.

Racemic 2,3-methanopyroglutamic acid (**52b**) has been used[128] to replace the natural amino acid at the N-terminus of thyroliberin (**TRH**) giving a mixture of distereomeric peptides significantly more stable than TRH to hydrolysis by pyroglutamyl aminopeptidase, known to degrade the native hormone. An NMR study of the peptide mixture, (±)–**∇Glp-His-Pro-NH$_2$**, showed that the rigid 2,3-

MeGlp residue was close to the C_4-H of the imidazole ring and the cis (at the His-Pro bond) content was very close to that of TRH. The mixture also showed a remarkable enhancement in central nervous system (CNS) activities (3-20 times) relative to TRH in three different *in vivo* bioassays.[129] The hormonal prolactin releasing activity, on the other hand, was greatly reduced. These intriguing biological results were attributed to stabilization of the "$Y_{1,2}$" conformation[130], confirmed by the NMR results, in the cyclopropane analog and, at least in part, to the observed increase in stability of the ∇Glp-His bond.

Conclusion

Cyclopropane amino acids are extremely interesting compounds. These amino acids may be found to inhibit, by various mechanisms, amino acid processing enzymes of medicinal interest. They also have potential as conformation restricting moieties in bioactive peptides causing the stabilization of the peptide toward enzyme cleavage. The presence of the strained electrophilic cyclopropane ring may lead to covalent attachment of the peptide to an enzyme active site leading to demise of enzymatic activity. If the true conformational effects of a cyclopropane amino acid on a peptide can be ascertained, receptor and/or active site mapping may be possible. Aspects like these and, I'm sure, others to be discovered make these compounds ripe for further exploitation.

Notes and References

1 The symbol ∇^Z or ∇^E prefixed to the abbreviation for an amino acid residue, as in ∇^ZPhe, means the Z-diastereomer of 2,3-methano- or cyclopropane-phenylalanine. It is used here only when the methanoamino acid appears in a peptide chain. The Δ^Z symbol indicates a dehydro amino acid as in Δ^ZPhe, meaning (Z)-2,3-dehydrophenylalanine.

2 Fowden, L., Lea, P. J. and Bell, E. A., "The Nonprotein Amino Acids of Plants", Advances in Enzymology, ed. by A. Meister, Wiley, New York, **1979**, pp. 117-175.

3 Burroughs, L. F.,Nature, **1957**, 360.

4 Adams, D. O. and Yang, S. F., Proc. Natl. Acad. Sci. USA, **1979**, *76*, 170.

5 Yang, S. F. in Ethylene, An Int. Symp., Ed. by Y. Fuchs and E. Chalutz, Nijhoff, The Hague, Neth, **1984**, 1-10.

6 Prochazka, Z., Chem. Listy, **1985**, *79*, 1042.

7 (a) Yang, S. F., Adams, D. O., Lizada, C., Yu, Y., Bradford, K. J., Cameron, A. C. and Hoffman, N. E. in Plant Growth Substances **1979**, Ed. by Skoog, K. F.,Springer, Berlin, 1980, pp. 219-229; (b) Lieberman, M., "Biosynthesis and Action of Ethylene" in Ann. Rev. Plant Physiol., **1979**, *30*, 533.

8 Yang, S. F., Hoffman, N. E., McKeon, T., Riov, J., Kao, C. H. and Yung, K. H. in Plant Growth Substances, **1982**, Ed. by P. F. Waring, Academic Press, New York, N. Y., pp. 239-248.

9 Ingold, C. K., Sako, S. and Thorpe, J. F., J. Chem. Soc., **1922**, *121*, 1177.

10 (a) Connors, T. A. and Ross, W. C. J., J. Chem. Soc., **1960**, 2119; (b) Cremlyn, R. J. W., J. Chem. Soc., **1962**, 3977; (c) X. Zhu and Q. Guo, Youji Huaxue, **1985**, 153; C. A. **103**(25): 214881g.

11 Wheeler, T. N. and Ray, J. A., Synthetic Commun., **1988**, *18*, 141.

12 Schöllkopf, U., Harms, R. and Hoppe, D., Liebigs Ann. Chem., **1973**, 611.

13 (a) Ramalingam, K., Kalvin, D. and Woodard, R. W., J. Labelled Compd. Radiopharm., **1984**, *21*, 833; (b) Subramanian, P. K., Kalvin, D. M., Ramalingam, K. and Woodard, R. W., J. Org. Chem., **1989**, *54*, 270.

14 O'Donnell, M. J. and Polt, R. L., J. Org. Chem., **1982**, *47*, 2663.

15 O'Donnell, M. J., Bruder, W. A., Eckrich, T. M., Shullenberger and Staten, G. S., Synthesis, **1984**, 127.

16 (a) Addlington, R. M., Baldwin, J. E. and Rawlings, B. J., J. Chem. Soc., Chem. Commun., **1983**, 290; (b) Baldwin, J. E., Adlington, R. M. and Rawlings, B. J., Tetrahedron Lett., **1985**, *26*, 481.

17 Bregovec , I. and Jakovcic, T., Monatsh. Chem., **1972**, *103*, 288.

18 Suzuki, M., Gooch, E. E. and Stammer, C. H., Tetrahedron Lett., **1983**, *24*, 3839.

19 (a) King, S. W., Riordan, J. M., Holt, E. M. and Stammer, C. H., J. Org. Chem., **1982**, *47*, 3270; (b) Kimura, H. and Stammer, C. H., J. Org. Chem., **1983**, *48*, 2440.

20 Hiyama, T. and Kai, M., Tetrahedron Lett., **1982**, *23*, 2103.

21 (a) Procházka, Z., Budesinsky, M., Smoliková, J., Trska, P. and Jost, K., Collect. Czech. Chem. Commun., **1982**, *47*, 2291; (b) Logusch, E. W., Tetrahedron Lett., **1986**, *27*, 5935; (c) DuBois, G. E., Crosby, G. A., McGarraugh, G. V., Ng, S. Y. W., Stephenson, R. A., Wang, P. C. and Wingard, Jr., R. E., J. Org. Chem., **1982**, *47*, 1319.

22 Srazewski, P. and Tamm, C., Synthesis, **1987**, 298.

23 Rinderknecht, H. and Niemann, C., J. Am. Chem. Soc., **1951**, *73*, 4259.

24 Rich, D. H. and Tam, J. P., Synthesis, **1978**, 46.

25 Haener, R. and Seebach, D., Chimia, **1985**, *39*, 356.

26 Seebach, D., Häner, R. and Vettiger, T., Helv. Chem. Acta, **1987**, *70*, 1507.

27 (a) Schöllkopf, U., Nozulak, J. and Groth, U., Tetrahedron, **1984**, *40*, 1409; (b) Schöllkopf, U., Tiller, T. and Bardenhagen, J., Tetrahedron, **1988**, *44*, 5293.

28 (a) Subramanian, P. K. and Woodard, R. W., Pep.: Struct. Funct., Proc. Am. Pept. Symp., 9th, Ed. by Dever, C. M., Hruby, V.J. and Kopple, K. D., Pierce Chem. Co.: Rockford, Ill.,**1985**, 437; (b) Subramanian, P. K. and Woodard, R. W., J. Org. Chem., **1987**, *52*, 15; (c) Subramanian, P. K., Ramalingam, K., Norton, S. and Woodard, R. W., Spectrosc. Lett., **1986**, *19*, 1059.

29 Valle, G., Crisma, M., Toniolo, C., Holt, E. M., Tamura, M., Bland, J. and Stammer, C. H., Int. J. Peptide Protein Res., **1989**, *34*, 56.

30 Hoffman, N. E., Yang, S. F., Ichihara, A. and Sakamura, S., Plant Physiol., **1982**, *70*, 195.

31 Peiser, G. D., Wang, T. T., Hoffman, N. E., Yang, S. F., Liu, H. and Walsh, C. T., Proc. Natl. Acad. Sci. U.S.A., **1984**, *81*, 3059.

32 (a) Pirrung, M. C. and McGeehan, G. M., J. Org. Chem., **1983**, *48*, 5143; (b) Pirrung, M. C., J. Am. Chem. Soc., **1983**, *105*, 7207; (c) Pirrung, M. C., Bioorg. Chem., **1985**, *13*, 219.

33 Pirrung, M. C. and McGeehan, G. M., J. Am. Chem. Soc., **1986**, *108*, 5647.

34 Pirrung, M. C., J. Org. Chem., **1987**, *52*, 4179.

35 Baldwin, J. E., Adlington, R. M., Lajoie, G. A. and Rawlings, B. J., J. Chem. Soc., Chem. Commun., **1985**, 1496; for a recent discussion of reactions of cypropanamines with hypochlorite, see Vaidyanathan, G. and Wilson, J. W., J. Org. Chem., **1989**, *54*, 1815.

36 Baldwin, J. E., Jackson, D. A., Adlington, R. M. and Rawlings, B. J., J. Chem. Soc., Chem. Commun, **1985**, 206.

[37] Vaidyanatathan, G. and Wilson, J. W., J. Org. Chem., **1989**, *54*, 1810.

[38] Baldwin, J. E., Adlington, R. M., Rawlings, B. J. and Jones, R. H., Tetrahedron Lett., **1985**, 485.

[39] Honma, M. and Shimomura, T., Agric. Biol. Chem., **1978**, *42*, 1825.

[40] (a) Walsh, C., Pascal, Jr., R. A., Johnston, R., Raines, M., Dikshit, R. D., Krantz, A. and Honma, M., Biochemistry, **1981**, *42*, 7509; (b) Lui, H., Auchus, R. and Walsh, C. T., J. Am. Chem. Soc., **1984**, *106*, 5335.

[41] Hill, R. K., Prakash, S. R., Wiesendanger, R., Angst, W., Martinoni, B., Arigoni, D., Liu, H. W. and Walsh, C. T., J. Am. Chem. Soc., **1984**, *106*, 795.

[42] (a) Khani-Oskouee, S., Jones, J. P. and Woodard, R. W., Biochem. Biophys. Res. Commun., **1984**, *121*, 181; (b) Wiesendanger,R., Martinoni, B., Boller, T. and Arigoni, D., J. Chem. Soc., Chem. Commun., **1986**, 238; (c) Wiesendanger, R., Martinoni, R. B., Boller, T. and Arigoni, D., Experientia, **1986**, *42*, 207.

[43] Khani-Oskouee, S., Ramalingam, K., Kalvin, D. and Woodard, R. W., Biorg. Chem., **1987**, *15*, 92.

[44] (a) Pirrung, M. C. and Trinks, U. P., J. Chem. Soc., Chem. Commun., **1989**, 857; (b) Pirrung, M. and McGeehan, G. M., Angew. Chem. Int. Ed. Engl., **1985**, *24*, 1044.

[45] Erion, M. D. and Walsh, C. T., Biochemistry, **1987**, *26*, 3417.

[46] Nadler, V., Kloog, Y. and Sokolovsky, M., European J. Pharm., **1988**, *157*, 115.

[47] (a) Hassall, C. H., Reyle, K. and Feng, P., Nature, **1954**, *173*, 356; (b) Hassall, C. H. and Reyle, K., Biochem. J., **1955**, *60*, 334.

[48] (a) Anderson, H. V., Johnson, J. L., Nelson, J. W., Olson, E. C., Speeter, M. E. and Vavra, J. J., Chemistry & Ind., **1958**, 330;(b) de Ropp, R. S., Van Meter, J. C., De Renzo, E. C., McKerns, K. W., Pidacks, C., Bell, P. H. Ullman, E. F., Safir, S. R., Fanshawe, W. J. and Davis, S. B., J. Amer. Chem. Soc., **1958**, *80*, 1004; (c) Ellington, E. V., Hassall, C. H. and Plimmer, J. R., Chemistry & Ind., **1958**, 329; (d) Hort, C. and Leppla, W., Angew. Chem., **1958**, *70*, 25; (e) Renner, U., Jöhl, A. and Stoll, W. G., Helv. Chim. Acta, **1958**, *41*, 588; (f) Wilkinson, S., Chemistry & Ind., **1958**, 17; (g) Eloff, J. N. and Fowden, L., Phytochemistry, **1970**, *9*, 2423.

[49] Gray, D. O. and Fowden, L., Biochem. J., **1962**, *82*, 385.

[50] Carbon, J. A., Martin,W. B. and Swett,L. R. , J. Am. Chem. Soc., **1958**, *80*, 1002.

[51] (a) Black, D. K. and Landor, S. R., Tetrahedron. Lett., **1963**, 1065; (b) Black, D. K. and Landor, S. R., J. Chem. Soc. (C), **1968**, 288.

[52] Ichihara, A., Shiraishi, K., Sato, H., Sakamura, S., Nishiyama, K., Sakai, R., Furusaki, A. and Matsumoto, T., J. Am. Chem. Soc., **1977**, *99*, 636.

[53] Ichihara, A , Shiraishi, K. and Sakamura, S., Tetrahedron. Lett., **1977**, 269.

[54] Jorgenson, E. C., Tetrahedron. Lett., **1971**, 863.

[55] Unlikely if one assumes that the S-configuration of the α-carbon atom of a cyclopropane amino acid corresponds to that of a natural L-amino acid.

[56] Ichihara, A., Shiraishi, K., Sakamura, S., Furusaki, A., Hashiba, N. and Matsumoto, T., Tetrahedron Lett., **1979**, 365.

[57] Shiraishi, K., Ichihara, A. and Sakamura, S., Agric. Biol. Chem., **1977**, *41*, 2497.

[58] Marco, J.L., Heterocycles, **1987**, *26*, 2579.

[59] (a) Ichihara, A., Kimura, R., Yamada, S. and Sakamura, S., J. Am. Chem. Soc., **1980**, *102*, 6353; (b) Jung, M. E. and Hudspeth, J. P., J. Am. Chem. Soc., **1980**, *102*, 2463.

[60] Mitchell, R. E., Pirrung, M. C. and McGeehan, G. M., Phytochemistry, **1987**, *26*, 2695.

[61] Parry, R. J. and Mafoti, R., J. Am. Chem. Soc., **1986**, *108*, 4681.

[62] Honma, M., Shimomura, T., Shiraishi, K., Ichihara,A. and Sakamura, S., Agric. Biol. Chem., **1979**, *43*, 1677.

[63] Shiraishi, K., Konoma, K., Sato, H., Ichihara, A., Sakamura, S., Nishiyama, K. and Sakai, R. Agric. Biol. Chem., **1979**, *43*, 1753.

[64] Wakamiya, T., Nakamoto, H. and Shiba, T., Tetrahedron Lett., **1984**, *25*, 4411.

[65] Wakamiya, T., Oda, Y., Fujita, H.and Shiba, T., Tetrahedron Lett., **1986**, *27*, 2143.

[66] Fowden, L., Smith, A., Millington, D. S. and Sheppard, R. C., Phytochemistry, **1969**, *8*, 437.

[67] Fowden, L., MacGibbon, C. M., Mellon, F. A. and Sheppard, R. C., Phytochemistry, **1972**, *11*, 1105.

[68] Landor, S. R., Landor, P. D. and Kalli, M., J. Chem. Soc. Perkin Trans. I, **1983**, 2921.

[69] Pellicciari, R., Curini, M., Natalini, B. and Ceccherelli, P., Ninth International Symposium on Medicinal Chemistry, West Berlin, **1986**, 118.

[70] Blocking groups and details of the synthesis have not been published in full.

[71] Kurokawa, N. and Ohfune, Y., Tetrahedron Lett., **1985**, *26*, 83.

[72] Yamanoi, K. and Ohfune, Y.,Tetrahedron Lett.,**1988**, *29*, 1181.

[73] Fujimoto, Y., Irreverre, F., Karle, J. M., Karle, I. L. and Witkop, B., J. Am. Chem. Soc., **1971**, *93*, 3471.

[74] Umezawa, H., Muraoka, Y., Fujii, A., Naganawa,H. and Takita, T., J. Antibiot., **1980**, *33*, 1079.

[75] Kato, K., Takita, T. and Umezawa, H., Tetrahedron Lett., **1980**, *21*, 4925.

[76] (a) Awad, W. I., Fateen, A. K. and Zayed, M. A., Tetrahedron, **1964**, *20*, 891; (b) Pages, R. A. and Burger, A., J. Med. Chem., **1966**, *9*, 766; (c) Pages, R. A. and Burger, A., *ibid.* **1967**, *10*, 435; (d) Elkaschef, M. A. F., Abdel-Megeid, F. M. E. and Yassin, S. M. A., J. Prakt. Chem., **1974**, *316*, 363; (e) Mustafa, A., Asker, W., Harhash, A. H. and Fleifel, A. M., Tetrahedron, **1965**, *21*, 2215.

[77] Carter, H. E. "Azlactones", in Organic Reactions, Vol. III, ed. by Adams, R., Wiley, New York, N.Y., **1946**, pp 198-239.

[78] Kochetkov, N. K., Budovskii, E. I., Khomutov, R. M., Ya. Karpeiskii, M. and Severin, E. S., General Chem., **1960**, 2557.

[79] (a) Prokof'ev, E. P. and Karpeiskaya, E. I., Tetrahedron Lett., **1979**, 737; (b) Brocklehurst, K., Price, H. S. and Williamson, K., J. Chem. Soc., Chem. Comm., **1968**, 884.

[80] (a) Brown, A. G. and Smale, T. C., J. Chem. Soc., Chem. Comm., **1969**, 1489; (b) Morgerstern, A. P., Schutij, C. and Nauta, W. Th., J. Chem. Soc., Chem. Comm., **1969**, 321.

[81] Brocklehurst, K., Bywater, R. P., Palmer, R. A. and Patrick, R., J. Chem. Soc., Chem. Comm., **1971**, 632.

[82] Hines, J. W., Breitholle, E. G., Sato, M. and Stammer, C. H., J. Org. Chem., **1976**, *41*, 1466.

[83] Bernabe, M., Fernandez-Alvarez, E. and Ullate, S. P., Anales de Quim. **1972**, *68*, 501, 1005.

[84] (a) King, S. W., Riordan, J. M., Holt , E. M. and Stammer, C. H., J. Org. Chem.,**1982**, *47*, 3270; (b) King, S. W., Dissertation submitted to the University of Georgia in partial fulfillment of the requirements for the Ph.D. degree, **1981**.

[85] (a) Tatsuoka, S. and Morimoto, A.,Yakugaku Zasshi, **1950**, *70*, 253; (b) Rao, Y. S. and Filler, R., Synthesis, **1975**, 749; also see (a) Rao, Y., J. Org. Chem., **1976**, *41*, 722; (b) Ullman, E. F. and Baumann, N., J. Am. Chem. Soc., **1970**, *92*, 5892.

[86] Fernandez, D., Pilar de Frutos, M., Marco, J. L., Fernandez-Alvarez, E. and Bernabe, M., Tetrahedron Lett., **1989**, *30*, 3101.

[87] Bernabe, M., Fernandez, O. C. and Fernandez-Alvarez, E., Anales de Quim., **1979**, *75*, 977.

[88] Arenal, I., Bernabe, M., Fernandez-Alvarez, E. and Gibello, A. M., Anales de Quim., **1983**, *79c*, 65.

[89] (a) Arenal, I., Bernabe, M., Fernandez-Alvarez, E., Izquierdo, M. L. and Penades, S., J. Heterocyclic Chem., **1983**, *20*, 607; (b) Cativiela, C., Diaz de Villegas, M. D. and Meléndez, E., J. Heterocyclic Chem., **1985**, *22*, 1655;

[90] Arenal, I., Bernabe, M., Cuevas, O. and Fernandez-Alvarez, E., Tetrahedron, **1983**, *39*, 1387.

[91] Cativiela, C., Diaz de Villegas, M. D., Mayoral, J. A. and Melendez, E., J. Org. Chem., **1985**, *50*, 3167.

[92] Suzuki, M., Orr, G. F. and Stammer, C. H., Bioorganic Chem., **1987**, *15*, 43.

[93] Suzuki, M., Kumar, S. D. and Stammer, C. H., J. Org. Chem., **1983**, *48*, 4769.

[94] Tsuge, O., Noguchi, M. and Moriyama, H., Heterocycles, **1981**, *16*, 209

[95] Izquierdo, M. L., Arenal, I , Bernabe, M. and Fernandez-Alvarez, E., Tetrahedron, **1985**, *41*, 215.

[96] Kaiser, C. and Zirkle, C. L., U. S. Patent, 3,313,842, Apr. 11, 1967; Chem. Abstr., **1965**, *63*, 149796.

[97] Shenvi, A. B., Confalone, P. N., Bockovich, N., Bergman, J. and O'Brien, J., American Chemical Society National Meeting, Toronto, Canada, **1987**, Section D, Organic Chemistry, paper #268.

[98] Mapelli, C., Turocy, G., Switzer, F. L. and Stammer, C. H., J. Org. Chem., **1989**, *54*, 145.

[99] Srivastava, V. P., Roberts, M., Holmes, T. and Stammer, C. H., J. Org. Chem., **1990**, in press.

[100] De Kimpe, N., Sulmon, P., Brunet, P , Lambein, F. and Schamp, N., Tetrahedron Lett., **1989**, *30*, 1863.

[101] Park, J-A, M. S. Thesis, presented in partial fulfillment of the requirements for a Master of Science Degree to the University of Georgia, **1988**.

[102] Unpublished results from the author's laboratories.

[103] (a) Cannon, J. G., Rege, A. B., Gruen, T. L. and Long, J. P., J. Med. Chem., **1972**, *15*, 71; (b) Cannon, J. G. and Garst, J. E. , J. Org. Chem., **1975**, *40*, 182.

[104] (a) Shroff, C.C., Stuart, W. S., Uhm, S. J. and Wheeler, J. W., J. Org. Chem., **1971**, *36*, 3356; (b) Kricheldorf, H., Makromolekulare Chem., **1973**, *173*, 13; (c) Gebhardt-Schmitz, A., Mennicken, H., Schmitz, F., Lipp, P. and Lipp, M., Tetrahedron Lett., **1962**, 227.

[105] (a) Elrod, L. F. III, Dissertation submitted to the Graduate School of the University of Georgia in partial fulfillment of requirements for the Ph. D. degree, **1987**; (b) Elrod, L. F. III, Holt, E. M., Mapelli, C. and Stammer, C. H., J. Chem. Soc., Chem. Commun., **1988**, 252; (c) Mapelli, C., Elrod, L. F. III, Holt, E. M. and Stammer, C. H., Tetrahedron, **1989**, *45*, 4377.

[106] Switzer, F. L., Van Halbeek, H., Holt , E. M. and Stammer, C. H., Tetrahedron, **1990**, Tetrahedron, **1989**, *45*, 6091.

[107] Bland, J. M., Varughese, K. I. and Stammer, C. H., J. Org. Chem., **1984**, *49*, 1634.

[108] Bland, J. M., Shah, A., Bortolussi, A. and Stammer, C. H., J. Org. Chem., **1988**, *53*, 992.

[109] Stewart, F. H. C., Aust. J. Chem., **1981**, *34*, 2431.

[110] Prochazka, Z., Lebl, M., Barth, T., Hlavacek, J., Trka, A., Budesinsky, M. and Jost, K., Coll. Czech. Chem. Comm., **1984**,*49*, 642.

[111] Tsang, J. W., Schmeid, M., Nyfeler, R. and Goodman, M., J. Med. Chem., **1984**, *27*, 1663.

[112] Mapelli, C., Newton, M. G., Ringold, C. E. and Stammer, C. H., Int. J. Peptide Protein Res., **1987**, *30*, 498.

[113] Ner, S. K., Suckling, C. J., Bell, A. R. and Wrigglesworth, R., J. Chem. Soc., Chem. Commun., **1987**, 480; for further discussion of inhibition of enzymes by cyclopropanes, see Breckenridge, R. J. and Suckling, C. J., Tetrahedron, **1986**, *42*, 5665 and Suckling, C. J., Biochem. Soc. Trans., **1986**, *14*, 402.

[114] Toniolo, C., Crisma, M., Valle, G., Bonora, G. M., Polinelli, S., Becker, E. L., Freer, R. J., Sudhanand, Rao, R. B., Balaram, P. and Sukumar, M. , Peptide Res., **1989**, *2*, 275.

[115] Varughese, K. I., Srinivasan, A. R. and Stammer, C. H., Int. J. Peptide Protein Res., **1985**, *26*, 242.

[116] (a) Barone, V., Franternali, B., Cristinziano, P. L., Lelj, F. and Rosa, A., Biopolymers, **1988**, *27*, 1673; (b) Toniolo, C. and Benedetti, E., ISI Atlas of Science, **1988**, 225; (c) Toniolo, C., Crisma, M., Valle, G., Bonora, G. M., Barone, V., Benedetti, E., DeBlasio, B., Pavone, V., Pedone, C., Santini, A., and Lelj, F., Peptide Chemistry, **1987**, 45.

[117] (a) Benedetti, E., DiBlasio, B., Pavone, V., Pedone, C. and Santini, A., Biopolymers, **1989**, *28*, 175-184; (b) Benedetti, E., DiBlasio, B., Pavone, V., Pedone, C., Santini, A., Barone, V., Fraternali, F., Lelj, F., Bavoso, A., Crisma, M. and Toniolo, C., Int. J. Biol. Macromol., **1990**, in press; (c) Crisma, M., Bonora, G. M., Toniolo, C., Barone, V., Benedetti, E., DiBlasio, B., Pavone, V., Pedone, C., Santini, A., Fraternali, F., Bavoso, A. and Lelj, F., Int. J. Biol. Macromol., **1990**, in press.

[118] Kimura, H., Stammer, C. H., Shimohigashi, Y., Ren-Lin, C. and Stewart, J., Biochem. Biophys. Res. Comm., **1983**, *115*, 112.

[119] Shimohigashi, Y., Costa, T., Pfeiffer, A., Herz, A., Kimura, H. and Stammer, C. H., FEBS Lett., **1987**, *222*, 71.

[120] Mapelli, C., Kimura, H. and Stammer, C. H., Int. J. Peptide Protein Res., **1986**, *28*, 347.

[121] (a) English, M. L. and Stammer, C. H., Biochem. Biophys. Res. Comm., **1978**, *85*, 780; (b) Chipkin, R. E., Stewart, J. M. and Stammer, C. H., *ibid.*, **1979**, *87*, 890.

[122] Varughese, K. I. , Wang, C. H., Kimura, H. and Stammer, C. H., Int. J. Peptide Protein Res., **1988**, *31*, 299.

[123] Shimohigashi, Y., Stammer, C. H. and Costa, T. "Synthetic Enkephalin Analogs: Designing New Drugs of Pharmacological Interest" in Synthetic Peptides in Biotechnology, A. R. Liss, Inc., **1988**, pp 203-223.

[124] Mapelli, C., Stammer, C. H., Lok, S., Mierke, D. F. and Goodman, M., Int. J. Peptide Protein Res., **1988**, *32*, 484.

[125] Goodman, M. J. Coddington, D. F. Mierke and W. D. Fuller, J. Am. Chem. Soc., **1987**, *109*, 4712.

[126] Edwards, J. V., Dailey, Jr., O. D., Bland, J. M. and Cutler, H. G.,"Approaches to Structure-Function Relationships for Naturally Occurring Cyclic Peptides" in ACS Symp. Series, **380**, "Biologically Active Natural Products" ed. by Cutler, H. G., American Chem. Soc., Washington, D. C., **1988**, 35.

[127] Ogawa, T., Shimohigashi, Y., Yoshitomi, H., Sakamoto, H., Kodama, H., Waki, M. and Stammer, C. H., Peptide Chemistry, **1988**, 25.

[128] Mapelli, C., Elrod, L. F., Holt, E. M., Switzer, F. L. and Stammer, C. H., Biopolymers, **1989**, *28*, 123.

[129] Mapelli, C., Iturrian, B. and Stammer, C. H., unpublished results.

[130] Ward, D. J., Finn, P. W., Griffiths, E. C. and Robson, B., Int. J. Peptide Protein Res., **1987**, *30*, 253.