53. Further *C*-Alkylations of Cyclotetrapeptides *via* Lithium and Phosphazenium (P4) Enolates: Discovery of a New Conformation

by **Dieter Seebach***, **Olivier Bezençon**[1]), **Bernhard Jaun**, **Thomas Pietzonka**[1]), **Jennifer L. Matthews**[2]), **Florian N. M. Kühnle** and **W. Bernd Schweizer**

Laboratorium für Organische Chemie der Eidgenössischen Technischen Hochschule, ETH-Zentrum, Universitätstrasse 16, CH–8092 Zürich

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Four cyclotetrapeptides containing one (**1**, **2**) or two (**3**, **4**) chiral amino acids have been *C*-alkylated or *C*-hydroxyalkylated through Li^+ or phosphazenium ($P4 \cdot H^+$) enolates. The reactions are completely diastereoselective (by NMR or HPLC analysis) with respect to the newly formed backbone stereogenic centres (*Tables 2* and *3*). The reactivity of the polylithiated species responsible for these alkylations is such that only highly reactive electrophiles (MeI, BnBr, primary allylic halides, aldehydes, CO_2) can be employed. It is shown that the position, and thus the chirality sense, of the newly formed stereogenic centre in a given cyclotetrapeptide backbone is controlled by the positioning of *N*-methyl groups in the starting material (*cf.* cyclo(-MeLeu-Gly-D-Ala-Sar-) (**3**) and cyclo(-Leu-Sar-MeDAla-Gly-) (**4**) in *Scheme 1*). With *Schwesinger*'s phosphazene P4-base, all NH groups are first benzylated and *C*-benzylation then takes place at a sarcosine, rather than an *N*-benzylglycine residue (*Table 3*). In contrast to open-chain *N*-benzyl peptides, the *N*-benzylated cyclotetrapeptides could not be debenzylated under dissolving-metal conditions (Na/NH_3). Conformational analysis (NMR spectroscopy and X-ray diffraction) shows that the prevailing species have *cis*/*trans*/*cis*/*trans* (*ctct*) peptide bonds (zigzag conformation of C_i backbone symmetry, *Figs. 2–4*). However, a hitherto unknown conformation of cyclotetrapeptides has been found in $CDCl_3$ solutions of the hydroxyalkylated products **18–21** (obtained with EtCHO and PhCHO as electrophiles; *Fig. 4*). The new conformation has four *trans* peptide bonds and is believed to result mainly from intramolecular H-bond formation, involving the newly generated alkyl- or arylserine residue. This assumption has also been supported by modelling (TRIPOS force field, SYBYL, see *Fig. 5* and *Table 6*). The structure may be considered as a β-turn mimic.

1. Introduction. – Cyclic tetrapeptides have been isolated from a variety of sources and some have been shown to exhibit significant biological activity[3]). Due to the potential uses in a wide range of chemical applications, the synthesis of cyclic tetrapeptides is an area of considerable interest [5] [6]. Yields of cyclisation reactions are often low and, therefore, extremely costly in terms of the preparation of the often non-proteinogenic amino acids found within the skeletons of these cyclotetrapeptides. It is often possible to enhance the yields by the presence of glycine, D-amino acids, sarcosine or cyclic amino acids within the chain[4]) and it is therefore advantageous to synthesize less highly substituted cyclic peptides and introduce the substituents at a later stage. This

[1]) Part of the Ph. D. Theses of *O. B.* (No. 11350, 1995) and *T. P.* (No. 10000, 1992).

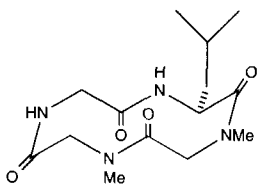
[2]) Royal Society (GB) Postdoctoral Research Fellow, ETH Zürich, 1995/96.

[3]) Two recent examples are cyclo(-Pro-Tyr-Pro-Val-), a potent tyrosine inhibitor [1] and trapoxin, cyclo(-L-phenylalanyl-L-phenylalanyl-D-pipecolinyl-L-2-amino-8-oxo-9,10-epoxydecanoyl-), an inhibitor of histone deacetylase [2]. For other examples, see [3] [4].

[4]) *E.g.*, the tripeptide Pro-Pro-Pro cyclises readily [7].

amounts to the modification of a given peptide rather than its synthesis from the corresponding components[5]).

In previous work, we have demonstrated [3] that modification of the cyclic tetrapeptides cyclo(-Leu-Sar-Sar-Gly-) (**1**), cyclo(-Val-Sar-Sar-Gly-) (**2**) and cyclo(-MeLeu-Gly-D-Ala-Sar-) (**3**) is possible. In all cases, reaction of a sarcosine enolate moiety adjacent to another *N*-methylamino acid gave, with highly reactive electrophiles, cyclic tetrapeptides with additional substituents and in good diastereoselectivity. As a continuation of this 'modification' strategy[6]), we set out to demonstrate that the stereoselective preparation of a series of cyclic tetrapeptides with either (*R*)- or (*S*)-configuration at the modified residue is possible.



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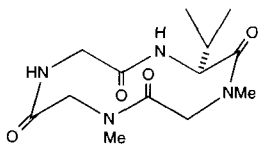

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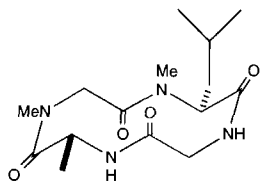

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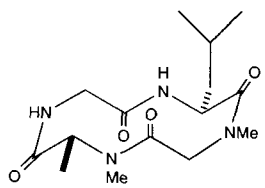

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We now report the preparation of cyclo(-Leu-Sar-MeDAla-Gly-) (**4**), a cyclic tetrapeptide which, if the *N*-methyl groups are not taken into account, has the same amino-acid sequence as cyclo(-MeLeu-Gly-D-Ala-Sar-) (**3**). This new cyclopeptide should, however, be alkylated from the *Si*-face. Furthermore, in order to achieve the ultimate goal of this work (the synthesis of cyclopeptides with opposite configurations), we have investigated the preparation and alkylation of *N*-benzylated cyclotetrapeptides and possible subsequent debenzylation procedures, and we now report further on these results[7]). In the final part, we discuss aspects of the conformational analysis of these cyclic tetrapeptides and describe the first example of a new conformation of a cyclic tetrapeptide which has an OH group on the newly introduced side chain.

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[5]) We envisage that this could become a method of building up a so-called library of cyclic peptides with varying substitution patterns [8].

[6]) This strategy has been discussed in a recent review [9].

[7]) A short communication has appeared on this topic [10] and it has been briefly mentioned as part of a longer review article on peptide modifications [9].

2. Preparation of Cyclo(-Leu-Sar-MeDAla-Gly-) (4). – In an analogous fashion to our previous work [3], this peptide was constructed in the N to C direction using Boc-protected amino acids and using BOP-Cl as the coupling reagent (see the linear precursors **5**–**12** in *Table 1*). The third peptide coupling, involving an easily epimerisable Me-D-Ala residue at the C-terminus, was performed at −10° with less than 5% epimerisation but in only 40% yield. The pentafluorophenyl ester, which has been recommended by *Schmidt* and coworkers [11] and subsequently by *Sheh* and *Mokotoff* [12], was used as activation for the cyclisation step and yielded the desired cyclopeptide **4** in 30–35% yield from Boc-Leu-Sar-MeDAla-Gly-OH.

Table 1. *Synthesis of the Linear Precursors of Cyclo(-Leu-Sar-MeDAla-Gly-)* (**4**).
The coupling agent was BOP-Cl (in CH_2Cl_2) and the deprotections were accomplished by catalytic hydrogenation (H_2, 10% Pd/C in MeOH). The pentafluorophenyl(PFP)-active ester was prepared by reaction of the peptide with pentafluorophenyl trifluoroacetate in pyridine.

Product No.	Leu	Sar	Me-D-Ala	Gly	Yield [%]
	Boc—OH H	—OBn			
5	Boc—	—OBn			60
6	Boc—	—OH H	—OBn		97
7	Boc—	—	—OBn		84
8	Boc—	—	—OH H	—OBn	98
9	Boc—	—	—	—OBn	40
10	Boc—	—	—	—OH	97
11	Boc—	—	—	—OPFP	[a])
12	H—	—	—	—OPFP	[a])

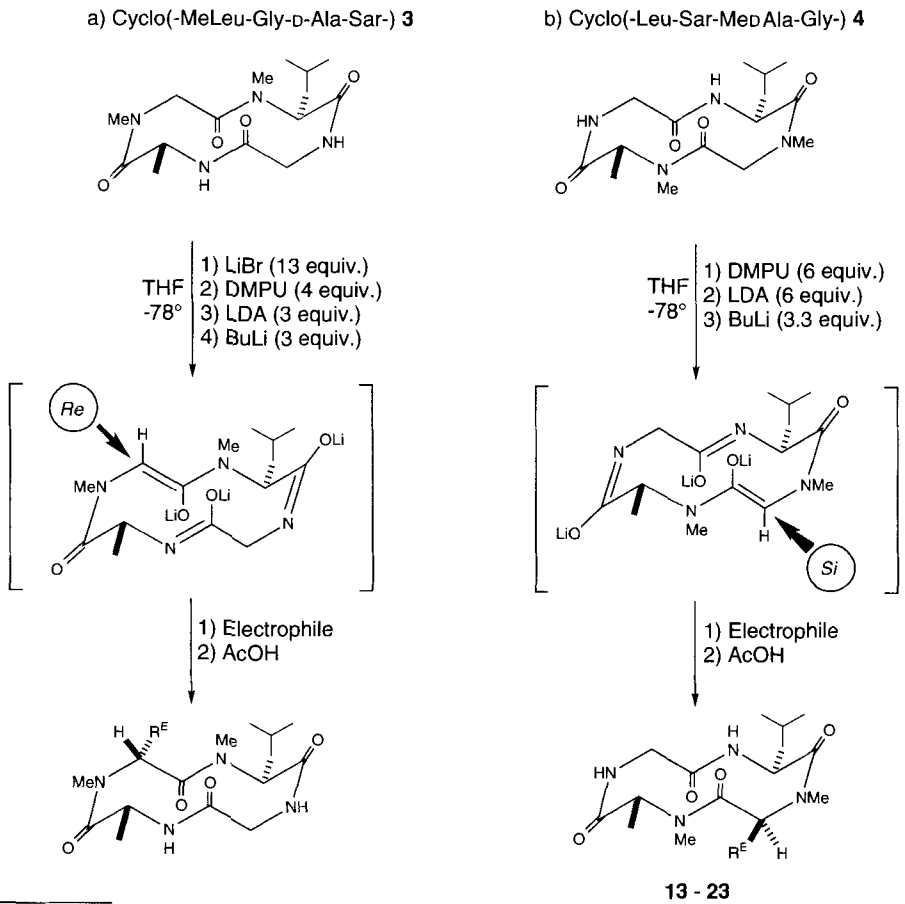
[a]) Used in next step without purification.

3. Solubility of Cyclo(-Leu-Sar-MeDAla-Gly-) (4). – It has been well established that the solubility of small peptides in THF, the solvent of choice for deprotonation and subsequent alkylation reactions, is usually strongly dependent upon the presence of salts [9]. We have also shown that the effect of LiBr on the solubility of cyclotetrapeptides **1**–**3** is highly ambiguous. Whereas the addition of LiBr resulted in the expected increase in solubility of **1** in THF at both room temperature and at −78°, adding LiBr to a solution of **2** or **3** decreased their solubility at room temperature and increased it at −78°. A study of the solubility of the cyclotetrapeptide **4** also led to different results: The addition of 2 equiv. of LiBr lowered the solubility from 6–9 mg/ml at room temperature, or from 3–4 mg/ml at −78°, to less than 1 mg/ml at both temperatures. At present, the reasons behind these observations are not fully understood[8]).

[8]) For a discussion, see the recent review article [9].

4. *C*-Alkylations of Cyclo(-Leu-Sar-MeDAla-Gly-) (4). – As the addition of LiBr to a solution of **4** results in the cyclic peptide becoming so insoluble (< 1 mg/ml), the alkylation experiments were performed in THF in the presence of 6 equiv. of 3,4,5,6-tetrahydro-1,3-dimethylpyrimidin-2(1*H*)-one (DMPU) without any additional Li salts. The solubility of this cyclotetrapeptide is so sensitive to the Li^+ concentration that even the addition of 6 equiv. of $(i\text{-Pr})_2NLi$ (LDA) as part of the alkylation procedure immediately provoked a turbidity, which is most likely to be a colloidal suspension of the trianionic Li-enolate of the peptide. This suspension does, however, react smoothly with alkylating reagents[9]). As 1–1.5 equiv. of H_2O, which could not be removed[10]), were present in the cyclopeptide, the addition of 6 equiv. of LDA was necessary to remove the proton

Scheme 1. *C-Alkylation of Cyclo(-MeLeu-Gly-D-Ala-Sar-)* (**3**) *and Cyclo(-Leu-Sar-MeDAla-Gly-)* (**4**) *where Alkylation Occurs from Opposite Faces.* For the specification of R^E, see *Table 2.*



[9]) The addition of LiBr at any stage of the reaction resulted in the cyclic tetrapeptide becoming so insoluble that no alkylation reactions occurred.

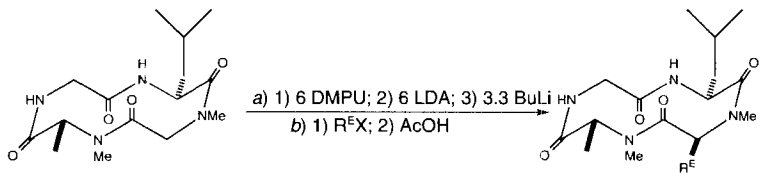
[10]) This was verified by both NMR and elemental analysis.

from C(α) of Sar and a further 3.3 equiv. of BuLi was added prior to the addition of the electrophile to prevent retrodonation of the proton by $(i\text{-Pr})_2NH$.

In our previous work [3], we speculated that the stereochemical outcome of the alkylation occurs due to the replacement of an outer H-atom in the α-position of the C=O group with retention of configuration. In the case of cyclo(-MeLeu-Gly-D-Ala-Sar-) (**3**), alkylation occurs at the *Re*-face to give products of (*R*)-configuration at the newly generated stereogenic centre. In a similar fashion, the reaction of cyclo(-Leu-Sar-MeDAla-Gly-) (**4**) occurs with replacement of an outer H-atom, but alkylation now takes place at the *Si*-face, hence generating products of (*S*)-configuration at the new centre of chirality (*Scheme 1*).

The yields (*Table 2*) of both deuteration using CF_3COOD (*Entry 1*) and alkylation with MeI or allyl bromide (*Entries 2* and *3*) of **4** are comparable to those obtained with

Table 2. *Alkylation of* **4** *with Various Electrophiles in THF at −78° in the Presence of DMPU.* For determination of the conformations, see *Sect. 6.*



4 **13–23**

Entry	Electrophile	Product			
		Number	R^E	Yield[a]) [%]	Conformation[b])
1	CF_3CO_2D	**13**	D	90[c])	*ctct*[d])
2	MeI	**14**	Me	76	*ctct*
3	CH_2=$CHCH_2Br$	**15**	CH_2=$CHCH_2$	51	*ctct*
4	MeCH=$CHCH_2Br$	**16**	MeCH=$CHCH_2$	48	*ctct*
5	Me_2C=$CHCH_2Br$	**17**	Me_2C=$CHCH_2$	80	*ctct*
6	1-(Bromomethyl)cyclohexene			[e])	
7	3-Bromocyclohex-1-ene			[e])	
8	(*E*)-1,4-Dibromobut-2-ene			[f])	
9	1,3-Bis(bromomethyl)benzene			[f])	
10	EtCHO	**18**	EtCH(OH)	(*S*,*R*): 24	*ctct*/*tttt* 1:1
		19	EtCH(OH)	(*S*,*S*): 28	*tttt*
11	PhCHO	**20**	PhCH(OH)	(*S*,*R*): 39[g])	*ctct*/*tttt* 15:85[h])
		21	PhCH(OH)	(*S*,*S*): 30	*tttt*
12	CO_2/CH_2N_2	**22**	MeO_2C	97	*ctct*/*xxxx*[i]) 35:65
13	AcCl	**23**[j])	H[j])	13[j])	*ctct*

[a]) Yields determined by HPLC after workup.
[b]) In $CDCl_3$ solution, see also *Sect. 6.*
[c]) D-Incorporation, determined by NMR.
[d]) Also in CD_3OD and in D_2O.
[e]) Intractable mixture of starting material and traces of monoalkylated product.
[f]) Intractable mixture of starting material with some monoalkylated product and traces of dialkylated product.
[g]) Absolute configuration determined by X-ray crystal-structure analysis (see *Sect. 6*).
[h]) *ctct* only in C_2D_5OD (see also *Sect. 6*).
[i]) There was insufficient data available to confirm whether a new conformer was present or whether formation of a different diastereoisomer had occurred.
[j]) R^E = MeC(O) at the N(α) of Gly; yield of the purified main product cyclo(-Leu-Sar-MeDAla-AcGly-).

3. Alkylation also proved to be possible with MeCH=CHCH$_2$Br and Me$_2$C=CHCH$_2$Br (*Entries 4* and *5*). In all of the previous four cases, the monoalkylated product was obtained with a diastereoselectivity greater than 95:5, the limit of detection by HPLC and, as was seen for **3**, only one conformation was observed[11]). Acylation of the cyclopeptide with AcCl (*Entry 13*) gave a mixture of products with the main one, cyclo(-Leu-Sar-MeDAla-AcGly-) (**23**), being isolated in 13% yield. GC Analysis of the single amino-acid residues derived from hydrolysis of **14** showed the presence of Leu, Me-L-Ala, Me-D-Ala and Gly in equal proportions. This is proof that the intermediate enolate is alkylated from the *Si*-face.

Attempts at alkylation with other alkyl halides demonstrate the present limitations of this 'modification' strategy [9]. As less reactive electrophiles were used, the yields rapidly decrease. Hence, the sterically more hindered 1-(bromomethyl)cyclohexene (*Entry 6*) and the secondary bromide 3-bromocyclohex-1-ene (*Entry 7*) did not react. Furthermore, the use of dielectrophiles such as (*E*)-1,4-dibromobut-2-ene (*Entry 8*) or 1,3-bis(bromomethyl)benzene (*Entry 9*) failed to yield isolable products. This is presumably a result of the need to use more than 10 equiv. of the electrophile to induce a reasonable conversion rate.

The use of aldehydes as the electrophile resulted in the formation of substituted serine derivatives[12]) (*Entries 10* and *11*). No stereoselectivity was observed with respect to the configuration at C(β) of the new side chain and all efforts to induce some selectivity at this position by the use of other metal derivatives (Ti or Mg) failed[13]).

Given the lack of stereoselectivity observed in the reactions of aldehydes, we decided to adopt an alternative strategy for the stereoselective introduction of functionality at the new side chain. Previous work in our group has shown that, as H–C(α) of the 2-amino-malonate residue of a linear oligopeptide is the most acidic, deprotonation with bases such as KO(*t*-Bu) or NaOMe and subsequent alkylation with a variety of electrophiles is possible [13]. Hence, the cyclotetrapeptide **4** was successfully converted to the methyl ester **22** of the (2*R*)-2-aminomalonic-acid derivative (*Entry 12*) by carboxylation with CO_2 and esterification with CH_2N_2. It is important to note that the ethereal CH_2N_2 had to be added at −78° immediately after quenching the reaction with AcOH. When no CH_2N_2 was added or when it was added after warming to room temperature, only the non-carboxylated starting material was isolated. This suggests that the malonic-acid derivative is able to adopt a conformation which facilitates a rapid decarboxylation under acidic conditions[14]). Unfortunately, attempts to employ this compound **22** for the further substitution of the cyclopeptide failed[15]).

Further attempts at the introduction of functionality on the new side chain lay with the reactions of the cyclopeptides **15**–**17**. We attempted to epoxidise the C=C bond in the

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[11]) For a more detailed discussion of the conformations of the alkylated products, see *Sect. 6*.

[12]) With tetradecanal ($C_{13}H_{27}CHO$), no hydroxyalkylation was observed. Upon addition of the electrophile, the solution became cloudy and only starting materials were recovered.

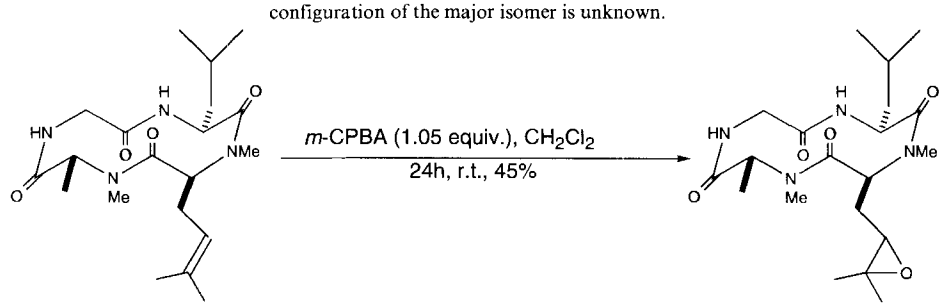
[13]) Interestingly, the addition of $TiCl_4$ resulted in the precipitation of a green material which suggests that the peptide trianion may have reduced Ti^{IV} to Ti^{III}.

[14]) The *ctct* conformation with an equatorial $R^E = CO_2H$, as drawn in *Scheme 1* and *Table 2*, would of course be the ideal one for decarboxylation. Interestingly, the major conformer of the corresponding methyl ester does not have this conformation, see *Entry 12* of *Table 2*.

[15]) Probably, too much steric hindrance would be introduced in an alkylation of our aminomalonate enolate generating a geminally disubstituted cyclotetrapeptide.

side chain of these three cyclopeptides. Reactions of **15** with 3-chloroperbenzoic acid (*m*-CPBA) in CH_2Cl_2, *t*-BuOOH/$TiCl_4$ [14], or PhSe(O)OH/H_2O_2 [15] failed. A very slow reaction was observed between **16** and *m*-CPBA, but, after 24 h, products of decomposition predominated and isolation of a pure compound from the reaction mixture proved to be impossible. However, the reaction of the methylbutenyl derivative **17** with *m*-CPBA (*Scheme 2*) proceeded in 45% yield to give a major diastereoisomer **24** (9:1) as an oil[16])[17]).

Scheme 2. *Epoxidation of* **17** *with* m-*CPBA*. Two diastereoisomers were formed in a ratio of 9:1. At present, the configuration of the major isomer is unknown.



17 **24**

5. *N*- and *C*-Benzylations of Cyclotetrapeptides with the Phosphazene P4-Base. – The major drawback of the 'Li-enolate peptide modification' strategy lies in the necessity to direct the site of the reactions by the use of *N*-methylated compounds. These *N*-methyl groups not only cause complications during synthesis of the precursors but there is also no method available for their removal whilst leaving the backbone of the cyclic peptide intact[18]). One way out would be to use a removable *N*-substituent.

In a previous short communication [10], we have mentioned that *N*-benzylation of cyclic peptides is possible using the metal-free, non-nucleophilic P4-base (*Table 3*) developed by *Schwesinger* and *Schlemper* [17] [18] and we now report on this in more detail. Cyclo(-Leu-Sar-Sar-Gly-) (**1**) was treated with 8 equiv. of benzyl bromide and 3 equiv. of P4-base at −100°. This yielded a mixture[19]) of di-*N*-benzylated product **25** (46% yield) and the tribenzylated product **26** (20% yield) (*Table 3*), with the two products being easily separated by chromatography[20]). The product which arises from a final *C*-benzylation was shown to be a single diastereoisomer. Similarly, the cyclopeptides **2** and **3** were

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[16]) At present, we have been unable to determine the absolute configuration at the new stereogenic centre.

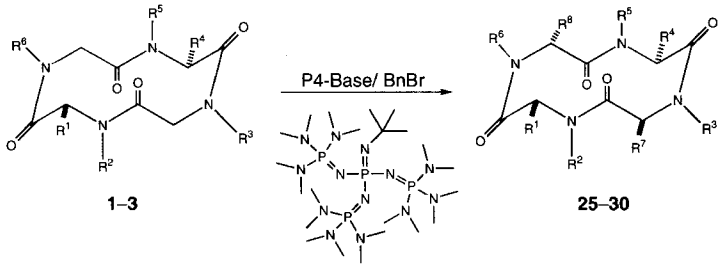
[17]) During the course of this work, other work within our group established that alkylation of a glycine residue neighbouring an *N*-alkylated amino-acid residue proceeded in higher yields than the corresponding reactions of a sarcosine residue [16]. We hoped to demonstrate the application of this approach (involving an aza-dienediolate reactive site) to the cyclopeptides. However, synthesis of cyclo(-MeLeu-MeAla-MeDAla-Gly-) proved to be impossible as the cyclisation of the *N*-methylleucine onto the glycine residue failed, with only linear polymeric material being isolated.

[18]) In proline-containing peptides, the *N*-substituent is built in and can function in the generation of an aza-dienediolate site [16].

[19]) The crystal structure of compound **25** has been solved and is discussed in *Sect. 6*.

[20]) *N*-Benzylation of these cyclic peptides dramatically increases both the solubility and the R_f values of the products.

Table 3. *Benzylation of Cyclo(-Leu-Sar-Sar-Gly-)* (**1**), *Cyclo(-Val-Sar-Sar-Gly-)* (**2**) *and Cyclo(-MeLeu-Gly-*D*-Ala-Sar-)* (**3**) *with $PhCH_2Br$/P4-Base.* Unless otherwise stated, 8 equiv. of BnBr were added to the cyclopeptide in THF at −78°, followed by 3 equiv. of P4-base at −100°. The mixture was then allowed to warm slowly to room temperature.



	R^1	R^2	R^3	R^4	R^5	R^6	R^7	R^8	Yield [%]
1	H	Me	Me	Me_2CHCH_2	H	H	–	–	–
2	H	Me	Me	Me_2CH	H	H	–	–	–
3	Me	H	H	Me_2CHCH_2	Me	Me	–	–	–
25	H	Me	Me	Me_2CHCH_2	Bn	Bn	H	H	46 (from **1**)
26	H	Me	Me	Me_2CHCH_2	Bn	Bn	Bn	H	20[a]) (from **1**)
27	H	Me	Me	Me_2CH	Bn	Bn	H	H	30[b]) (from **2**)
28	H	Me	Me	Me_2CH	Bn	Bn	Bn	H	80 (from **2**)
29	Me	Bn	Bn	Me_2CHCH_2	Me	Me	H	H	52 (from **3**)
30	Me	Bn	Bn	Me_2CHCH_2	Me	Me	H	Bn	20[a]) (from **3**)

[a]) Obtained as minor product during formation of dibenzylated product.

[b]) Obtained with 4 equiv. of BnBr and 2.3 equiv. of P4-base. The temperature was maintained at −78° for 2 h, and the reaction was then quenched.

benzylated to yield products **27**–**30** of di- and tribenzylation (*Table 3*). A variation in the proportion of reagents added in the reaction of **2** dramatically affected the ratio of di- and tribenzylation. The use of 2.3 equiv. of P4-base and 4 equiv. of BnBr yielded 30% of dibenzylated **27** and no tribenzylated product. However, 3 equiv. of P4-base and 8 equiv. of BnBr resulted in the formation of the tribenzylated **28** in 80% yield with no dibenzylated product being found. In the cases of **1** and **3**, the proportion of reagents added seems to have much less effect on the ratio of products observed; 3 equiv. of P4-base and 8 equiv. of BnBr resulted in a *ca.* 2:1 mixture of dibenzylated and tribenzylated products for **1** and a 2.5:1 mixture for **3**. A further experiment with compound **1** established that a benzyl group is introduced first at the N-atom of glycine (R^6 = Bn) and then at the N-atom of leucine (R^5 = Bn). The structure of the dibenzylation product **25** from **1** was determined by crystal-structure analysis (see *Fig. 3* in *Sect. 6*). Thus, it is possible not only to *N*-benzylate cyclotetrapeptides but also to generate peptide enolates with the phosphazene base and to achieve highly diastereoselective *C*-benzylations (a single diastereoisomer **28** was obtained from **2** in 80% yield!).

Having succeeded in our aim of introducing benzyl groups onto the cyclopeptide skeleton, we then moved on to investigate possible methods for the removal of these *N*-protecting groups. The debenzylation of linear *N*-benzyl peptides is possible using the procedure developed by *Weygand* and *Steglich* (Na/liq. NH_3) [19]. However, this proce-

dure, when applied to the cyclotetrapeptide derivatives, did not result in the removal of the *N*-benzyl groups but instead gave products arising from decomposition of the cyclopeptide skeleton. Although the very polar decomposition products (TLC) were not identified, we believe that, as the distance between the two sides of the cyclic peptide in the zigzag conformation is only 3.1 Å, it is plausible that a transannular pinacol coupling may be causing the problem.

These results mean that, at present, we have still not succeeded in finding a method for the removal of the *N*-protecting groups from the cyclotetrapeptides **25–30**. A solution to this problem might be the use of 4-methoxybenzyl groups which are known to be removed from amide N-atoms with cerium ammonium nitrate.

6. Conformational Analysis. – The analysis of the conformation of cyclopeptides has received considerable attention as a method of achieving a greater understanding about the reasons behind the biological activity of larger, often highly complex peptides and proteins[21]) [23]. Whilst cyclodipeptides and cyclotripeptides contain only *cis* amide bonds, cyclic tetrapeptides may contain *trans* amide bonds and are, therefore, capable of displaying a variety of conformations. It is not unusual to find that several conformers exist in an equilibrium mixture and complexation [24] or solvation [4] [25] [26] methods can be used

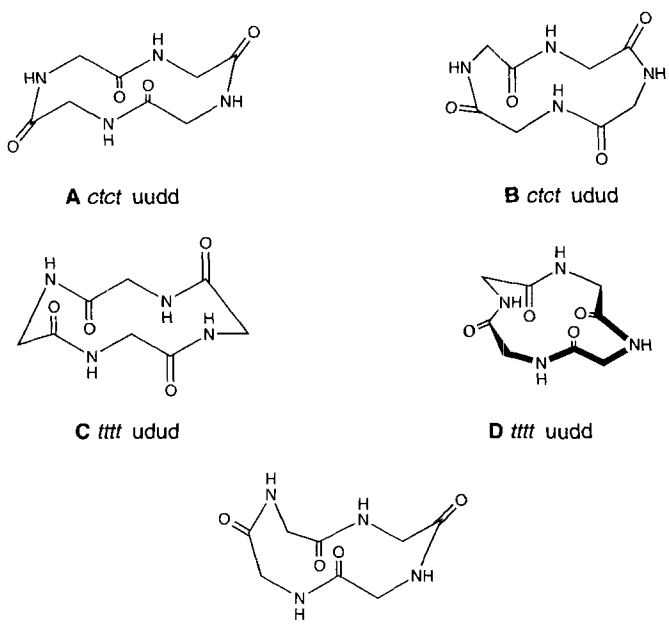


Fig. 1. *Five possible conformers of cyclotetrapeptides*: ctct *uudd* (zigzag) *conformation* **A** (symmetry C_i), ctct *udud conformation* **B** (symmetry C_2), tttt *udud conformation* **C** (symmetry C_2), tttt *uudd conformation* **D** (symmetry C_i) *and* ctct *uuud conformation* **E**

[21]) This approach has not been limited to peptides. See, *e.g.*, the study of cyclic oligomers of 3-hydroxybutanoic acid as a way of gaining new information about the possible structure of poly(hydroxybutanoic acid) ion channels [20–22].

to shift such equilibria. In the past, attempts have been made to lay down rules allowing the prediction of the conformation of a cyclopeptide with a known sequence of amino acids [27] [28]. However, these rules have found only limited use and this is maybe not so surprising when the dramatic changes in conformation arising from a very minor structural modification are taken into account [27] [29].

Calculations on cyclotetraglycine *in vacuo* [30] have been used to propose that the most stable conformer is that in which the amide bonds are all *trans* (*tttt*) and the carbonyl groups are oriented in an up-down-up-down (udud) fashion (0 kcal/mol) (see **C** in *Fig. 1*). A *cis-trans-cis-trans* (*ctct*) centrosymmetric zigzag arrangement **A** exists at 1.29 kcal/mol and a *tttt* centrosymmetric arrangement **D** at 2.21 kcal/mol.

Experimentally, *ctct* conformations of the ring backbone of cyclotetrapeptides have been shown to predominate [31]. The two most commonly occurring *ctct* conformers[22]), one with carbonyl groups oriented uudd (centrosymmetric, see **A**) and the other with the udud orientation (C_2 symmetric, see **B**), have been found in crystalline forms of cyclotetrapeptides and also in $CDCl_3$ or (D_6)DMSO solution. The *tttt* arrangements have also been experimentally observed[23]). In the majority of cases, the udud (C_2 symmetric) orientation of the carbonyl groups is observed and this is particularly true in solution[24]). Other conformations are seen far less frequently. A *cccc* arrangement has been found for cyclo(-Pro-Sar-Pro-Sar-) in the solid state [38] and in (D_6)DMSO [39]. Studies of equilibrium mixtures have led to the observation of a *ccct* conformation [27] [39] and a *cctt* orientation has also been suggested [24]. We now report on the conformational analysis of some of the cyclopeptides mentioned in the previous sections.

The NMR spectra of cyclo(-Leu-Sar-MeDAla-Gly-) (**4**) in the three solvents $CDCl_3$, CD_3OD and D_2O all suggest the presence of a single identical conformer regardless of the nature of the solvent. The chemical shifts of the backbone protons closely correspond to those reported by *Rayudu* [40] as being typical for the centrosymmetric zigzag *ctct* uudd conformation of cyclic tetrapeptides (*cf.* **A** in *Fig. 1*). The same single conformation was found for cyclic peptides **1**–**3** (described earlier [3]) and for all the alkyl derivatives of cyclo(-Leu-Sar-MeDAla-Gly-) (**4**) which have been prepared in this study. 1D-NOE Difference spectra of **14** corroborate this conformational assignment; the Leu-MeAla and MeDAla-Gly amide bonds are both *cis*, the two others *trans* (*ctct* uudd), as shown in

Table 4. *^{1}H-NMR Chemical Shifts* (δ [ppm]) *of Backbone Protons of the Cyclopeptides* **14** *and* **20** *in $CDCl_3$*

		MeXxx[a])		Me-D-Ala		Gly			Leu	
		MeN	H–C(α)	MeN	H–C(α)	NH	H_{Si}–C(α)	H_{Re}–C(α)	NH	H–C(α)
14	*ctct*	3.11	4.55	2.83	5.42	6.46	3.87	3.63	6.50	4.84
20	*ctct*	[b])	3.23	[b])	5.38	6.30	3.83	3.61	6.03	4.80
	tttt	2.53	3.70	2.64	5.32	6.91	3.29	4.41	6.51	4.75

[a]) **14**: MeXxx = MeAla; **20**: MeXxx = MeSer(3-Ph).
[b]) Could not be unambiguously assigned.

[22]) We have also reported the observation of a *ctct* uuud conformation **E** (*Fig. 1*) [3]. The unambiguous identification of this conformer has been relatively infrequently reported [29] [32] [33].
[23]) For a recent example, see [34]; see also [24].
[24]) See, *e.g.*, in CD_3CN [35], $CDCl_3$ [4] [26] [35–37] and (D_6)DMSO [37].

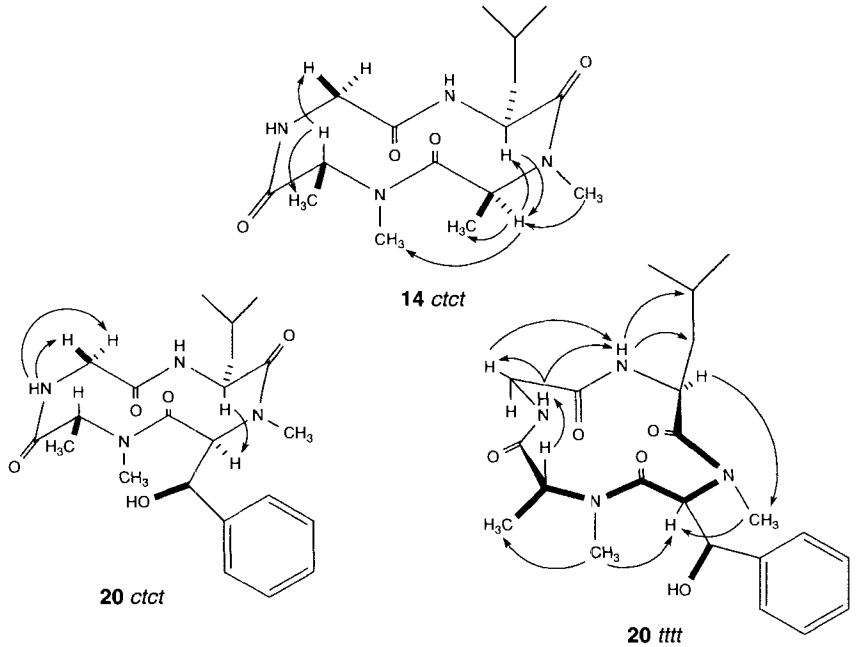


Fig. 2. *Conformations of cyclo(-Leu-MeAla-MeDAla-Gly-)* (**14**) *and cyclo(-Leu-MeSer(3-Ph)-MeDAla-Gly-)* (**20**) *as determined by NMR NOE measurements*

Fig. 2. This *ctct* uudd conformation was also found to be present in the crystalline form of the di-*N*-benzylated peptide **25** (*Fig. 3*).

However, in $CDCl_3$ solution, cyclopeptides carrying an OH group at the β-position of the newly introduced side chain (see **18–21** in *Table 2*) showed ^{1}H-NMR spectra (*Tables 4* and *5*) indicating the presence of a new conformer which is unlike any of the conformation types described so far for cyclotetrapeptides[25]). Since, in $CDCl_3$, the β-substituted serine derivatives **18–21** were all at least partially present in this unknown conformation and since we were able to crystallise from EtOH the cyclotetrapeptide **20** which carries a PhCH(OH) group, we selected this compound for a combined study by both NMR and

Table 5. *^{1}H-NMR Coupling Constants Observed Between the Protons of the Cyclopeptides* **14** *and* **20**

		Gly			Leu
		J(NH,H_{Si}–C(α))	J(NH,H_{Re}–C(α))	J(H_{Si}–C(α),H_{Re}–C(α))	J(NH,H–C(α))
14	*ctct*	8.85	7.03	17.53	9.45
20	*ctct*	8.90	7.10	17.40	8.80
	tttt	2.15	10.55	12.70	9.80

[25]) For cyclopeptides **19** and **21**, only this new conformer was observed, whereas for compounds **18** and **20** this unknown conformer coexisted with the standard *ctct* uudd form, as deduced from chemical shifts, coupling constants and NOE of the corresponding subspectra (see *Tables 4* and *5* and *Fig. 2*). A similar conformer has been observed only once previously and this was for a cyclodepsipeptide [41].

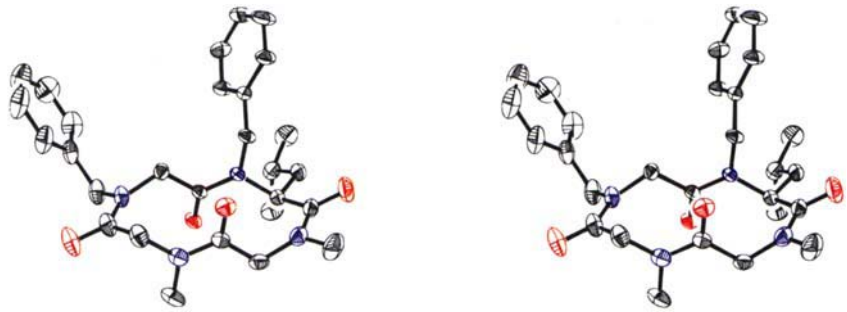


Fig. 3. *Stereo ORTEP representation of the crystal structure of* **25**. O-Atoms are shown in red, N-atoms in blue; H-atoms are omitted for clarity. The thermal ellipsoids are drawn to the 25% probability level.

X-ray crystal-structure analysis. Interestingly, the X-ray analysis revealed that in the crystalline state, cyclopeptide **20** attains the standard *ctct* uudd conformation (see *Fig. 2*) with one molecule of EtOH H-bonded to the carbonyl O-atom of Me-D-Ala (*Fig. 4*). Presuming that this intermolecular H-bond might be the reason for the different conformations in the crystal and in $CDCl_3$ solution, we checked the ^{1}H-NMR spectrum of **20** in (D_6)ethanol and found that, as in the crystals, only the *ctct* uudd conformation was detectable in this D-bonding solvent.

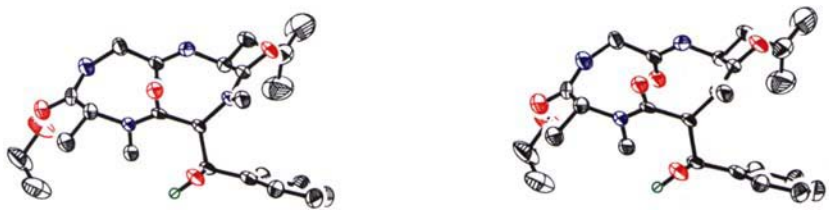


Fig. 4. *Stereo ORTEP representation of the crystal structure of* **20**. O-Atoms are shown in red, N-atoms in blue; H-atoms are omitted for clarity. The thermal ellipsoids are drawn to the 25% probability level.

The nature of the unknown major conformer of **20** found in $CDCl_3$ solution was deduced by ^{1}H-NMR analysis through coupling constants, longitudinal (NOESY and 1D-NOE difference) and transverse (ROESY) NOE spectroscopy: this conformer exhibits four *trans* amide bonds and the carbonyl O-atoms of Leu and MeSer(3-Ph) point up, whereas those of Gly and Me-D-Ala point down (*tttt* uudd), as shown in *Fig. 2* (see also **D** in *Fig. 1*).

A strong NOE between H–C(α) of Me-D-Ala and NH of Gly demonstrates that the MeDAla-Gly amide bond of the major isomer of **20**, must be *trans*. The NH of Gly exhibits a large coupling (10.6 Hz) but no NOE with one of the 2 H–C(α) of Gly, while the coupling to the other H–C(α) of Gly, exhibiting a strong NOE, is near 0 Hz (see *Table 5*). The local conformation in the Gly residue must be that shown in *Fig. 2* for the *tttt* conformation, with dihedral angles near 150 and 90° between NH and H_{Re}– and H_{Si}–C(α) of Gly, respectively. Both, NH and H_{Si}–C(α) of Gly exhibit a NOE to NH of Leu which, therefore, points to the same side of the macrocycle as NH of Gly. This is only possible if the Gly-Leu peptide bond is also *trans*. The H–C(α) of Leu shows a strong coupling but no NOE with NH of Leu which in turn shows NOE with CH_2(β) and H–C(γ) of Leu. This indicates that NH and H–C(α) of Leu are *anti*-periplanar to each other. The H–C(α) of Leu exhibits a NOE with the MeN of MeSer(3-Ph) and the MeN groups of both MeSer(3-Ph) and Me-D-Ala show strong NOE to H–C(α) of MeSer(3-Ph). The MeN of Me-D-Ala exhibits a NOE with Me(β) of Me-D-Ala but not with H–C(α) of Me-D-Ala. This

chain of NOE correlations proves that H–C(α) of Leu, MeN and H–C(α) of MeSer(3-Ph), and MeN of Me-D-Ala all point towards the same side of the macrocycle and is only consistent with *trans* amide bonds for both Leu-MeSer(3-Ph) and MeSer(3-Ph)-MeDAla.

With the help of molecular-mechanics calculations, we estimated the energy difference between the conformation *ctct* from the X-ray structure and the all-*trans* conformer of **20**. A model of the *tttt* uudd conformation was minimised using the TRIPOS force field (SYBYL [42]) and *Gasteiger-Marsili* charges [43] followed by a molecular-dynamics calculation over a period of 10000 fs starting at a temperature of 600 K. The resulting local energy minimum conformation (28.7 kcal/mol) (*Fig. 5, a*) corresponds to the all-*trans* structure. Starting from this model, a random conformation search found two *ctct* conformers with lower energy. The first one (*Fig. 5, b*), which corresponds to the X-ray

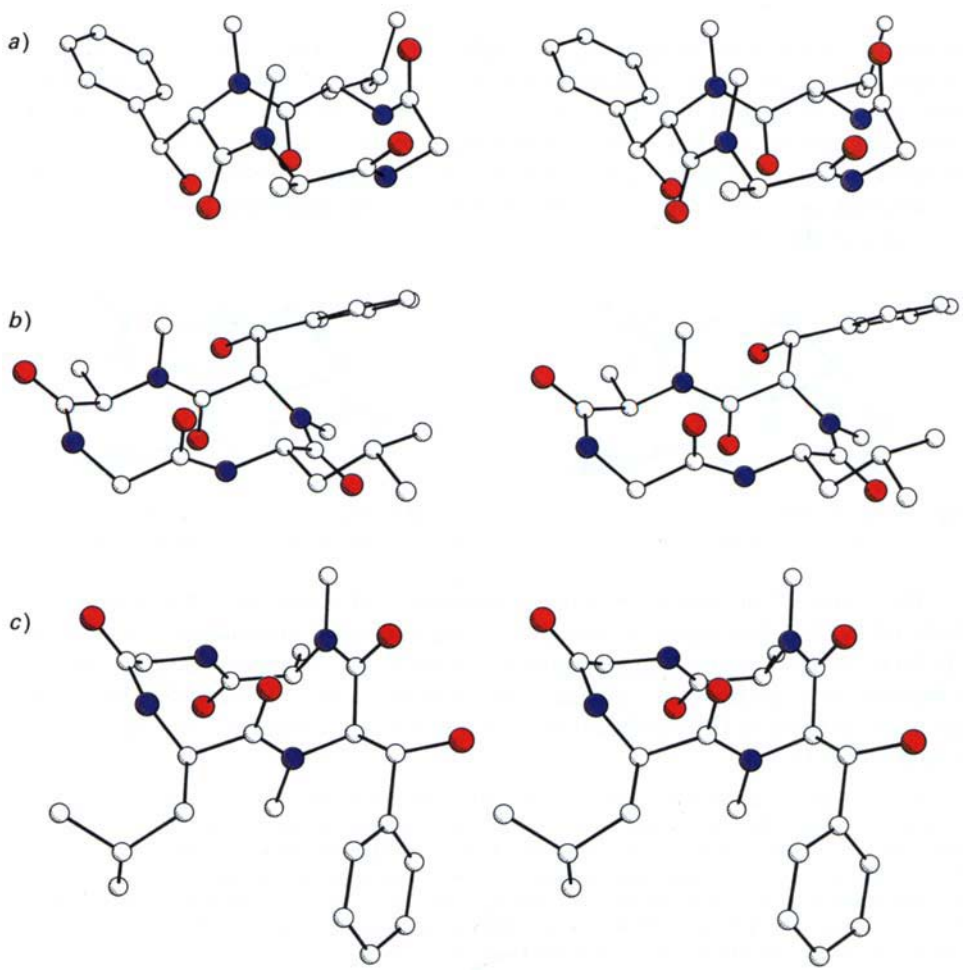


Fig. 5. *Stereo PLUTO plots of the three conformers of* **20** *found by molecular modelling:* a) *The* tttt *udud conformation* (energy = 28.7 kcal/mol), b) *the* ctct *uudd conformation* (energy = 27.1 kcal/mol) *and* c) *the* ctct *uuud conformation* (energy = 22.0 kcal/mol)

crystal structure with the carbonyl groups oriented uudd, is not significantly lower in energy (27.1 kcal/mol). The second one (*Fig. 5, c*), with the carbonyl groups uuud, is, at 22.0 kcal/mol, the lowest in energy. The energy difference between the last one, which has not been found experimentally, and the two other ones might be explained by solvation effects. For the minimised *tttt* conformation, these calculations indicated two intramolecular H-bonds, one between the alcohol function of the MeSer(3-Ph) and its adjacent carbonyl group and the second rather unfavourable one between the NH of Gly and the CO of Leu, which indicates that the sequence Leu-MeSer(3-Ph)-MeDAla-Gly may be considered as a β-turn equivalent. As the amide bonds are *N*-methylated in this region, it is not possible to determine the dihedral angles by NMR, but those of the calculated structure (*Table 6*) correspond to none of the well-defined β-turn types.

Table 6. *Calculated Dihedral Angles in the Structure of* **20**

	ϕ [°]	ψ [°]	ω [°]
Leu	−85.6	141.3	148.0
MeSer(3-Ph)	42.1	53.1	−172.3
Me-D-Ala	125.1	−71.5	−168.4
Gly	−99.7	−72.8	160.4

The interconversion of the two conformers is slow compared with T_1. No NOE exchange cross-peaks between the constitutionally corresponding CH protons were observed in the NOESY and ROESY spectra and even in the NOE difference spectra (irradiation time 2 s), no saturation transfer was observed. At 27° as well as at −10°, no other conformer such as a possible intermediate *cttt* form was observed.

7. Conclusion. – We have shown that progress towards our ultimate goal of the synthesis of identical substituted cyclopeptides of opposite configuration has been made. Furthermore, during this work, we established the existence of a new conformer of the arylserine derivative **20** in $CDCl_3$ solution. This conformer, *tttt* uudd, appears to result from the effects of two intramolecular H-bonds. These H-bonds lead to the sequence Leu-MeSer(3-Ph)-MeDAla-Gly being suggested as a potential β-turn mimic.

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Experimental Part

1. *Abbreviations*: AcGly (*N*-acetylglycine); Ahx(4-en) ((*S*)-2-aminohex-4-enoic acid); Ahx(5-Me,4-en) ((*S*)-2-amino-5-methylhex-4-enoic acid); Ape(4-en) ((*S*)-2-aminopent-4-enoic acid); BOP-Cl (bis(2-oxooxazolidin-3-yl)phosphinic chloride); *c* (*cis*); *m*-CPBA (3-chloroperbenzoic acid); DIPEA (*N,N*-diisopropylethylamine); DMAP (4-(dimethylamino)pyridine); DMPU (3,4,5,6-tetrahydro-1,3-dimethylpyrimidin-2(1*H*)-one); FC (flash chromatography); GP (general procedure); h.v. (high vacuum, 0.01–0.1 Torr); LDA (lithium diisopropylamide); Ser(3-Et) (3-ethylserine); Ser(3-Ph) (3-phenylserine); *t* (*trans*).

2. *General.* All alkylations were carried out under Ar in oven- or flame-dried glassware. Solvents used for reactions and crystallisations were purchased from *Fluka* (*puriss.*). Dry THF for alkylations was freshly distilled from K under Ar. Solvents used for extraction and FC were distilled as follows: AcOEt from P_2O_5, Et_2O from

$KOH/FeSO_4$, hexane and pentane from *Sikkon*, MeOH was purchased from *Fluka* (*puriss.*). $(i\text{-}Pr)_2NH$ was distilled from CaH_2 prior to use. DMPU (*BASF*) was distilled from CaH_2 (110°/5 Torr) and stored over activated molecular sieves (4 Å). LiBr (*Fluka, purum*) was dried at 160°/h.v. for 3 h, dissolved in THF (*ca.* 0.7M) and stored in the dark under Ar. Alkyl halides were filtered through *Alox* (*Woelm*) prior to use. Benzaldehyde and propionaldehyde were distilled from $CaCl_2$ and stored over molecular sieves. Basic *Dowex* refers to *Fluka Dowex* ion exchange resin *1* × *8* (20–50 mesh), washed with 6M NaOH, H_2O and MeOH. Acidic *Dowex* refers to *Fluka Dowex* ion exchange resin *50* × *8* (20–50 mesh), washed with 6M HCl, H_2O and MeOH. BuLi (15% in hexane, *Chemetall Gesellschaft*) was titrated according to the procedure of *Ronald* and coworkers [44] using 2,5-dimethoxybenzyl alcohol as indicator. CH_2N_2 was prepared using the method of *Boer* and *Baker* [45]. CF_3COOH was purchased from *Solvay GmbH*. Amino acids were donated by *Degussa*. Cyclo(-Leu-Sar-Sar-Gly-) (**1**), cyclo(-Val-Sar-Sar-Gly-) (**2**), and cyclo(-MeLeu-Gly-D-Ala-Sar-) (**3**) were synthesized according to previously published procedures [3]. Other chemicals were purchased from *Fluka*.

FC: *Fluka* silica gel *60*. TLC: *Merck* silica gel *60-F_{254}* anal. plates; detection by UV and/or by placing in a Cl_2 tank for 5–30 min, then staining with a soln. of bis[4-(dimethylamino)phenyl]methane [46]. Anal. HPLC: *Knauer* equipment using a reversed-phase *Nucleosil®-100-5-C_8* column (*Macherey-Nagel*, 250 × 4 mm). Prep. HPLC: *Knauer* equipment using a reversed phase *Nucleosil®-100-7-C_8* column (*Macherey-Nagel*, 250 × 100 mm). GC: hydrolyses and formation of derivatives of the peptides for GC analysis were performed as described in [47]; *Carlo-Erba-Fractovap-4160-HR-GC* equipment using a *Chirasil-Val* column (*Macherey-Nagel*, 25 m, 0.4 mm); temp. program: 5 min 160°, 2°/min, 45 min 200°. Cryostat: *Frigomix®-S* apparatus (*B. Braun*). M.p.: *Büchi-510* apparatus; uncorrected. Optical rotations: 10 cm, 1-ml cell, *Perkin-Elmer-241* polarimeter at r.t. (20°). ^1H- and ^{13}C-NMR: *Bruker-AMX-II-500*, *Bruker-AMX-400*, *Bruker-WM-300* or *Varian-Gemini-200* spectrometers; chemical shifts in ppm, coupling constants in Hz. FAB-MS: *VG-ZAB2-SEQ* in a 3-nitrobenzyl alcohol matrix in *m/z* (% of the highest peak).

3. *Preparation of the Linear Precursor. Boc-Leu-Sar-OCH_2Ph* (**5**): Boc-Leu-OH·H_2O (35.0 g, 0.140 mol) and DIPEA (49.0 ml, 0.287 mol) were dissolved in CH_2Cl_2 (700 ml) and cooled to 0°. BOP-Cl (39.5 g, 0.155 mol), H-Sar-OCH_2Ph·TosOH (54.3 g, 0.155 mol) and DIPEA (27.0 ml, 0.162 mol) were added to this soln. The mixture was allowed to warm to r.t. overnight while stirring, partially evaporated and washed twice with 1M H_2SO_4 and twice with sat. $NaHCO_3$ soln. The aq. phases were extracted with CH_2Cl_2. The combined org. phases were dried ($MgSO_4$) and evaporated and the residue was purified by FC (Et_2O/hexane 1:3, then 1:2) to yield **5** as a colourless oil that was dried overnight under h.v. (32.8 g, 60%). R_f 0.45 (AcOEt/hexane 1:2). $[\alpha]_D^{r.t.} = -16.8$ (*c* = 1.0, MeOH). ^1H-NMR (200 MHz, $CDCl_3$): 7.35 (*s*, 5 H); 5.15 (br. *s*, 3 H); 4.70 (*m*, 1 H); 4.50 (*d*, *J* = 18, 1 H); 4.00, 3.80 (2*d*, *J* = 18, 1 H); 3.15, 2.95 (2*s*, 3 H); 1.70 (*m*, 1 H); 1.45 (*s*, 9 H); 1.20 (*m*, 2 H); 1.00–0.80 (br. *m*, 6 H).

Boc-Leu-Sar-OH (**6**). Pd/C (1.6 g) was added to a soln. of **5** (31.5 g, 0.104 mol) in MeOH (200 ml) under Ar, the flask was evacuated and purged twice with H_2. The mixture was stirred overnight under H_2. Filtration through *Celite* and evaporation yielded a residue which was dried overnight under h.v.: **6** (23.5 g, 97%). Colourless oil. ^1H-NMR (200 MHz, $CDCl_3$): 6.05 (br. *s*, 1 H); 5.40 (*d*, *J* = 8, 1 H); 4.70 (*m*, 1 H); 4.40 (*d*, *J* = 18, 1 H); 3.85 (*d*, *J* = 18, 1 H); 3.15, 2.95 (2*s*, 3 H); 1.70 (*m*, 1 H); 1.45 (br. *s*, 11 H); 0.95, 0.90 (2*d*, *J* = 7, 6 H).

Boc-Leu-Sar-MeDAla-OCH_2Ph (**7**). A mixture of **6** (10.6 g, 35.0 mmol) and HCl·H-MeDAla-OCH_2Ph [23] (14.0 g, 61.0 mmol) in CH_2Cl_2 (250 ml) was cooled to −10°. BOP-Cl (15.6 g, 61.1 mmol) and DIPEA (24.6 ml, 140 mmol) were added and the mixture was allowed to warm to r.t. overnight while stirring. The soln. was washed twice with 1M H_2SO_4, twice with sat. $NaHCO_3$ soln. and once with brine. The aq. phases were extracted with CH_2Cl_2. The combined org. phases were dried ($MgSO_4$) and evaporated and the residue was purified by FC (AcOEt/hexane 1:1) to give **7** as a colourless oil which was dried overnight under h.v. (14.1 g, 84%). R_f 0.25 (AcOEt/hexane 1:1). $[\alpha]_D^{r.t.} = +10.2$ (*c* = 0.6, EtOH). ^1H-NMR (200 MHz, $CDCl_3$): 7.35 (*s*, 5 H); 5.25–5.10 (*m*, 4 H); 4.70 (*m*, 1 H); 4.40 (*d*, *J* = 18, 1 H); 4.00 (*d*, *J* = 18, 1 H); 3.10, 3.00 (2*s*, 3 H); 2.90, 2.80 (2*s*, 3 H); 1.75 (*m*, 1 H); 1.50 (br. *m*, 14 H); 1.00, 0.95, 0.85 (3*d*, *J* = 7, 6 H). FAB-MS: 478 (16, *M*H$^+$), 378 (32), 285 (11), 265 (47), 263 (14), 229 (79), 194 (27), 130 (23), 91 (72), 86 (27), 58 (25), 57 (100).

Boc-Leu-Sar-MeDAla-OH (**8**). As described for **6**, with Pd/C (0.70 g), **7** (14.0 g, 29.3 mmol) and MeOH (100 ml): **8** (11.2 g, 98%). Colourless oil. $[\alpha]_D^{r.t.} = +5.0$ (*c* = 0.75, EtOH). ^1H-NMR (200 MHz, $CDCl_3$): 5.70 (*d*, *J* = 10, 1 H); 5.20 (*m*, 1 H); 4.60 (*m*, 2 H); 3.75 (*d*, *J* = 18, 1 H); 3.15, 3.10 (2*s*, 3 H); 2.95, 2.90, 2.85 (3*s*, 3 H); 1.75 (*m*, 1 H); 1.50 (br. *m*, 14 H); 0.95 (*m*, 6 H). FAB-MS: 410 (100, *M*Na$^+$), 388 (10, *M*H$^+$), 332 (13), 288 (32), 285 (13), 230 (10), 229 (67), 225 (14), 185 (18), 175 (59), 173 (14), 130 (26), 104 (28), 100 (14), 86 (42).

Boc-Leu-Sar-MeDAla-Gly-OCH_2Ph (**9**). A soln. of **8** (5.0 g, 13 mmol) and H-Gly-OCH_2Ph·TosOH (5.0 g, 16 mmol) in CH_2Cl_2 (300 ml) was cooled to −20° and BOP-Cl (4.0 g, 16 mmol) and DIPEA (7.0 ml, 41 mmol) were added to it. The mixture was stirred for 48 h between −15 and −5° (cryostat), then washed twice with 1M H_2SO_4,

twice with sat. $NaHCO_3$ soln. and once with brine. The org. phase was dried ($MgSO_4$) and evaporated and the residue purified by FC (AcOEt/hexane 3:1). Drying the residue overnight under h.v. afforded **9** (3.7 g, 53%). Colourless oil. R_f 0.20 (AcOEt/hexane 3:1). GC (after hydrolysis and formation of derivatives): D-Ala (t_R 8 min 32 s)/L-Ala (t_R 8 min 24 s) 6:1. $[\alpha]_D^{r.t.} = +28.6$ ($c = 1.1$, EtOH). ^{1}H-NMR (200 MHz, $CDCl_3$): 7.35 (*s*, 5 H); 6.85 (*t*, $J = 7$, 1 H); 5.40–5.10 (*m*, 2 H); 5.15 (*s*, 2 H); 4.70 (*m*, 1 H); 4.45 (*d*, $J = 18$, 1 H); 4.35–3.80 (*m*, 2 H); 3.70 (*d*, $J = 18$, 1 H); 3.20, 3.00 (2*s*, 3 H); 2.90 (*s*, 3 H); 1.75 (*m*, 1 H); 1.55–1.25 (*m*, 14 H); 1.00, 0.90, 0.85 (*d* or *m*, $J = 8$, 6 H). FAB-MS: 557 (5, MNa$^+$), 535 (3, MH$^+$), 435 (23), 322 (14), 314 (30), 251 (22), 229 (56), 132 (32), 130 (21), 91 (39), 86 (18), 58 (21), 57 (100).

*Boc-Leu-Sar-Me*D*Ala-Gly-OH* (**10**). As described for **6**, with Pd/C (0.3 g), **9** (5.75 g, 10.8 mmol) and MeOH (100 ml): **10** (4.63 g, 97%). Oil. $[\alpha]_D^{r.t.} = +34.0$ ($c = 1.2$, EtOH). ^{1}H-NMR (200 MHz, $CDCl_3$): 6.95 (*t*, $J = 6$, 1 H); 5.40 (*d*, $J = 8$, 1 H); 5.25 (*q*, $J = 7$, 1 H); 4.65 (*m*, 2 H); 4.30–3.60 (*m*, 3 H); 3.15 (*s*, 3 H); 3.00, 2.95 (2*s*, 3 H); 1.75 (*m*, 1 H); 1.55–1.20 (*m*, 14 H); 1.05–0.80 (*m*, 6 H). FAB-MS: 467 (63, MNa$^+$), 445 (5, MH$^+$), 367 (15), 345 (26), 314 (25), 232 (25), 230 (12), 229 (67), 185 (19), 161 (30), 157 (11), 130 (23), 100 (15), 86 (33), 58 (38), 57 (100).

*Boc-Leu-Sar-Me*D*Ala-Gly-OC$_6$F$_5$* (**11**). A soln. of **10** (4.5 g, 10 mmol) in pyridine (40 ml) was cooled to 0°. Pentafluorophenyl trifluoroacetate [48] (3.4 g, 12 mmol) was added and the soln. allowed to warm to r.t. overnight while stirring. The solvent was evaporated, the resulting oil dissolved in Et_2O and the soln. washed once with H_2O, twice with 5% citric acid, once with brine, twice with sat. $NaHCO_3$ soln. and once with brine. The org. phase was then dried ($MgSO_4$) and evaporated: **11** as an oil (8.0 g) that was used in the next step without further purification.

*H-Leu-Sar-Me*D*Ala-Gly-OC$_6$F$_5$ Trifluoroacetate* (**12**·CF_3COOH). The crude **11** (8.0 g) was dissolved in CH_2Cl_2 (40 ml) and the soln. cooled to 0°. CF_3COOH (40 ml) was added and the mixture stirred at 0° for 100 min. The solvent was evaporated and the residue dried overnight under h.v.: **12** as a yellow-brown oil (8.3 g) that still contained 15% of pyridine (^{1}H-NMR). FAB-MS: 511 (100, M^+), 398 (21), 327 (15), 185 (23), 100 (23), 86 (38), 58 (15).

4. *Cyclisation. Cyclo(-Leu-Sar-Me*D*Ala-Gly-)* (**4**). In a 10-l flask (overhead stirrer, precision addition funnel), a soln. of dioxane (3.50 l), pyridine (0.60 l), CF_3CH_2OH (90 ml) and DMAP (0.5 g, 4.0 mmol) was heated to 90–100°. A soln. of **12** (4.1 g, 5.0 mmol from **10**) in dioxane (1.00 l) was added dropwise (1 drop/s) within 36 h, with continuous fast stirring and heating. After the addition was complete, the soln. was heated for 1 h, cooled to r.t. and evaporated. The resulting crude brown residue was filtered through basic *Dowex* in MeOH, the filtrate evaporated, the residue filtered through acidic *Dowex* in MeOH and the filtrate evaporated. Purification by FC (CH_2Cl_2/MeOH 95:5) followed by crystallisation from MeOH/hexane and purification of the mother liquors by HPLC afforded **4** as a white powder (0.61 g, 36%) that was dried overnight over P_2O_5 (110°/10^{-5} Torr). R_f 0.12 (CH_2Cl_2/MeOH 95:5). $[\alpha]_D^{r.t.} = +12.4$ ($c = 0.85$, EtOH). HPLC (H_2O/MeCN 95:5, isocratic): t_R 28.7 min. IR (KBr): 3370*s*, 3080*w*, 2955*m*, 2880*w*, 1690*m*, 1660*s*, 1615*m*, 1560*w*, 1485*m*, 1445*m*, 1400*m*, 1375*w*, 1365*w*, 1355*w*, 1315*w*, 1285*w*, 1260*m*, 1230*w*, 1220*w*, 1135*w*, 1125*w*, 1090*m*, 965*w*. ^{1}H-NMR (400 MHz, $CDCl_3$): 6.20 (*d*, $J = 9.1$, NH, Leu); 6.11 (*t*, $J = 8.0$, NH, Gly); 5.43 (*q*, $J = 6.7$, H–C(α), D-Ala); 4.80 (*m*, H–C(α), Leu); 4.37 (*d*, $J = 17.6$, H–C(α), Sar); 3.90 (*dd*, $J = 17.6$, 9.3, H–C(α), Gly); 3.62 (*d*, $J = 17.7$, H–C(α), Sar); 3.62 (*dd*, $J = 17.6$, 6.8, H–C(α), Gly); 3.08 (*s*, MeN, Sar); 2.73 (*s*, MeN, D-Ala); 1.76–1.68 (*m*, H–C(γ), Leu); 1.64 (*s*, 1.5 H, H_2O); 1.51 (*m*, CH_2(β), Leu); 1.27 (*d*, $J = 6.7$, Me(β), D-Ala); 0.94 (*t*, $J = 6.0$, 2 Me(δ), Leu). ^{1}H-NMR (200 MHz, D_2O): 5.45 (*q*, $J = 7$, H–C(α), D-Ala); 4.90 (*m*, H–C(α), Leu); 4.40 (*d*, $J = 17$, H–C(α), Sar); 4.05 (*d*, $J = 17$, H–C(α), Sar); 3.85 (*s*, CH_2(α), Gly); 3.05 (*s*, MeN, Sar); 2.70 (*s*, MeN, D-Ala); 1.75–1.40 (*m*, CH_2(β), H–C(γ)); 1.25 (*d*, $J = 8$, Me(β), D-Ala); 0.95 (*d*, $J = 8$, 2 Me(δ), Leu). ^{1}H-NMR (200 MHz, CD_3OD): 5.35 (*q*, $J = 7$, H–C(α), D-Ala); 4.85 (*m*, H–C(α), Leu); 4.35 (*d*, $J = 18$, H–C(α), Sar); 3.90 (*d*, $J = 17$, H–C(α), Sar); 3.80 (*d*, $J = 17$, H–C(α), Gly); 3.60 (*d*, $J = 17$, H–C(α), Gly); 3.05 (*s*, MeN, Sar); 2.70 (*s*, MeN, D-Ala); 2.75–1.35 (*m*, CH_2(β), H–C(γ)); 1.20 (*d*, $J = 7$, Me(β), D-Ala); 0.90 (*m*, 2 Me(δ), Leu). ^{13}C-NMR (100 MHz, $CDCl_3$): 172.2 (CO, D-Ala); 170.6 (CO, Leu); 168.3 (CO, Gly); 167.3 (CO, Sar); 50.9 (C(α), Sar); 48.6 (C(α), Leu or D-Ala); 48.2 (C(α), Leu or D-Ala); 43.8 (C(α), Gly); 41.0 (C(β), Leu); 36.8 (MeN, Sar); 29.0 (MeN, D-Ala); 24.3 (C(γ), Leu); 23.2 (C(δ), Leu); 22.1 (C(δ), Leu); 13.4 (C(β), D-Ala). FAB-MS: 349 (21, MNa$^+$), 327 (100, MH$^+$), 326 (11), 325 (15), 214 (10), 157 (15), 155 (12), 154 (21), 137 (20), 136 (20), 91 (19), 86 (36), 77 (10), 58 (33), 57 (11), 55 (11). Anal. calc. for $C_{15}H_{26}N_4O_4$: C 53.20, H 8.03, N 17.17; found: C 53.14, H 7.82, N 16.04, H_2O 3.60.

5. *C-Alkylations. GP I.* Unless otherwise stated, the following procedure was applied: Peptide **4** in THF, DMPU (6 equiv.) was added and the mixture was cooled to −78°. A 0.4M LDA soln. was prepared at −78° in THF from (i-Pr)$_2$NH and BuLi; it was stirred for 15 min and then added (6 equiv.) to the peptide soln. *via* syringe. This mixture was stirred for 2 h at −78°. BuLi (3.3 equiv.) and the electrophile (12 equiv.) were added. The soln. was stirred overnight at −78° and AcOH (0.2–0.5 ml) was added. The solvent was evaporated, the residue dissolved in MeOH and filtered through basic and then acidic *Dowex*, and the MeOH evaporated. The crude material was purified by HPLC with subsequent lyophilisation.

Cyclo(-Leu-(D$_1$)Sar-MeDAla-Gly-) (**13**). According to *GP I*, with **4** (50 mg, 0.15 mmol), DMPU (0.11 ml, 0.92 mmol), LDA (0.4M, 2.3 ml, 0.9 mmol), BuLi (1.2M, 0.42 ml, 0.5 mmol) and CF_3CO_2D (0.17 ml, 2.2 mmol). After addition of the electrophile, the mixture was immediately worked up. HPLC yielded **4**/**13** (35 mg, 70%). ^{1}H-NMR: *ca.* 90% D-incorporation. HPLC (H_2O/MeCN 95:5, isocratic): t_R 28.7 min. ^{1}H-NMR (200 MHz, $CDCl_3$): 6.20 (*d*, *J* = 10, 1 H); 6.40 (*t*, *J* = 8, 1 H); 5.35 (*q*, *J* = 7, 1 H); 4.80 (*m*, 1 H); 4.35 (*s*, 1 H); 3.90 (*dd*, *J* = 17, 10, 1 H); 3.60 (*dd*, *J* = 17, 7, 1 H); 3.05 (*s*, 3 H); 2.70 (*s*, 3 H); 1.80–1.40 (*m*, 3 H); 1.25 (*d*, *J* = 7, 3 H); 0.95 (*m*, 6 H). FAB-MS: 350 (11, *M*Na$^+$), 328 (63, *M*H$^+$), 327 (14), 289 (12), 155 (32), 139 (17), 138 (39), 137 (75), 124 (11), 120 (14), 107 (26), 91 (14), 90 (17), 89 (23), 86 (17), 78 (11), 77 (22), 58 (13), 57 (12).

Cyclo(-Leu-MeAla-MeDAla-Gly-) (**14**). According to *GP I*, with **4** (200 mg, 0.613 mmol), DMPU (0.44 ml, 3.7 mmol), LDA (0.4M, 9.2 ml, 3.7 mmol), BuLi (1.0M, 2.0 ml, 2.0 mmol) and MeI (0.47 ml, 7.5 mmol). Anal. HPLC: 76% of **14**, 24% of **4**. Peptide **14** was obtained as a colourless powder (150 mg, 59%). HPLC (H_2O/MeCN 95:5 → 60:40 within 40 min): t_R 14.8 (**4**), 17.8 min (**14**). Hydrolysis of the peptide, formation of derivatives and GC analysis proved the absolute configuration of the new amino acid. ^{1}H-NMR (500 MHz, $CDCl_3$): 6.50 (*d*, *J* = 9.5, 1 H); 6.46 (*t*, *J* = 7.9, 1 H); 5.42 (*q*, *J* = 6.7, 1 H); 4.84 (*m*, 1 H); 4.55 (*q*, *J* = 10.9, 1 H); 3.87 (*dd*, *J* = 17.5, 8.8, 1 H); 3.63 (*dd*, *J* = 17.5, 7.1, 1 H); 3.11 (*s*, 3 H); 2.83 (*s*, 3 H); 1.72 (*m*, 1 H); 1.62–1.47 (*m*, 2 H); 1.52 (*d*, *J* = 7.1, 3 H); 1.24 (*d*, *J* = 6.7, 3 H); 0.96 (*d*, *J* = 6.4, 3 H); 0.93 (*d*, *J* = 6.5, 3 H); NOE: *ctct* conformation. ^{13}C-NMR (125 MHz, $CDCl_3$): 172.9 (*s*); 171.7 (*s*); 171.0 (*s*); 167.5 (*s*); 52.3 (*d*); 49.6 (*d*); 48.0 (*d*); 43.8 (*t*); 41.3 (*t*); 31.0 (*q*); 30.8 (*q*); 24.4 (*d*); 23.2 (*q*); 22.4 (*q*); 15.3 (*q*); 13.4 (*q*). FAB-MS: 363 (30, *M*Na$^+$), 341 (98, *M*H$^+$), 339 (17), 155 (18), 141 (10), 139 (11), 138 (22), 114 (15), 89 (14), 86 (30), 77 (14), 58 (100), 57 (13), 56 (18), 55 (14).

Cyclo(-Leu-Ape(4-en)-MeDAla-Gly-) (**15**). According to *GP I*, with 4 (100 mg, 0.307 mmol), DMPU (0.22 ml, 1.8 mmol), LDA (0.4M, 4.6 ml, 1.8 mmol), BuLi (1.35M, 0.75 ml, 1.0 mmol) and allyl bromide (0.31 ml, 3.8 mmol). Anal. HPLC: 51% of **15**, 49% of **4**. Peptide **15** was isolated as a colourless powder (46 mg, 41%). HPLC (H_2O/MeCN 95:5 → 60:40 within 40 min): t_R 14.9 (**4**), 25.8 min (**15**). ^{1}H-NMR (400 MHz, $CDCl_3$): 6.55–6.40 (*m*, 2 H); 5.70 (*m*, 1 H); 5.41 (*q*, *J* = 6.7, 1 H); 5.22 (*d*, *J* = 8.7, 1 H); 5.19 (*d*, *J* = 1.0, 1 H); 4.83 (*m*, 1 H); 4.45 (*dd*, *J* = 11.5, 2.7, 1 H); 3.87 (*dd*, *J* = 17.4, 8.9, 1 H); 3.65 (*dd*, *J* = 17.5, 7.0, 1 H); 3.13 (*s*, 3 H); 2.83 (*s*, 3 H); 2.71 (*m*, 1 H); 2.50 (*dd*, *J* = 14.8, 4.7, 1 H); 1.76 (*m*, 1 H); 1.57 (*m*, 1 H); 1.46 (*m*, 1 H); 1.24 (*d*, *J* = 6.7, 3 H); 0.95 (*d*, *J* = 6.5, 3 H); 0.92 (*d*, *J* = 6.6, 3 H); NOE: *ctct* conformation. ^{13}C-NMR (100 MHz, $CDCl_3$): 172.8 (s); 171.7 (*s*); 170.5 (*s*); 167.5 (*s*); 132.5 (*d*); 118.9 (*t*); 57.1 (*d*); 49.7 (*d*); 48.0 (*d*); 43.8 (*t*); 41.2 (*t*); 31.9 (*t*); 31.2 (*q*); 30.9 (*q*); 24.5 (*d*); 23.0 (*q*); 22.4 (*q*); 13.4 (*q*). FAB-MS: 389 (100, *M*Na$^+$), 367 (77, *M*H$^+$), 138 (13), 137 (16), 107 (13), 90 (11), 89 (18), 86 (30), 85 (10), 84 (77), 82 (11), 78 (19), 77 (21), 63 (11), 58 (33), 56 (16), 55 (18).

Cyclo(-Leu-Ahx(4-en)-MeDAla-Gly-) (**16**). According to *GP I*, with **4** (34 mg, 0.10 mmol), DMPU (0.08 ml, 0.7 mmol), LDA (0.4M, 1.55 ml, 0.6 mmol), BuLi (1.35M, 0.25 ml, 0.3 mmol) and 1-bromobut-2-ene (0.13 ml, 1.3 mmol). Anal. HPLC: 48% of **4**, 52% of **16**. Peptide **16** was isolated as a colourless powder (15 mg, 38%). HPLC (H_2O/MeCN 95:5 → 60:40 within 40 min): t_R 16.3 (**4**), 36.8 min (**16**). ^{1}H-NMR (200 MHz, $CDCl_3$): 6.70 (*d*, *J* = 10, 1 H); 6.60 (*t*, *J* = 8, 1 H); 5.40 (*q*, *J* = 8, 1 H); 5.00–4.70 (*m*, 2 H); 4.35 (*dd*, *J* = 12, 3, 1 H); 3.85 (*dd*, *J* = 18, 8, 1 H); 3.60 (*dd*, *J* = 18, 6, 1 H); 3.10 (*s*, 3 H); 2.80 (*s*, 3 H); 2.80–2.35 (*m*, 2 H); 1.85–1.30 (*m*, 3 H); 1.25 (*d*, *J* = 8, 3 H); 0.90 (*m*, 6 H). ^{13}C-NMR (125 MHz, $CDCl_3$): 173.0 (*s*); 171.8 (*s*); 170.7 (*s*); 167.6 (*s*); 129.7, 128.0 (2*d*); 125.0, 124.2 (2*d*); 57.6 (*d*); 49.7 (*d*); 48.0 (*d*); 43.8 (*t*); 41.1 (*t*); 31.2 (*q*); 30.9 (*t*); 30.9 (*t*); 24.5 (*d*); 22.9 (*q*); 22.5 (*q*); 18.0 (*q*); 13.4 (*q*). FAB-MS: 403 (15, *M*Na$^+$), 381 (100, *M*H$^+$), 379 (11), 307 (13), 155 (20), 154 (56), 138 (21), 137 (37), 136 (41), 107 (16), 99 (11), 98 (74), 97 (11), 91 (11), 90 (11), 89 (15), 86 (29), 77 (16), 69 (10), 58 (30), 57 (13), 56 (11), 55 (18).

Cyclo(-Leu-Ahx(5-Me,4-en)-MeDAla-Gly-) (**17**). According to *GP I*, with **4** (37 mg, 0.11 mmol), DMPU (0.08 ml, 0.7 mmol), LDA (0.4M, 1.70 ml, 0.7 mmol), BuLi (1.35M, 0.28 ml, 0.4 mmol) and 1-bromo-3-methylbut-2-ene (0.16 ml, 1.4 mmol). Anal. HPLC: 20% of **4**, 80% of **17**. Peptide **17** was obtained as a colourless powder (27 mg, 60%). HPLC (H_2O/MeCN 95:5 → 60:40 within 40 min): t_R 16.3 (**4**), 36.8 min (**17**). ^{1}H-NMR (200 MHz, $CDCl_3$): 6.70 (*d*, *J* = 10, 1 H); 6.60 (*t*, *J* = 8, 1 H); 5.40 (*q*, *J* = 8, 1 H); 5.00–4.70 (*m*, 2 H); 4.35 (*dd*, *J* = 12, 3, 1 H); 3.85 (*dd*, *J* = 18, 8, 1 H); 3.60 (*dd*, *J* = 18, 6, 1 H); 3.10 (*s*, 3 H); 2.80 (*s*, 3 H); 2.80–2.35 (*m*, 2 H); 1.85–1.30 (*m*, 3 H); 1.25 (*d*, *J* = 8 Hz, 3 H); 0.90 (*m*, 6 H); NOE: *ctct* conformation. ^{13}C-NMR (125 MHz, $CDCl_3$): 173.0 (*s*); 171.8 (*s*); 170.9 (*s*); 167.6 (*s*); 135.9 (*s*); 118.3 (*d*); 57.5 (*d*); 49.7 (*d*); 47.9 (*d*); 43.8 (*t*); 41.1 (*t*); 31.1 (*q*); 30.9 (*q*); 26.9 (*t*); 25.8 (*d*); 24.5 (*q*); 23.0 (*q*); 22.5 (*q*); 18.3 (*q*); 13.4 (*q*). FAB-MS: 417 (15, *M*Na$^+$), 395 (100, *M*H$^+$), 393 (12), 155 (11), 154 (25), 138 (12), 137 (17), 136 (20), 113 (13), 112 (75), 86 (27), 81 (12), 58 (26), 55 (11).

Cyclo(-Leu-MeSer((3R)- and (3S)-3-Et)-MeDAla-Gly-) (**18** and **19**, resp.). According to *GP I*, with **4** (26 mg, 0.080 mmol), DMPU (0.06 ml, 0.5 mmol), LDA (0.4M, 1.2 ml, 0.5 mmol), BuLi (1.1M, 0.24 ml, 0.3 mmol) and propionaldehyde (0.06 ml, 0.8 mmol). Anal. HPLC: 48% of **4**, 24% of **18**, 28% of **19**. Peptides **18** and **19** were obtained as white powders (4 mg of each, 13%).

18: HPLC (H_2O/MeCN 95:5 → 60:40 within 40 min): t_R 18.6 min. ^{1}H-NMR (500 MHz, $CDCl_3$): 6.49 (*d*, *J* = 10.5, 0.5 H); 6.19 (*m*, 1 H); 5.96 (*d*, *J* = 8.2, 0.5 H); 5.48, 5.13 (2*q*, *J* = 6.8, 1 H); 4.82 (*m*, 1 H); 4.50, 3.49 (2*d*,

J = 4.1, 1 H); 4.45, 3.84 (2*dd*, J = 12.7, 10.7 and J = 17.4, 9.1, resp., 1 H); 4.33 (*m*, 1 H); 3.60, 3.31 (2*dd*, J = 17.5, 7.0 and J = 15.8, 1.8, resp. 1 H); 3.29, 3.26 (2*s*, 3 H); 2.87, 2.71 (2*s*, 3 H); 1.78 (*m*, 1 H); 1.63 (*m*, 3 H); 1.43 (*m*, 1 H); 1.28, 1.26 (2*d*, J = 6.8, 3 H); 1.07–0.91 (*m*, 9 H). ^{13}C-NMR (125 MHz, $CDCl_3$): 177.0 (*s*); 173.7 (*s*); 172.9 (*s*); 172.1 (*s*); 171.6 (*s*); 170.1 (*s*); 169.5 (*s*); 167.4 (*s*); 72.8 (*d*); 72.4 (*d*); 66.7 (*d*); 61.1 (*d*); 53.3 (*d*); 50.7 (*d*); 50.0 (*d*); 48.0 (*d*); 45.5 (*t*); 43.8 (*t*); 41.2 (*t*); 39.5 (*t*); 39.3 (*q*); 34.3 (*q*); 31.2 (*q*); 30.3 (*q*); 27.9 (*t*); 27.6 (*t*); 24.9 (*d*); 24.5 (*d*); 23.2 (*q*); 22.9 (*q*); 22.4 (*q*); 21.7 (*q*); 13.6 (*q*); 12.2 (*q*); 10.7 (*q*); 10.6 (*q*). FAB-MS: 407 (73, MNa$^+$), 385 (100, MH$^+$), 383 (12), 367 (29), 327 (12), 307 (20), 289 (13), 192 (18), 176 (21), 156 (17), 155 (31), 152 (11), 143 (13), 139 (17), 138 (32), 137 (61), 124 (12), 120 (13), 115 (10), 114 (13), 113 (10), 107 (26), 102 (70), 91 (17), 90 (18), 89 (25), 86 (46), 85 (10), 84 (16), 79 (10), 78 (14), 77 (27), 69 (12), 67 (12), 65 (12), 64 (12), 63 (12), 58 (46), 57 (18), 56 (13), 55 (22), 51 (13), 44 (12), 43 (18), 42 (18), 39 (50).

19: HPLC (H_2O/MeCN 95:5→60:40 within 40 min): t_R 20.1 min. ^{1}H-NMR (500 MHz, $CDCl_3$): 6.33 (*d*, J = 9.4, 1 H); 6.00 (*m*, 1 H); 5.18 (*q*, J = 6.8, 1 H); 4.82 (*m*, 1 H); 4.50–4.39 (*m*, 2 H); 3.87 (br. *s*, 1 H); 3.32 (*d*, J = 8.5, 1 H); 3.27 (*s*, 3 H); 3.24 (*dd*, J = 13.0, 1.7, 1 H); 2.70 (*s*, 3 H); 1.75–1.62 (*m*, 1 H); 1.61–1.47 (*m*, 2 H); 1.46–1.40 (*m*, 1 H); 1.39–1.31 (*m*, 1 H); 1.26 (*d*, J = 6.8, 3 H); 1.06 (*t*, J = 7.3, 3 H); 0.97 (*m*, 6 H). ^{13}C-NMR (125 MHz, $CDCl_3$): 176.4 (*s*); 173.4 (*s*); 171.9 (*s*); 170.9 (*s*); 70.1 (*d*); 68.2 (*d*); 53.0 (*d*); 50.1 (*d*); 45.1 (*t*); 39.7 (*q*); 39.6 (*t*); 30.5 (*q*); 25.6 (*t*); 24.9 (*d*); 23.2 (*q*); 21.8 (*q*); 12.1 (*q*); 10.1 (*q*). FAB-MS: 407 (73, MNa$^+$), 385 (100, MH$^+$), 383 (12), 367 (29), 327 (12), 307 (20), 289 (13), 192 (18), 176 (21), 156 (17), 155 (31), 152 (11), 143 (13), 139 (17), 138 (32), 137 (61), 124 (12), 120 (13), 115 (10), 114 (13), 113 (10), 107 (26), 102 (70), 91 (17), 90 (18), 89 (25), 86 (46), 85 (10), 84 (16), 79 (10), 78 (14), 77 (27), 69 (12), 67 (12), 65 (12), 64 (12), 63 (12), 58 (46), 57 (18), 56 (13), 55 (22), 51 (13), 44 (12), 43 (18), 42 (18), 39 (50).

*Cyclo(-Leu-MeSer((3*R*)- and (3*S*)-3-Ph)-Me*D*Ala-Gly-)* (**20** and **21**, resp.). According to *GP I*, with **4** (100 mg, 0.307 mmol), DMPU (0.22 ml, 1.8 mmol), LDA (0.4M, 4.6 ml, 1.8 mmol), BuLi (1.35M, 0.75 ml, 1.0 mmol) and PhCHO (0.37 ml, 3.7 mmol). Anal. HPLC: 31% of **4**, 39% of **20** and 30% of **21**. Peptides **20** and **21** were obtained as white powders (41 mg (31%) of **20** and 32 mg (24%) of **21**). Crystals for X-ray analysis of **20** were obtained from EtOH.

20: HPLC (H_2O/MeCN 95:5→60:40 within 40 min): t_R 29.2 min. ^{1}H-NMR (500 MHz, $CDCl_3$): 7.47–7.30 (*m*, 5 arom. H); 6.91, 6.30 (*d* and *t*, J = 9.3 and J = 7.9, resp., NH, Gly); 6.52, 6.03 (2*d*, J = 9.8 and J = 9.3, resp., NH, Leu); 5.58, 5.52 (2*d*, J = 2.5 and J = 6.6, resp., H–C(β), Ser(3-Ph)); 5.37, 5.31 (2*q*, J = 6.8, H–C(α), D-Ala); 5.13 (br. *s*, OH); 4.82–4.72 (*m*, H–C(α), Leu); 4.41, 3.84 (2*dd*, J = 12.7, 10.5 and J = 17.3, 8.9, 1 H–C(α), Gly); 3.74, 3.70 (2*d*, J = 2.6, H–C(α), Ser(3-Ph)); 3.61, 3.29 (2*dd*, J = 17.3, 7.1 and J = 12.7, 2.2, 1 H–C(α), Gly); 3.23, 2.52 (2*s*, MeN, Ser(3-Ph)); 2.64, 2.34 (2*s*, MeN, D-Ala); 1.80 (*m*, H–C(γ), Leu); 1.69 (*m*, 1 H–C(β), Leu); 1.41 (*m*, 1 H–C(β), Leu); 1.28, 1.14 (2*d*, J = 6.8, Me(β), D-Ala); 1.00, 0.94 (2*d*, J = 6.6, Me(δ), Leu); 0.96, 0.91 (2*d*, J = 6.6, Me(δ), Leu); NOESY, ROESY, NOE: *tttt* conformation. ^{1}H-NMR (500 MHz, (D_6)Ethanol): 7.45 (*d*, J = 7.8, 2 arom. H); 7.31 (*t*, J = 10.0, 2 arom. H); 7.24 (*t*, J = 7.3, 1 arom. H); 5.52 (*d*, J = 5.8, H–C(β), Ser(3-Ph)); 5.32 (*q*, J = 4.5, H–C(α), D-Ala); 5.25 (*s*, OH); 4.90 (*d*, J = 6.3, H–C(α), Ser(3-Ph); 4.81 (*m*, H–C(α), Leu); 3.79 (*d*, J = 17.4, 1 H–C(α), Gly); 3.61 (*d*, J = 17.7, 1 H–C(α), Gly); 3.06 (*s*, MeN); 2.59 (*s*, MeN); 1.76–1.65 (*m*, H–C(γ), Leu); 1.56–1.50 (*m*, 1 H–C(β), Leu); 1.33–1.28 (*m*, 1 H–C(β), Leu); 1.16–1.11 (*m*, 2 Me(δ), Leu, EtOH); 0.92 (*d*, J = 7.0, Me(β), D-Ala). ^{13}C-NMR (125 MHz, $CDCl_3$; only major conformer): 177.5 (CO, D-Ala); 174.0 (CO, Leu); 172.0 (CO, Ser(3-Ph)); 170.0 (CO, Gly); 141.1 (arom.); 128.8 (arom.); 128.1 (arom.); 126.0 (arom.); 73.6 (C(β), Ser(3-Ph)); 69.6 (C(α), Ser(3-Ph)); 53.8 (C(α), Leu); 50.5 (C(α), D-Ala); 45.5 (C(α), Gly); 39.7 (C(β), Leu); 38.4 (MeN, Ser(3-Ph)); 30.5 (MeN, D-Ala); 24.8 (C(γ), Leu); 23.3 (C(δ), Leu); 21.8 (C(δ), Leu); 12.2 (C(β)- D-Ala). ^{13}C-NMR (125 MHz, [D_6]Ethanol); 174.5 (CO, D-Ala); 172.8 (CO, Leu); 170.6 (CO, Gly); 169.4 (CO, Ser(3-Ph)); 142.1 (arom.); 129.2 (arom.); 128.2 (arom.); 126.6 (arom.); 71.2 (C(β), Ser(3-Ph)); 64.3 (C(α), Ser(3-Ph)); 51.2 (C(α), Leu); 48.5 (C(α), D-Ala); 44.1 (C(α), Gly); 42.0 (C(β), Leu); 33.9 (MeN, Ser(3-Ph)); 31.3 (MeN, D-Ala); 25.4 (C(γ), Leu); 23.1, 23.0 (C(δ), Leu); 13.4 (C(β), D-Ala).

21: HPLC (H_2O/MeCN 95:5→60:40 within 40 min): t_R 36 min 20 s. ^{1}H-NMR (500 MHz, $CDCl_3$): 7.35 (*m*, 5 H); 6.42 (*d*, J = 10.4, 1 H); 6.05 (*d*, J = 10.3, 1 H); 5.53 (*d*, J = 8.9, 1 H); 5.24 (*q*, J = 6.8, 1 H); 4.67 (*m*, 1 H); 4.47 (*dd*, J = 12.8, 10.7, 1 H); 4.17 (br. *s*, 1 H); 3.44 (*d*, J = 8.8, 1 H); 3.30 (*dd*, J = 12.8, 1.8, 1 H); 2.65 (*s*, 3 H); 2.36 (*s*, 3 H); 1.70 (*m*, 1 H); 1.58 (*m*, 1 H); 1.30 (*d*, J = 6.8, 3 H); 1.23 (*m*, 1 H); 0.95 (*d*, J = 6.7, 3 H); 0.91 (*d*, J = 6.6, 3 H). ^{13}C-NMR (125 MHz, $CDCl_3$): 176.3 (*s*); 173.8 (*s*); 171.6 (*s*); 171.0 (*s*); 139.4 (*s*); 128.8 (*d*); 128.5 (*d*); 127.0 (*d*); 71.1 (*d*); 69.7 (*d*); 53.3 (*d*); 50.5 (*d*); 45.6 (*t*); 39.1 (*t*); 37.8 (*q*); 30.3 (*q*); 24.7 (*d*); 23.2 (*q*); 21.6 (*q*); 12.1 (*q*).

*Cyclo(-Leu-Sar((*R*)-2-CO_2Me)-Me*D*Ala-Gly-)* (**22**). According to *GP I*, with **4** (100 mg, 0.307 mmol), DMPU (0.22 ml, 1.8 mmol), LDA (0.4M, 4.6 ml, 1.8 mmol) and BuLi (1.0M, 1.0 ml, 1.0 mmol). CO_2 was bubbled through the mixture for 1 h. AcOH and then CH_2N_2 were added until the soln. kept its yellow colour for more than 10 s. The mixture was allowed to warm up. Anal. HPLC: 97% of **22**, 3% of **4**. Peptide **22** was obtained as a colourless powder (100 mg, 85%). HPLC (H_2O/MeCN 95:5→60:40 within 40 min): t_R 16.2 (**4**), 12.3 min (**22**).

^{1}H-NMR ($CDCl_3$, 500 MHz): 8.64 (*d*, *J* = 6, 0.5 H, NH); 7.62 (*d*, *J* = 9, 0.5 H, NH); 6.17 (br. *s*, 0.5 H, H–C(α), D-Ala); 6.14 (*s*, 0.5 H, H–C(α), Sar(2-CO_2Me)); 5.40 (*q*, *J* = 7, 0.5 H, H–C(α), D-Ala); 5.08 (*s*, 0.5 H, H–C(α), D-Ala); 4.86 (*m*, 0.5 H, H–C(α), Leu); 4.69 (*dd*, *J* = 7, 10, 0.5 H, H–C(α), Gly); 4.15 (*m*, 1 H, 2 × 0.5 H–C(α), Leu and Gly); 3.86, 3.81 (2*s*, 3 H, MeO); 3.64 (*dd*, *J* = 17.5, 6.9, 0.5 H, H–C(α), Gly); 3.28 (*d*, *J* = 14, 0.5 H, H–C(α), Gly); 3.12, 3.02, 2.99, 2.67 (4*s*, 6 H, 2 MeN); 2.00–1.40 (*m*, *ca.* 4 H, $CH_2(\beta)$, H–C(γ), Leu, H_2O); 1.52, 1.30 (2*d*, *J* = 3 and *J* = 7, resp., 3 H, Me(β), D-Ala); 1.00–0.90 (*m*, 6 H, 2 Me(δ), Leu). ^{13}C-NMR ($CDCl_3$, 125 MHz): 173.6 (*s*); 172.6 (*s*); 172.4 (*s*); 170.9 (*s*); 170.0 (*s*); 167.7 (*s*); 167.4 (*s*); 166.3 (*s*); 166.0 (*s*); 165.8 (*s*); 62.4 (*d*); 58.5 (*d*); 55.1 (*d*); 53.6 (*q*); 52.7 (*q*); 51.9 (*d*); 49.7 (*t*); 47.7 (*t*); 43.7 (*t*); 42.6 (*t*); 41.1 (*t*); 40.1 (*t*); 33.2 (*q*); 32.5 (*q*); 30.2 (*q*); 30.0 (*q*); 24.6 (*d*); 24.3 (*d*); 23.3 (*q*); 23.2 (*q*); 22.1 (*q*); 20.7 (*q*); 17.2 (*q*); 13.1 (*q*). FAB-MS: 407 (44, *M*Na$^+$), 385 (100, *M*H$^+$), 383 (12), 307 (16), 289 (11), 272 (24), 215 (19), 176 (22), 164 (16), 155 (27), 154 (68), 152 (10), 139 (14), 138 (29), 137 (53), 136 (56), 120 (12), 114 (11), 107 (21), 102 (28), 91 (14), 90 (15), 89 (21), 86 (39), 78 (11), 77 (21), 65 (10), 58 (39), 57 (18), 56 (16), 55 (18).

Cyclo(-Leu-Sar-MeD Ala-AcGly-) (**23**). According to *GP I*, with **4** (27 mg, 0.083 mmol), DMPU (0.06 ml, 0.5 mmol), 0.4M LDA (1.25 ml, 0.5 mmol), BuLi (1.35M, 0.21 ml, 0.3 mmol) and AcCl (80 mg, 0.7 ml, 1.0 mmol). Anal. HPLC: 3 unidentified peaks. The isolation of the main one afforded **23** as a white powder (4 mg, 13%). HPLC (H_2O/MeCN 95:5→60:40 within 40 min): t_R 16.2 (**4**), 25.7 min (**23**). ^{1}H-NMR (200 MHz, $CDCl_3$): 5.95 (*d*, *J* = 8.5, 1 H); 5.55 (*q*, *J* = 8.4, 1 H); 4.95 (*d*, *J* = 18.0, 1 H); 4.75 (*m*, 1 H); 4.40 (*d*, *J* = 18.1, 1 H); 3.75 (*d*, *J* = 17.5, 1 H); 3.55 (*d*, *J* = 17.5, 1 H); 3.05 (*s*, 3 H); 2.70 (*s*, 3 H); 2.55 (*s*, 3 H); 1.90–1.35 (*m*, 3 H); 1.35 (*d*, *J* = 7.3, 3 H); 0.90 (*d*, *J* = 8.2, 6 H); NOE: *ctct* conformation. ^{13}C-NMR (100 MHz, $CDCl_3$): 173.3 (*s*); 173.1 (*s*); 170.6 (*s*); 165.5 (*s*); 165.1 (*s*); 52.1 (*d*); 50.8 (*t*); 48.2 (*d*); 44.5 (*t*); 40.9 (*t*); 36.7 (*q*); 29.2 (*q*); 26.8 (*q*); 24.3 (*d*); 23.1 (*q*); 22.3 (*q*); 14.1 (*q*). FAB-MS: 369 (100, *M*H$^+$), 329 (16), 313 (11).

Cyclo(-Leu-{(1S)-2-[(3,3-dimethyloxiran-2-yl)methyl]sarcosyl}-MeD Ala-Gly-) (**24**). *m*-CPBA (*ca.* 17 mg, 0.07 mmol) was added to a soln. of **17** (25 mg, 0.064 mmol) in CH_2Cl_2. After 1 h, Na_2SO_3 (*ca.* 50 mg) was added and the mixture filtered through acidic and basic *Dowex*. Anal. HPLC: two stereoisomers 96:4; the major one, **24**, was isolated by prep. HPLC as a white powder (12 mg, 45%). HPLC (H_2O/MeCN 95:5→60:40 within 40 min): t_R 25.5 min (**24**). ^{1}H-NMR ($CDCl_3$, 400 MHz): 6.39 (br. *s*, 1 H); 6.28 (br. *s*, 1 H); 5.40 (*q*, *J* = 9, 1 H); 4.94 (*m*, 1 H); 4.64 (*dd*, *J* = 15, 3, 1 H); 3.86 (*dd*, *J* = 22, 8, 1 H); 3.63 (*dd*, *J* = 22, 7, 1 H); 3.19 (*s*, 3 H); 2.78 (*s*, 3 H); 2.67 (*dd*, *J* = 9, 3, 1 H); 2.36 (*ddd*, *J* = 14, 14, 3, 1 H); 1.8–1.4 (*m*, 4 H); 1.34 (*s*, 3 H); 1.33 (*s*, 3 H); 1.23 (*d*, *J* = 5, 3 H); 0.97 (*d*, *J* = 6, 3 H); 0.93 (*d*, *J* = 6, 3 H). ^{13}C-NMR ($CDCl_3$, 125 MHz): 172.7 (*s*); 172.2 (*s*); 170.5 (*s*); 167.4 (*s*); 60.3 (*d*); 59.9 (*d*); 55.1 (*d*); 49.6 (*d*); 48.1 (*d*); 43.7 (*t*); 41.2 (*t*); 31.7 (*q*); 30.9 (*q*); 27.4 (*t*); 24.6 (*d*); 24.5 (*q*); 23.0 (*q*); 22.4 (*q*); 19.3 (*q*); 13.5 (*q*). FAB-MS: 433 (92, *M*Na$^+$), 411 (93, *M*H$^+$), 409 (16), 399 (10), 155 (10), 154 (13), 143 (16), 141 (11), 136 (12), 129 (12), 128 (98), 127 (12), 114 (18), 86 (73), 58 (100).

6. *Benzylations Using the P4-Base. GP II.* The cyclic peptide was dissolved or suspended in THF (5 ml) and cooled to −78°. $PhCH_2Br$ was added and the mixture cooled to −100°. P4-Base, which had been previously dissolved in THF (5 ml) under Ar, was added dropwise such that the temp. did not exceed −90°. After stirring for 3 h at −78°, the mixture was allowed to warm to r.t. and Et_2O (10 ml) was added. Most of the P4-base salt was removed by filtration and the resulting soln. was evaporated. The alkylated products were isolated by FC and the residual salt of the P4-base could be removed from the column by elution with MeOH with subsequent regeneration allowing the P4-base to be used in further reactions.

Cyclo(-MeLeu-PhCH$_2$Gly-PhCH$_2$D Ala-Sar-) (**29**) *and cyclo(-MeLeu-PhCH$_2$Gly-PhCH$_2$D Ala-MeD Phe-)* (**30**). According to *GP II* with cyclo(-MeLeu-Gly-D-Ala-Sar-) (**3**; 98 mg, 0.30 mmol), $PhCH_2Br$ (0.30 ml, 2.5 mmol) and P4-base (0.91 mmol). FC (Et_2O/MeOH 95:5) yielded **29** (80 mg, 52%) and **30** (35 mg, 20%) as white powders.

29: M.p. 248°. $[\alpha]_D^{r.t.} = +39.6$ (*c* = 0.72, $CHCl_3$). ^{1}H-NMR ($CDCl_3$, 300 MHz): 7.28–6.94 (*m*, 10 H); 5.66 (*q*, *J* = 6.6, 1 H); 5.50 (*d*, *J* = 14.5, 1 H); 5.27 (*m*, 1 H); 4.55 (*d*, *J* = 18, 1 H); 4.35 (*dd*, *J* = 27, 8.5, 2 H); 4.14 (*d*, *J* = 18, 1 H); 3.64 (*d*, *J* = 18, 1 H); 3.60 (*d*, *J* = 14.5, 1 H); 3.12 (*s*, 3 H); 3.08 (*d*, *J* = 18.5, 1 H); 2.76 (*s*, 3 H); 1.68 (*m*, 1 H); 1.35–1.50 (*m*, 2 H); 1.35 (*d*, *J* = 6.6, 3 H); 0.96 (*d*, *J* = 6.6, 3 H); 0.94 (*d*, *J* = 6.6, 3 H). ^{13}C-NMR ($CDCl_3$, 75 MHz): 170.8; 169.9; 169.0; 167.5; 137.1; 136.2; 129.1; 128.9; 128.5; 127.6; 127.3; 125.7; 52.1; 50.8; 50.3; 49.6; 47.0; 46.3; 37.4; 36.8; 29.0; 24.6; 23.3; 22.5; 15.3. FAB-MS: 507 (81, *M*H$^+$), 309 (8), 199 (22), 197 (10), 155 (13), 154 (41), 137 (26), 136 (35), 134 (27), 127 (18), 106 (18), 100 (39), 91 (100).

30: $[\alpha]_D^{r.t.} = -53.3$ (*c* = 0.8, $CHCl_3$). ^{1}H-NMR ($CDCl_3$, 300 MHz): 7.28–6.94 (*m*, 15 H); 5.66 (*q*, *J* = 6.6, 1 H); 5.46 (*d*, *J* = 14, 1 H); 5.35 (*dd*, *J* = 9, 6, 1 H); 4.88 (*dd*, *J* = 10.5, 4, 1 H); 4.25 (*s*, 2 H); 3.57 (*d*, *J* = 14, 1 H); 3.14 (*s*, 3 H); 3.13 (*dd*, *J* = 25, 11, 2 H); 3.08 (*d*, *J* = 18, 1 H); 2.95 (*s*, 3 H); 1.85–1.55 (*m*, 2 H); 1.33 (*d*, *J* = 6.5, 3 H); 1.52–1.21 (*m*, 1 H); 0.99 (*d*, *J* = 6.6, 3 H); 0.97 (*d*, *J* = 6.6, 3 H). ^{13}C-NMR ($CDCl_3$, 75 MHz): 172.2; 170.2; 169.7; 169.1; 137.2; 136.2; 129.2; 129.1; 129.0; 128.5; 127.7; 127.6; 127.4; 125.7; 58.1; 53.0; 50.6; 49.9; 47.2; 46.2; 37.8; 33.3; 32.2; 31.4; 24.1; 23.4; 22.6; 15.0. FAB-MS: 597 (65, *M*H$^+$), 406 (6), 217 (17), 203 (12), 190 (12), 162 (13), 156 (18), 154 (33), 138 (13), 137 (21), 136 (30), 134 (61), 120 (20), 106 (16), 100 (47), 91 (100).

Cyclo(-PhCH$_2$Val-Sar-Sar-PhCH$_2$Gly-) (**27**). Initially according to *GP II* with cyclo(-Val-Sar-Sar-Gly-) (**2**; 90 mg, 0.30 mmol), $PhCH_2Br$ (0.30 ml, 2.6 mmol) and P4-base (0.67 mmol). Two h after addition of the P4-base, the reaction was quenched with NH_4BF_4 soln. the mixture extracted, and the soln. evaporated. FC (Et_2O/MeOH 95:5) yielded **27** (39 mg, 27%). White powder. $[\alpha]_D^{r.t.} = +59.0$ ($c = 1.8$, $CHCl_3$). ^{1}H-NMR ($CDCl_3$, 400 MHz): 7.25–6.93 (*m*, 10 H); 5.40 (*d*, $J = 14.5$, 1 H); 5.32 (*d*, $J = 14.5$, 1 H); 4.59 (*d*, $J = 10$, 1 H); 4.42 (*d*, $J = 17$, 1 H); 4.22 (*d*, $J = 17$, 1 H); 4.17 (*d*, $J = 18$, 1 H); 3.71 (*d*, $J = 14.5$, 1 H); 3.70 (*d*, $J = 18$, 1 H); 3.34 (*d*, $J = 18$, 1 H); 3.30 (*d*, $J = 14.2$, 1 H); 3.12 (*s*, 3 H); 2.99 (*s*, 3 H); 2.10 (*m*, 1 H); 0.96 (*d*, $J = 7$, 3 H); 0.93 (*d*, $J = 7$, 3 H). ^{13}C-NMR ($CDCl_3$, 100 MHz): 170.2; 169.6; 168.9; 167.2; 136.6; 135.9; 129.1; 128.7; 128.6; 127.6; 127.5; 127.4; 60.7; 50.9; 50.8; 50.4; 48.6; 47.4; 36.6; 33.5; 27.8; 20.1; 19.5. FAB-MS: 479 (37, MH$^+$), 391 (81), 371 (21), 279 (8), 167 (23), 154 (22), 149 (100), 137 (27), 129 (34), 113 (21), 91 (25), 67 (27), 57 (68), 55 (43).

Cyclo(-PhCH$_2$Val-MePhe-Sar-PhCH$_2$Gly-) (**28**). According to *GP II*, with cyclo(-Val-Sar-Sar-Gly-) (84 mg, 0.28 mmol), $PhCH_2Br$ (0.27 ml, 2.3 mmol) and P4-base (1.1 mmol). FC (Et_2O/MeOH 95:5) yielded **28** (128 mg, 80%). Colourless oil. $[\alpha]_D^{r.t.} = +47.3$ ($c = 1.0$, $CHCl_3$). ^{1}H-NMR ($CDCl_3$, 400 MHz): 7.39–6.97 (*m*, 15 H); 5.40 (*d*, $J = 14.5$, 1 H); 5.31 (*d*, $J = 14.5$, 1 H); 5.00 (*d*, $J = 10.5$, 1 H); 4.98 (*t*, $J = 7.8$, 1 H); 4.33 (*d*, $J = 17.1$, 1 H); 4.08 (*d*, $J = 18$, 1 H); 4.06 (*d*, $J = 17$, 1 H); 3.68 (*d*, $J = 14.5$, 1 H); 3.37 (*d*, $J = 14.5$, 1 H), 3.35 (*d*, $J = 18.0$, 1 H); 3.20 (*d*, $J = 7.5$, 2 H); 3.13 (*s*, 3 H); 3.11 (*s*, 3 H); 2.12 (*m*, 1 H); 0.95 (*d*, $J = 6.6$, 3 H); 0.82 (*d*, $J = 6.6$, 3 H). ^{13}C-NMR ($CDCl_3$, 100 MHz): 170.6; 170.3, 169.4; 169.2; 136.5; 136.2; 135.8; 129.1; 129.1; 129.0; 128.8; 128.6; 128.4; 127.8; 127.7; 127.6; 127.5; 127.4; 61.2; 57.6; 50.9; 50.5; 48.7; 47.3; 35.0; 33.9; 31.6; 27.8; 19.9; 19.5. FAB-MS: 591 (4, MNa$^+$), 569 (47, MH$^+$), 162 (21), 134 (28), 91 (100), 55 (25).

Cyclo(-PhCH$_2$Leu-Sar-Sar-PhCH$_2$Gly-) (**25**) *and Cyclo(-PhCH$_2$Leu-MePhe-Sar-PhCH$_2$Gly-)* (**26**). According to *GP II*, with cyclo(-Leu-Sar-Sar-Gly-) (**1**; 100 mg, 0.27 mmol), $PhCH_2Br$ (0.30 ml, 2.0 mmol) and P4-base (1.0 mmol). FC (Et_2O/MeOH 95:5) yielded **25** (70 mg, 52%) as a white powder and **26** (37 mg, 23%) as a colourless oil.

25: M.p. 207°. $[\alpha]_D^{r.t.} = +18.1$ ($c = 0.98$, MeOH). ^{1}H-NMR ($CDCl_3$, 300 MHz): 7.27–6.95 (*m*, 10 H); 5.52 (*t*, $J = 7.0$, 1 H); 5.43 (*d*, $J = 14.5$, 1 H); 5.27 (*d*, $J = 14.5$, 1 H); 4.59 (*d*, $J = 18$, 1 H); 4.06 (*dd*, $J = 18$, 2 H); 3.71 (*d*, $J = 18$, 1 H); 3.60 (*d*, $J = 14.5$, 1 H); 3.30 (*d*, $J = 14.5$, 1 H); 3.14 (*d*, $J = 18.5$, 1 H); 3.11 (*s*, 3 H); 2.99 (*s*, 3 H); 1.88–1.76 (*m*, 1 H); 1.71–1.60 (*m*, 1 H); 1.48–1.40 (*m*, 1 H); 1.00 (*d*, $J = 6.6$, 3 H); 0.91 (*d*, $J = 6.6$, 3 H). ^{13}C-NMR (CDCl, 75 MHz): 170.3; 169.3; 168.8; 167.1; 137.1; 135.9; 129.1; 128.9; 128.5; 127.6; 127.4; 125.9; 52.4; 50.9; 50.8; 50.3; 48.0; 46.8; 38.4; 36.7; 33.6; 24.8; 23.0; 22.5. FAB-MS: 493 (100, MH$^+$), 307 (10), 176 (22), 155 (14), 154 (41), 138 (18), 137 (30), 136 (33), 120 (14), 107 (13), 91 (71), 89 (13), 77 (14).

26: $[\alpha]_D^{r.t.} = +40.4$ ($c = 2.3$, MeOH). ^{1}H-NMR ($CDCl_3$, 300 MHz): 7.42–6.87 (*m*, 15 H); 5.53 (*m*, 1 H); 5.43 (*d*, $J = 14.5$, 1 H); 5.36 (*d*, $J = 14.5$, 1 H); 4.98 (*m*, 1 H); 4.21 (*d*, $J = 18$, 1 H); 4.06 (*d*, $J = 18$); 3.95 (*dd*, $J = 19$, 1 H); 3.52 (*d*, $J = 14.5$, 1 H); 3.35 (*d*, $J = 14.5$, 1 H); 3.17 (*m*, 2 H); 3.17 (*d*, $J = 19$, 1 H); 3.16 (*s*, 3 H); 3.12 (*s*, 3 H); 1.91 (*m*, 1 H); 1.56 (*m*, 1 H); 1.39–1.18 (*m*, 1 H); 0.93 (*d*, $J = 6.6$, 3 H); 0.86 (*d*, $J = 6.6$, 3 H). ^{13}C-NMR ($CDCl_3$, 75 MHz): 171.2; 169.4; 169.3; 169.1; 137.1; 136.3; 135.8; 129.2; 129.0; 128.5; 127.6; 127.4; 125.9; 58.1; 52.4; 50.9; 50.4; 48.0; 46.6; 38.4; 35.1; 33.9; 31.8; 24.9; 23.2; 21.8. FAB-MS: 583 (100, MH$^+$), 258 (6), 189 (13), 176 (21), 154 (26), 138 (10), 137 (17), 136 (21), 134 (28), 120 (12), 105 (9), 91 (92).

7. *X-Ray Structure Analyses.* 7.1. *Cyclo(-Leu-Ser-((3R)-3-Ph)-Me*D*Ala-Gly-)* (**20**; $C_{22}H_{32}O_5N_4 \cdot C_2H_6O$). Determination of the cell parameters and collection of the reflection intensities were performed on an *Enraf-Nonius-CAD4* four-circle diffractometer (graphite monochromatised MoK_α radiation, $\lambda = 0.7107$ Å). Colourless crystal, 0.4 × 0.4 × 0.5 mm, orthorhombic, space group $P2_12_12_1$, $a = 9.354(3)$ Å, $b = 15.369(10)$ Å, $c = 18.304(6)$ Å, $V = 2631$ Å^3, $Z = 4$, $\rho_{calc.} = 1.208$ gcm^{-3}, $\mu = 0.087$ mm^{-1}, $F(000) = 1032$. Number of unique reflections 1701 (ω scan, $4 < 2\theta < 50°$, $T = 293$ K), of which 1701 with $I > 3\sigma(I)$ were used for the determination (direct methods, SHELXS-86 [49]). SHELXL 93 [50] was used for structure refinement (full-matrix least-squares). The non-H-atoms were refined anisotropically, the H-atoms were added to the molecule with constant isotropic temperature factors on idealised positions and refined according to the riding model (afix 3). The refinement converged at $R = 0.096$ ($wR^2 = 0.264$), min. and max. rest electron density −0.57, 0.77 eÅ^{-3}, number of variables 307.

7.2. *Cyclo(-PhCH$_2$Leu-Sar-Sar-PhCH$_2$Gly-)* (**25**; $C_{28}H_{36}O_4N_4$). Determination of the cell parameters and collection of the reflection intensities were performed on an *Enraf-Nonius-CAD4* four-circle diffractometer (graphite monochromatised MoK_α radiation, $\lambda = 0.7107$ Å). Colourless crystal, orthorhombic, space group $P2_12_12_1$, $a = 10.029(1)$ Å, $b = 13.259(2)$ Å, $c = 20.493(3)$ Å, $V = 2725.0(6)$ Å^3, $Z = 4$, $\rho_{calc.} = 1.201$ gcm^{-3}, $\mu = 0.081$ mm^{-1}, $F(000) = 1056$. Number of unique reflections 1990 (ω scan, $4 < 2\theta < 56°$, $T = 293$ K), of which 1990 with $I > 2\sigma(I)$ were used for the determination (direct methods, SHELXS-86 [49]). SHELXL 93 [50] was used for structure refinement (full-matrix least-squares). The non-H-atoms were refined anisotropically, the H-atoms were added to the molecule with constant isotropic temperature factors on idealised positions and refined according to the riding model (afix 3). The refinement converged at $R = 0.038$ ($wR^2 = 0.117$), min. and max. rest electron density −0.16, 0.16 eÅ^{-3}, number of variables 458.

REFERENCES

[1] H. Kawagishi, A. Somoto, J. Kuranari, A. Kimura, S. Chiba, *Tetrahedron Lett.* **1993,** *34,* 3439.
[2] M. Kijima, M. Yoshida, K. Sugita, S. Horinouchi, T. Beppu, *J. Biol. Chem.* **1993,** *268,* 22429.
[3] S.A. Miller, S.L. Griffiths, D. Seebach, *Helv. Chim. Acta* **1993,** *76,* 563.
[4] R.E. Shute, M. Kawai, D.A. Rich, *Tetrahedron* **1988,** *44,* 685.
[5] U. Schmidt, *Pure Appl. Chem.* **1986,** *58,* 295.
[6] U. Schmidt, *Nachr. Chem. Tech. Lab.* **1989,** *37,* 1034.
[7] M. Rothe, K.-D. Steffen, I. Rothe, *Angew. Chem.* **1965,** *77,* 347; *ibid. Int. Ed.* **1965,** *4,* 356.
[8] R.A. Houghten, *Gene* **1993,** *137,* 7.
[9] D. Seebach, A.K. Beck, A. Studer, in 'Modern Synthetic Methods 1995', Eds. B. Ernst and C. Leumann, VCHA (Basel) and VCH (Weinheim), 1995, Vol. 7, p. 1.
[10] T. Pietzonka, D. Seebach, *Angew. Chem.* **1992,** *104,* 1543; *ibid. Int. Ed.* **1992,** *31,* 1481.
[11] U. Schmidt, A. Lieberknecht, H. Gressner, J. Talbiersky, *J. Org. Chem.* **1982,** *47,* 3261.
[12] L. Sheh, M. Mokotoff, *Tetrahedron Lett.* **1985,** *26,* 5755.
[13] H.G. Bossler, P. Waldmeier, D. Seebach, *Angew. Chem.* **1994,** *106,* 455; *ibid. Int. Ed.* **1994,** *33,* 439.
[14] U. Schöllkopf, T. Tiller, Y. Bardenhagen, *Tetrahedron* **1988,** *44,* 5293.
[15] P. Grieco, Y. Yokoyama, S. Gilman, M. Nishizawa, *J. Org. Chem.* **1977,** *42,* 2034.
[16] H. Bossler, D. Seebach, *Helv. Chim. Acta* **1994,** *77,* 1124.
[17] R. Schwesinger, H. Schlemper, *Angew. Chem.* **1987,** *99,* 1212.
[18] R. Schwesinger, H. Schlemper, *Nachr. Chem. Tech. Lab.* **1990,** *38,* 1214; R. Schwesinger, J. Willaredt, H. Schlemper, M. Keller, D. Schmidt, H. Fritz, *Chem. Ber.* **1994,** *127,* 2435.
[19] F. Weygand, W. Steglich, J. Bjarnason, R. Akhtar, N. Khan, *Tetrahedron Lett.* **1984,** *25,* 4709.
[20] H.-M. Müller, D. Seebach, *Angew. Chem.* **1993,** *105,* 483; *ibid. Int. Ed.* **1993,** *32,* 477.
[21] D.A. Plattner, A. Brunner, M. Dobler, H.-M. Müller, W. Petter, P. Zbinden, D. Seebach, *Helv. Chim. Acta* **1993,** *76,* 2004.
[22] D. Seebach, T. Hoffmann, F.N.M. Kühnle, U.D. Lengweiler, *Helv. Chim. Acta* **1994,** *77,* 2007.
[23] H. Kessler, *Angew. Chem.* **1982,** *94,* 509; *ibid. Int. Ed.* **1982,** *21,* 512.
[24] J. McEwen, K. Ottoson, *Biopolymers* **1993,** *33,* 1377.
[25] W. Maestle, U. Link, W. Witschel, U. Thewald, T. Weber, M. Rothe, *Biopolymers* **1991,** *31,* 735.
[26] D.H. Rich, R.D. Jasensky, *J. Am. Chem. Soc.* **1980,** *102,* 1112.
[27] Y. Terada, M. Kawai, D.H. Rich, *Int. J. Pept. Protein Res.* **1989,** *33,* 3.
[28] T. Kato, A. Tone, Y. Kodera, S. Lee, Y. Shimohigashi, N. Izumiya, *Chem. Lett.* **1985,** 1209.
[29] C. Chiang-Chan, I.L. Karle, *Int. J. Pept. Protein Res.* **1982,** *20,* 133.
[30] D.N.J. White, C. Morrow, *Computers and Chemistry* **1979,** *3,* 33.
[31] D.H. Rich, P. Bhatnagar, in 'Peptides (Chemistry, Structure, Biology)', Eds. M. Goodman and J. Meienhofer, Wiley & Sons (N.Y.), 1977, p. 340.
[32] K. Titlestad, *Acta Chem. Scand., Ser. B* **1977,** *31,* 641.
[33] D.H. Rich, P.K. Bhatnagar, *J. Am. Chem. Soc.* **1978,** *100,* 2218.
[34] S. Gupta, G. Peiser, T. Nakajima, Y.-S. Hwang, *Tetrahedron Lett.* **1994,** *35,* 6009.
[35] B. Di Blasio, A. Lombardi, G. D'Auria, M. Saviano, C. Isernia, O. Maglio, L. Pollino, C. Pedone, V. Pavone, *Biopolymers* **1993,** *33,* 621.
[36] Y. Fusaoka, E. Ozeki, S. Kimaru, Y. Imanishi, *Int. J. Pept. Protein Res.* **1989,** *34,* 104.
[37] D.H. Rich, M. Kawai, R.D. Jasensky, *Int. J. Pept. Protein Res.* **1983,** *21,* 35.
[38] K. Ueno, T. Shimizu, *Biopolymers* **1983,** *22,* 633.
[39] T. Shimizu, Y. Tanaka, K. Tsuda, *Biopolymers* **1983,** *22,* 617.
[40] S. Rayudu, Ph.D. Thesis, The University of Arkansas, 1982.
[41] Z. Karimov, A.M. Mikhailov, G.N. Tishchenko, *Bioorgan. Khim.* **1976,** *2,* 1033.
[42] 'SYBYL', *TRIPOS Association Inc.*, St. Louis, Missouri, USA, 1991.
[43] J. Gasteiger, M. Marsili, *Tetrahedron* **1980,** *36,* 3219.
[44] M.R. Winkle, J.M. Lansinger, R.C. Ronald, *J. Chem. Soc., Chem. Commun.* **1980,** 87.
[45] T.J. Boer, H.J. Baker, *Org. Synth.* **1963,** *4,* 250.
[46] E.V. Arx, M. Faupel, M. Brugger, *J. Chromatogr.* **1976,** *120,* 224.
[47] S. Seebach, H. Bossler, H. Gründler, S.-i. Shoda, R. Wenger, *Helv. Chim. Acta* **1991,** *74,* 197.
[48] S. Sakakibara, N. Inukai, *Bull. Chem. Soc. Jpn.* **1965,** *38,* 1979; E.R. Fallardeau, D.D. Desmarteu, *J. Fluorine Chem.* **1976,** *7,* 409.
[49] SHELX-86, G.M. Sheldrick, Program for Crystal Structure Solution, University of Göttingen, 1986.
[50] 'SHELXL-93', G.M. Sheldrick, *Curr. Cont. Phys. Chem. Earth Sci.* **1989,** *29,* 14.