CHIMIA 39 (1985) Nr.4 (April)

Uncommon Amino Acids in the Strategy of Natural Product Synthesis**

Henricus C. J. Ottenheijm*

The chemistry of amino acids occuring in proteins is well established. Much less is known, however, about amino acids that occur in lower organisms and that have a functionality in addition to the amino and carboxy group. A pathway linking protein amino acids and uncommon, non-protein amino acids deserves attention as an outline of a biological relationship between these amino acids as well as a «chemosynthetic chart». This is illustrated by a biogenetic approach to gliotoxin and sporidesmin B; our approach features the transposition of a N-hydroxy group to a α-functionality in amino acids. Some novel reactions with a cycloadduct from indole derivatives and nitroso olefins open a new approach to indole alkaloids, e.g. tryptathionine, both in terms of strategy and methodology. Finally, one of our syntheses of sparsomycin is outlined. This bacterial metabolite contains a monooxodithioacetal moiety which can be considered as a derivative of D-*cysteine. Consequently, our approach features nucleophilic ring-opening of the sultine prepared from* D-*cysteine. Structure-activity relationship studies of sparsomycin have resulted in the synthesis of two analogues that have a higher antitumor activity and a lower toxicity than sparsomycin itself.*

1. Introduction

When the organic chemist uses the term «natural products», he usually means secondary metabolites. These *secondary metabolites* are distinct from primary metabolites in that they are frequently of relatively complex structure; in addition their distribution is restricted and more characteristic of specific sources. Whereas there are no sharp lines delimiting either class of compounds, *primary metabolites* are nearly universal in their distribution; they are the products of, and participants in, the cellular activities of nearly all living organisms, from microorganisms to man.

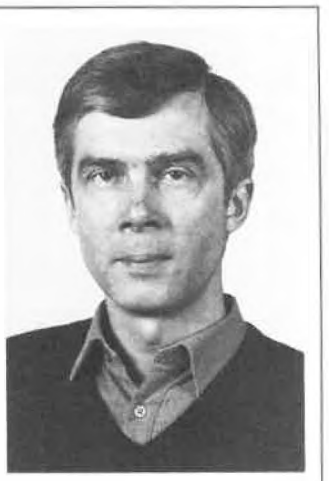
Amino acids fit very well into this classification[1a]. The common amino acids – in their L-configuration – fall into the category of primary metabolites. Their occurrence – e.g. as constituents of proteins – is widespread in all living cells; their chemistry is well established and their biochemical importance is fully understood. Much less is known, however, about the chemical and biochemical behaviour of the variety of α-amino acids featuring structures, which are uncommon to protein amino acids; most of them can be accomodated within the general structures **1**–**5** and fall into the category of secondary metabolites.

Illustrative examples of secondary metabolites characterized by structure **2** are gliotoxin (**6**), sporidesmin B (**7**), and sirodesmin A.

Numerous α,β-dehydroamino acids **5** have been identified in recent years as constituents of fungal metabolites[1b], e.g. neoechinulin B. In most of these metabolites D-amino acids **1** also occur. Another class of uncommon amino acids, i.e. *N*-hydroxyamino acids **4**, can be recognized in microbial metabolites, e.g. mycelianamide and astechrome.

Several examples of secondary metabolites containing the β-functionalized amino acid derivative **3** are represented by phalloin (**8**), sparsomycin (**9**), and γ-glutamylmarasmine (**10**).

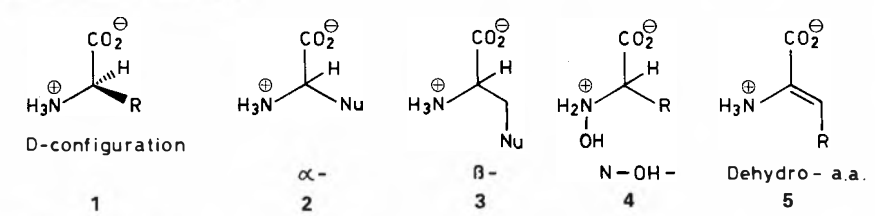
A fascinating question is whether there is a biogenetical or chemical relationship between L-amino acids and the uncommon amino acids **1**–**5**. Let us first address a possible biogenetical relationship.



Henricus (Harry) Ottenheijm: Born on April 30, 1943 in Maastricht, the Netherlands. Vordiplom at the University of Groningen (1963) and Dipl.-Chem. at the University of Frankfurt am Main (1965); Dr. rer. nat. at the University of Frankfurt am Main, 1968, with Th. Wieland, Thesis on «Die Konstitution des Antamanids und die Synthese eines Analogons». Postdoctoral positions at (1) the University of California, Berkeley, 1968–1969, with M. Calvin and A. Bassham, research on photosynthesis, Fellow of the U.S. Atomic Energy Commission; (2) Cornell University, Ithaca, 1969–1970, with J. Meinwald, synthesis of pheromones; (3) National Institutes of Health, Bethesda, 1970–1971, with B. Witkop, natural products synthesis, research associate. Since 1971 senior research associate at the University of Nijmegen, the Netherlands; 1974 tenured position, reader. Niels Stensen Foundation Fellow, January–August 1983, visiting faculty at the California Institute of Technology with D. Evans. Research interests: bioorganic chemistry, structure elucidation and synthesis of biologically active natural products, biosynthesis, structure-activity relationship studies; computer-assisted organic synthesis and computer-assisted molecular modelling.

α-Amino acids as building blocks of natural products

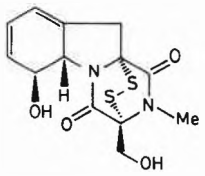
uncommon amino acids



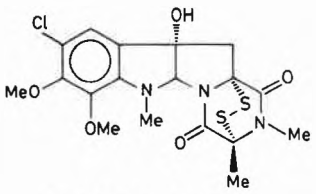
* *Correspondence:* Dr. H. C. J. Ottenheijm
Laboratory of Organic Chemistry
University of Nijmegen
Toernooiveld, NL-6525 ED Nijmegen
(The Netherlands)

** This article is based on a lecture presented at the University of Zürich, Department of Organic Chemistry, June 26, 1984.

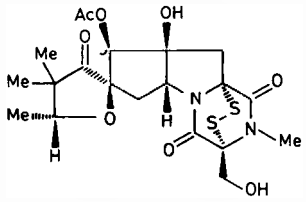
uncommon amino acid moieties of fungal metabolites



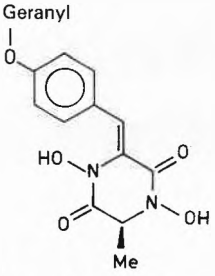
6 gliotoxin



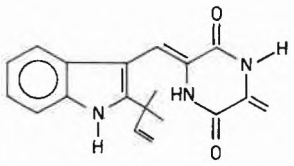
7 sporidesmin B



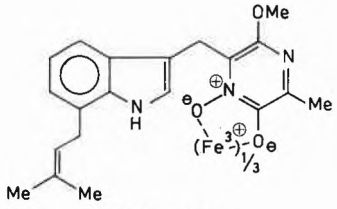
sirodesmin A



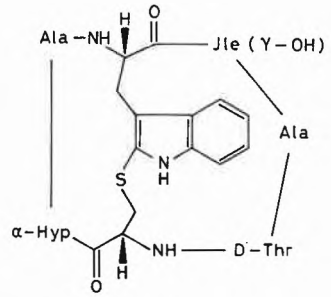
mycelianamide



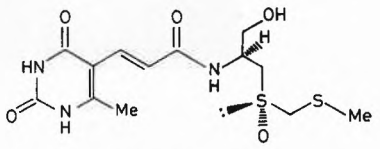
neo-echinulin B



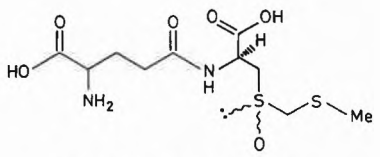
astechrome



8 phalloin



9 sparsomycin



10 γ-glutamyl-marasmine

One pathway for the formation of α-functionalized amino acids **2** from L-amino acids is by direct oxidation, as has been discussed in the biosynthesis of the tripeptide part of the ergotalkaloids[2]. A second pathway might involve a β-elimination reaction from serine, cysteine or threonine to yield dehydroamino acids **5**. A third possible pathway, which has been discussed too by *Schmidt* et al.[1b] is depicted in Scheme 1. It features oxidation of an amino acid to give a *N*-hydroxyamino acid **4**. Whereas the first two routes undoubtedly are of biogenetic relevance, the last mentioned route might also account for at least part of the metabolism of amino acids in lower organisms. This hypothesis is based on the following considerations:

First, of the three routes mentioned, only the last one links *N*-hydroxyamino acids **4** with the other uncommon amino acids **2**, **3**, and **5**. Second, it has been shown[3] that *N*-hydroxylation of amino acids is an important reaction in amino acid metabolism in microorganisms and plants. Third, several of the organisms that produce fungal metabolites featuring the uncommon amino acids **1**–**3** and **5** also produce *N*-hydroxyamino acid **4** containing metabolites. The demonstration of *N*-hydroxyamino acids as intermediates in metabolic pathways, which has not received much attention until recently, has been impeded by the instability of these compounds, by the lack of a general synthesis and proper analytical techniques, and by their occurrence in only minute quantities in biological material.

In the first part of this report we wish to demonstrate that Scheme 1 deserves attention not only as an outline of a biological relationship between L-amino acids and the uncommon amino acids **2**–**5**, but also as a chemosynthetic chart. Although the direct oxidation of amino acids into *N*-hydroxyamino acids **4** has not yet been achieved, it is our intention to show that *N*-hydroxyamino acids **4** are good synthons for the other uncommon amino acids **2**, **3**, and **5**.

In the second part of this report we will disclose some new reactions we developed recently in the domain of indole alkaloids.

In the third part of this report the synthesis of sparsomycin (**9**), a natural product featuring the β-mercapto-amino acid monooxodithioacetal moiety **11** will be discussed. So far, only two representatives of this class of compounds are known, i.e. sparsomycin (**9**) and γ-glutamyl-marasmin (**10**). In addition results of our structure-activity relationship studies of this antitumor compound **9** are disclosed.

2. α-Functionalized Amino Acid Derivatives

2.1. A Biogenetic Approach

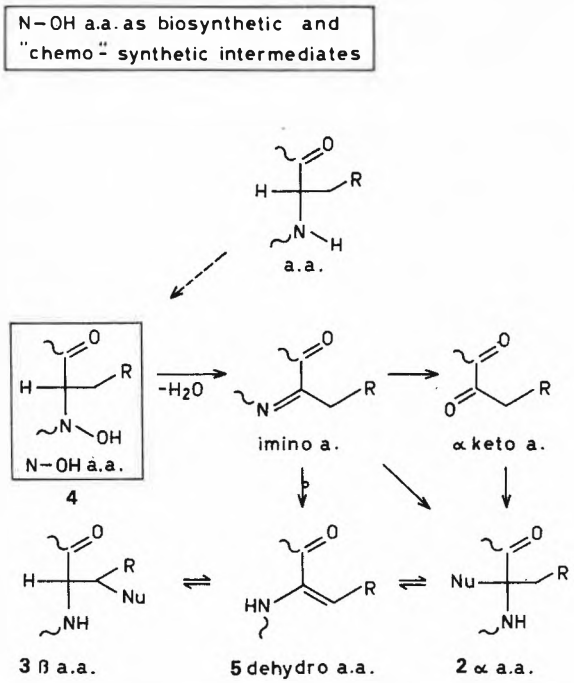
One of the targets that we settled upon originally was gliotoxin (**6**), in part because of its structure and in part because of its biological activity: it inhibits reverse transcriptase, an enzyme characteristic for RNA-viruses.

Gliotoxin can be regarded as an oxidized condensation product of two α-mercapto-α-amino acid derivatives **12**. However, neither of these derivatives is evidently capable of independent existence; unacylated α-mercapto-α-amino acids could not yet be synthesized.

Accordingly, we felt that a synthetic procedure for gliotoxin analogues would have to create a functional group at the indoline C-2 position, convertible to a mercapto group, simultaneously with the acylation of the indoline nitrogen atom by an α-mercapto-α-amino acid equivalent. Our initial, successful approach[4] involved the addition of pyruvoyl chloride[5] to the imine bond of an indolenine and the intramolecular cyclization of an amide nitrogen with the pyruvoyl α-carbonyl group (see **13**).

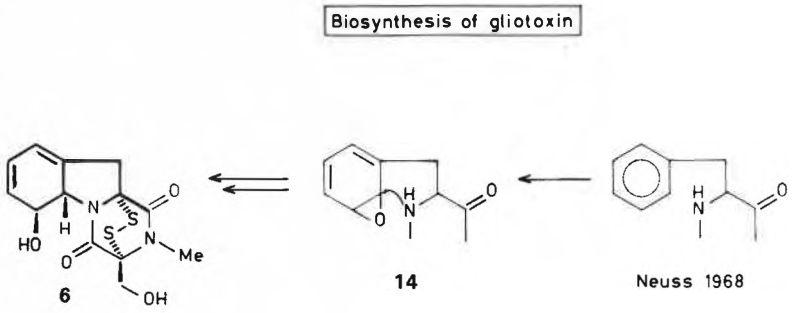
This approach – which is certainly not biogenetic – led us to speculate on the biosynthesis of gliotoxin. *cyclo*-L-Phe-L-Ser has been shown to be an efficient precursor of gliotoxin (**6**), and further labeling studies have demonstrated that the *N*-methyl group is derived from methionine, whereas the sulfur atoms are delivered by cysteine[6]. The most likely explanation for the formation of the dihydroaromatic sys-

Scheme 1



tem has been provided by *Neuss* et al.[7], who invoked the intermediacy of a benzene oxide **14** (Scheme 2).

Scheme 2



We have proposed a mechanism for the introduction of the sulfur bridge[8]. This proposal features oxidation of the dioxopiperazine amide nitrogen in **15** to form the hydroxamic acid **16** followed by dehydration to the acylimine **17** (Scheme 3).

We reasoned that the proposed role of *N*-hydroxy-α-amino acids **16** in the biosynthetic conversion of α-amino acids **15** into α-functionalized α-amino acids **18** might gain in probability if the latter could be obtained «chemosynthetically» by starting from *N*-hydroxy-α-amino acids. In Scheme 4 some results toward this directive are depicted, which lent support to our hypothesis.

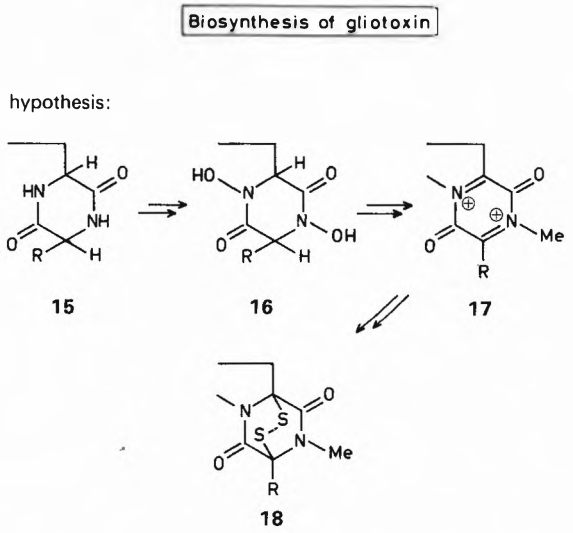
Selective reduction[9] of the oxime C=N bond of **19** and subsequent *N*-acylation of **20** yielded *N*-hydroxy-α-amino acid derivatives **21**. Treatment with base (*t*-BuOK in MeOH) afforded the α-methoxy-α-amino acid derivative **24**, a reaction for which we proposed the acylimine **22** as an intermediate[10]. If this acylimine is formed in the absence of a nucleophile (1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) in dioxane) it rearranges into a dehydro-α-amino acid **23**[11].

Subsequently we were able to show[8] that this approach for α-functionalization by transposition of an *N*-functionality could also be achieved in dioxopiperazines, e.g. **25**, as outlined in Scheme 5.

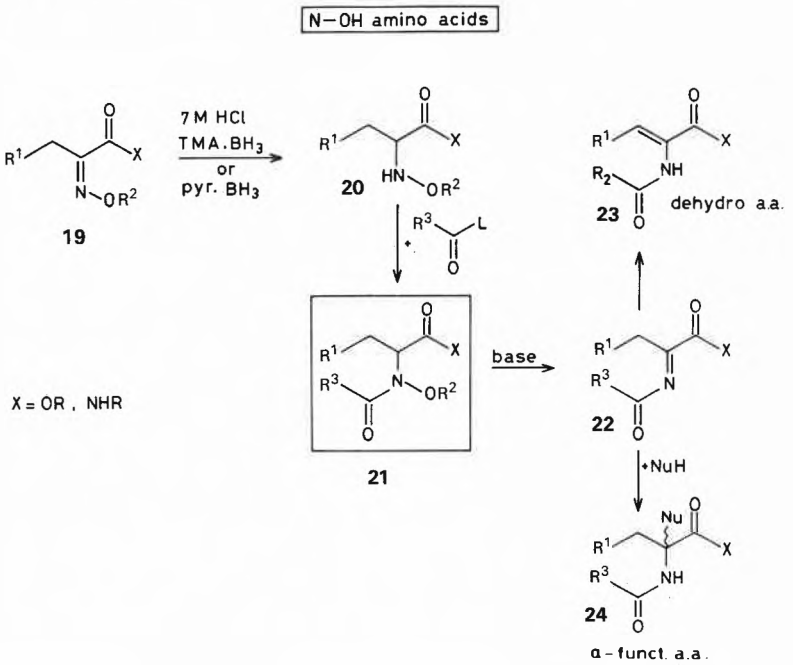
The conversion **26**→**27** deserves an explanation. The Markownikoff-type H_2S addition to the *exo*-methylene bond of **26** was found to proceed in a diastereoselective fashion; only the *cis*-dithiol **27** could be detected. This $ZnCl_2$-catalyzed reaction could be explained according to Scheme 6.

A zinc complex with the SH-group in **28** might direct the incoming SH-group from the same face by complexation, yielding **27**. A second role for the catalyst is to convert H_2S into a stronger acid; when **28** is exposed to H_2S in the absence of zinc chloride no reaction took place[4].

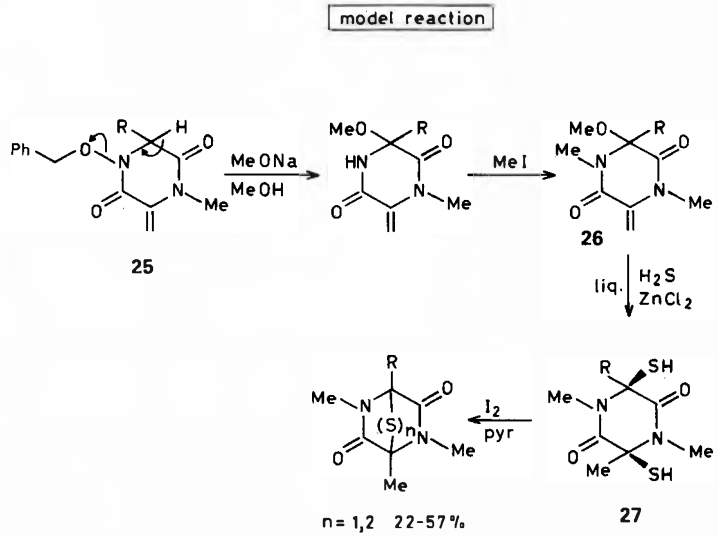
Scheme 3



Scheme 4



Scheme 5



Presently, the conversion of Bis(*N*-hydroxy)dioxopiperzines[12] **29** into sulfur bridged dioxopiperazines **30** is being studied.

2.2. A Synthetic Approach to Sporidesmin B

Encouraged by these results, we selected sporidesmin B (**7**) as our next target and focussed on the conversion of *N*-hydroxytryptophan **31** into the corresponding α-functionalized amino acid.

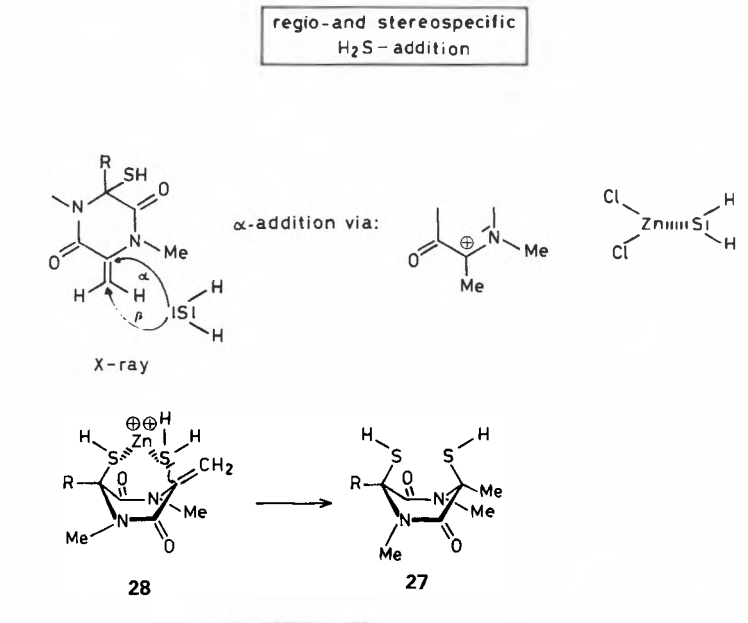
Two syntheses of **31** were developed[13]. One procedure started from indole-3-pyruvic acid which was converted into the corresponding *O*-benzyl oxime ethyl ester. Methylation of the indole nitrogen and reduction of the oxime C=N bond with $(CH_3)_3N\text{-}BH_3$ ($TMA \cdot BH_3$) afforded the *N*-benzyloxy amino acid ester (cf. **19**→**20**). The second route (Scheme 7) features[14] the reaction of *N*-methylindole (**32**, $R^1 = H$) with the transient nitroso olefin **34**, prepared from **33**. The adduct **35** cannot be isolated; base catalyzed rearomatization afforded **36**. Reduction gave **37** in good yields.

Pyruvoyl chloride[5] and **37** in CH_2Cl_2/ether reacted at room temperature to form **38** (Scheme 8); no *O*-acylation was observed. About 24 h after mixing the intermediate was converted completely into **39**. When stirred for several days after the addition of methanol, the corresponding α-methoxy derivative was formed, which was benzylated to give **40**. Diastereomeric induction was observed; the indolyl group and the methyl group are in a *cis*-relationship in the major isomer (90%).

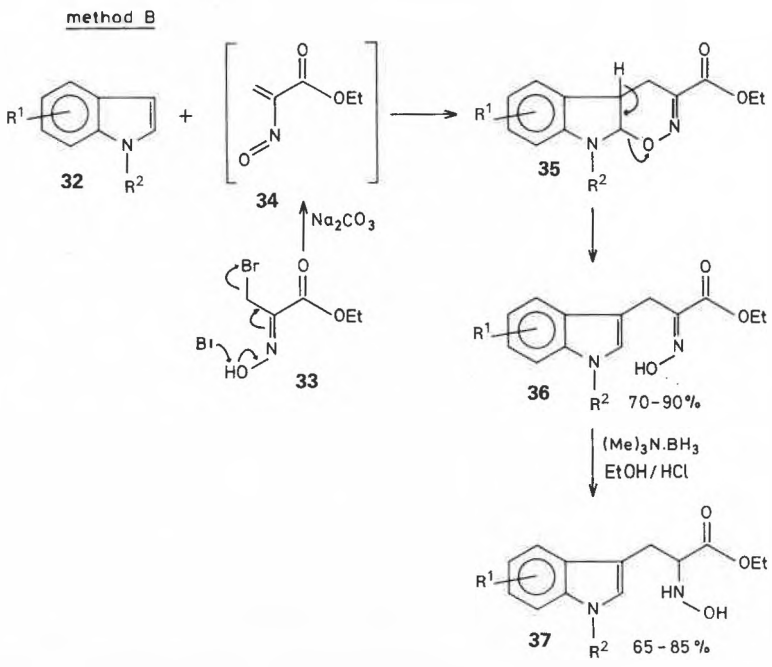
Transposition of the *N*-functionality in **40** to an α-functionality (→**41**) was achieved by treatment with CH_3ONa/CH_3OH (Scheme 9). The diastereomers were separated and each diastereomer was subjected to the following reaction. For closing of the five-membered ring singlet oxygen was used[15]. A reliable procedure involves methylene blue as sensitizer and a reaction temperature of −78 °C; reduction of the peroxo function in **42** led smoothly to **43**.

Hopefully the remaining stages will directly follow the sequence outlined in Scheme 5 to allow conversion of **43** into the corresponding disulfide bridged dioxopiperazine. A total synthesis of sporidesmin B (**7**) seems then feasible by using the properly substituted indole derivative[16] instead of **32** (see Scheme 7).

Scheme 6



Scheme 7



3. Reactions of a New Synthon: Syntheses of Indole Alkaloids

In the synthesis of the *N*-hydroxytryptophan derivative **37** from indole **32** we observed the primarily formed cycloadduct **35** as an intermediate that escaped isolation. Intrigued by the potential usefulness of this adduct for other reaction sequences, we have explored the chemistry of **44** formed by reaction of the corresponding indole derivative and the nitroso olefin **34** (Scheme 10). This cycloaddition proceeds smoothly even with the sterically demanding isopentenyl group at the C-2 position of indole; the adduct undergoes base catalysed ring opening followed by rearomatization to afford **45**. Work is in progress to convert **45** into fungal metabolites of the neo-echinulin class[13].

A new rearrangement was observed[17] in the cycloaddition of the nitroso olefin **34** with indoles having an alkylthio substituent at C-3. This reaction yielded **46**, in which the alkylthio group had migrated from C-3 to C-2. This rearrangement occurs in good yields under mild conditions (room temperature, CH_2Cl_2, Na_2CO_3) and has been used recently by us[18] in a novel approach to the 2-(*S*-cysteinyl)tryptophan derivative **53**, which is the characteristic structural element of the toxic principles (e.g. Phalloin **8**) of members of the genus *Amanita*. The results are summarized in Scheme 11 and Scheme 12.

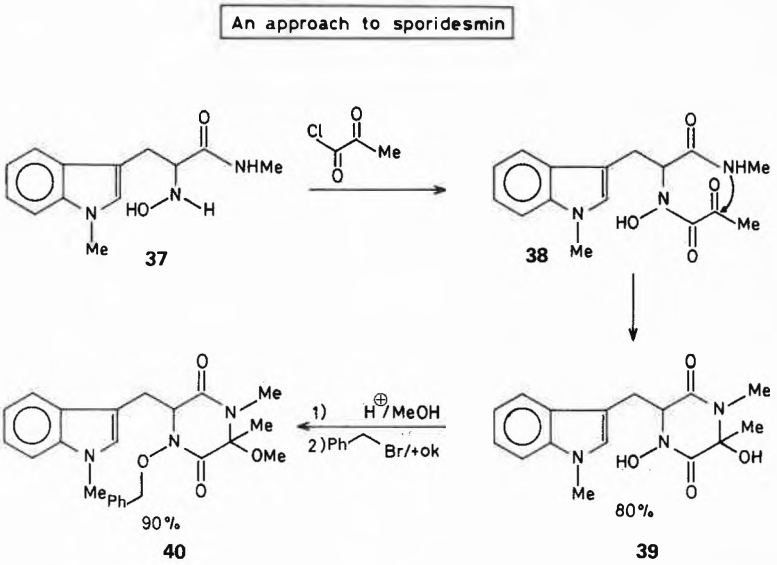
Stable cycloadducts **44** are formed when the substituent X has a low migratatory aptitude, e.g. when X is an alkyl or acetoxy group. A borderline case arises when the alkyl group is an isopentenyl group; treatment with trifluoroacetic acid causes an 1,2-allylshift to yield **47** (Scheme 10). Work is in progress to use this reaction in a synthetic approach to members of the fumitremorgen class which have **47** as structural feature.

The adduct **52** (**44** with X = OAc) has also been studied (Scheme 13). It is a stable compound which shows no tendency to rearrangement. Interestingly, the dihydro-1,2-oxazin ring of **52** is susceptible to nucleophilic attack: treatment with hydrides and hydrosulfides afforded the indole derivative **54**. We have used this approach for a second synthesis of tryptathionine (**51**→**52**→**50**; Scheme 11).

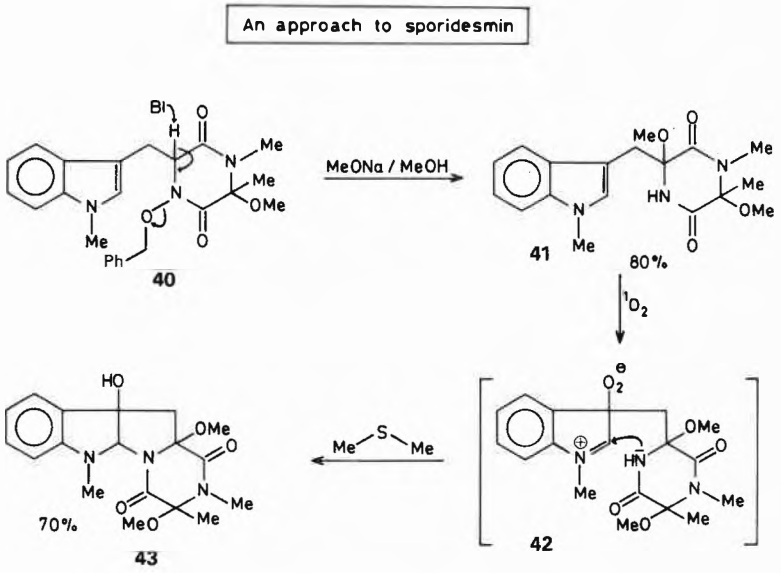
A mechanistic rationale for the reactions depicted in Schemes 10 and 13 is presented in Scheme 14. The indolenine derivative **44a** (X = SR), in equilibrium with **55a** forms the episulfonium species **56**, which undergoes rearomatization to yield **46**. In case of X = OAc (**44b**), the intermediate indolenine **55b** has a lower tendency to rearrange so that an external nucleophile can be added instead to yield **57**. Subsequent elimination of AcOH yields **54**.

In conclusion the adduct **44** opens a new approach to indole alkaloids, both in terms of strategy and methodology. We feel that *N*-hydroxyamino acids in general are valuable synthons for a wide range of natural products.

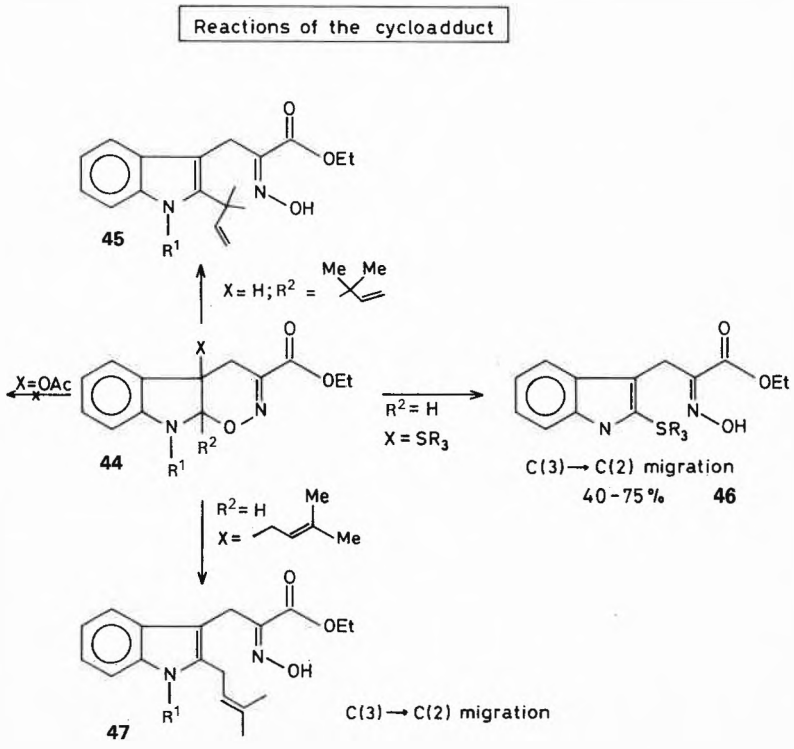
Scheme 8



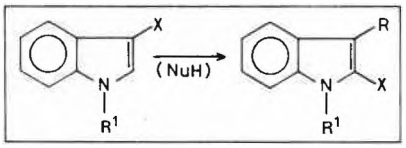
Scheme 9

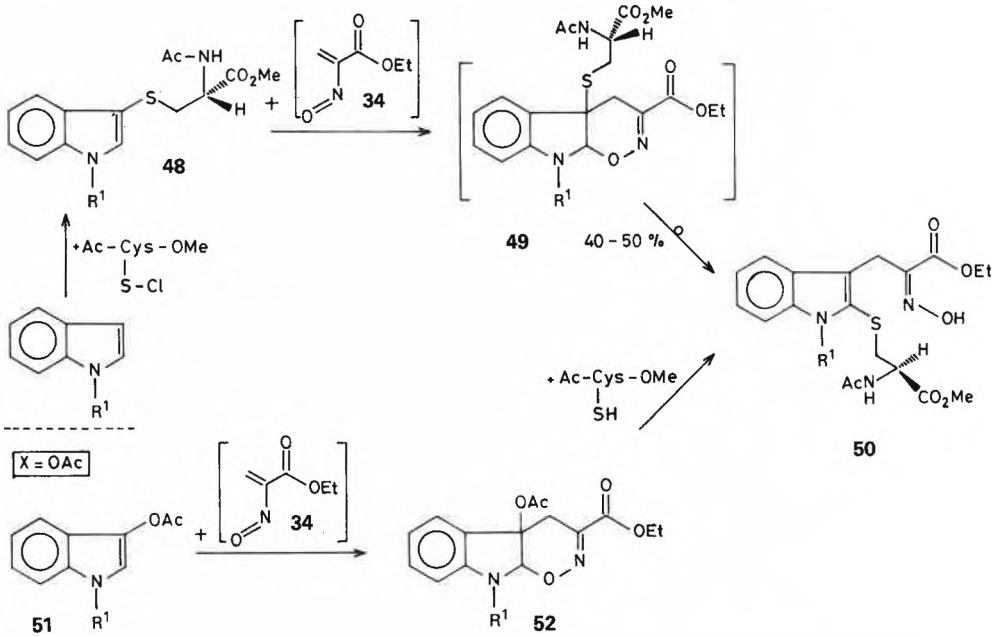


Scheme 10

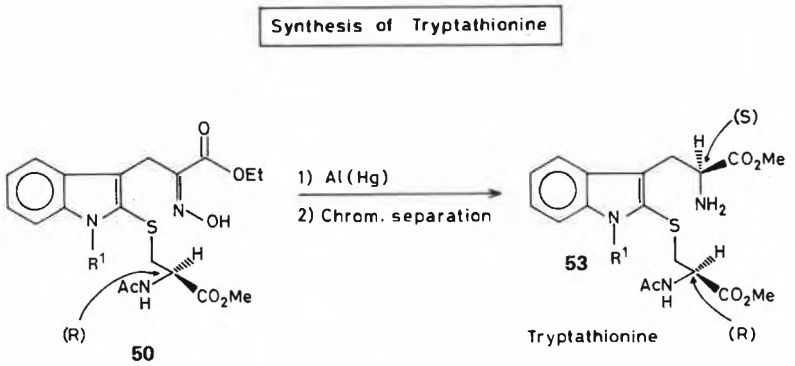


Scheme 11

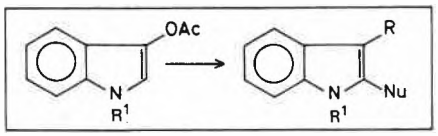


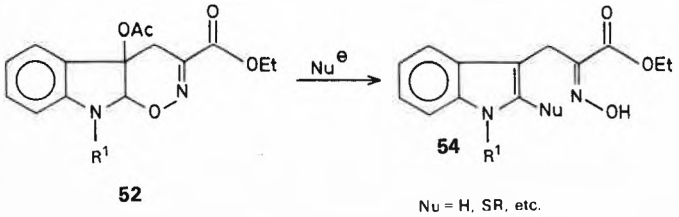


Scheme 12



Scheme 13





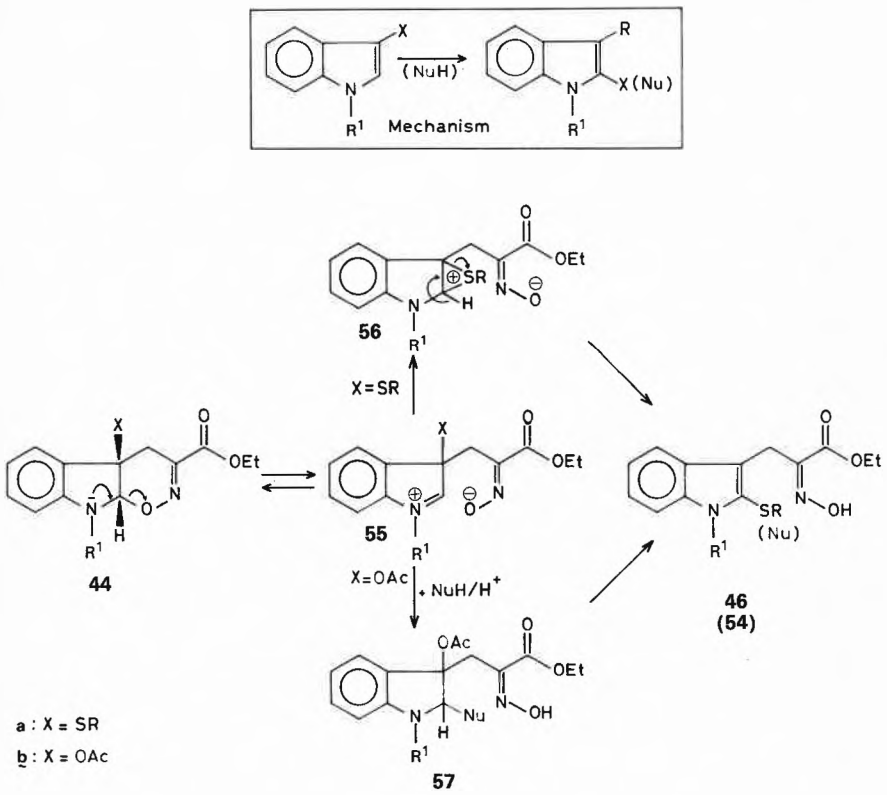
4. Sparsomycin: Synthesis and Structure-Activity-Relationship Studies

Sparsomycin (**9**), a metabolite of *Streptomyces sparsogenes* or *Streptomyces cuspidospores*, has attracted much attention, partly because of its unusual structure – notice the in nature rarely encountered $-S(O)CH_2S-$moiety – and partly because of its biological activity of which the antitumor activity and the inhibition of protein biosynthesis are the most striking properties[19]. On the basis of spectroscopic and degradation studies[20] the presently accepted structure **9** was proposed; however, the chirality of the sulfoxide sulfur atom remained undetermined in these studies.

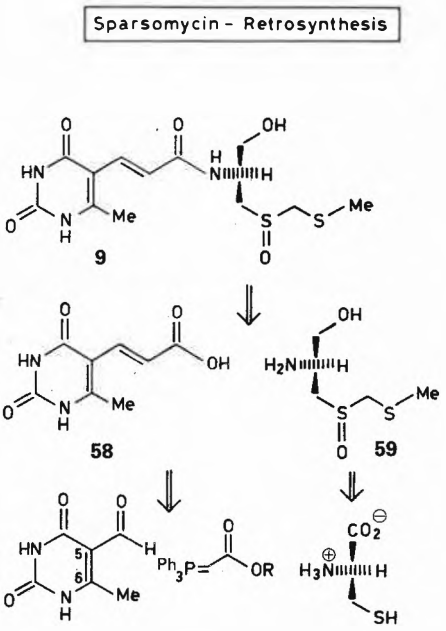
In an analysis of the synthetic problem (Scheme 15) sparsomycin can be considered as an amide derived from the acid component **58** and the amine component **59**. The latter can be regarded as a derivative of D-cysteine having its D CO_2H-function reduced and its D SH-function alkylated and oxidized.

Component **58** could be prepared by using a Wittig condensation[19]. More challenging was the synthesis of component **59**, since the unsymmetrically substituted $-S(O)CH_2S-$moiety is acid labile and may also undergo β-elimination – thermal or base-induced – to which sulfoxides are prone. An attractive approach to a protected derivative of **59**, i.e. **59a**, appeared to be nucleophilic ring opening of a cyclic sulfinate **60**, a γ-sultine (Scheme 16). This compound has a sulfur atom activated toward nucleophilic attack as well as a protected alcohol function. This approach was a viable one: we have synthesized sultines of type **60** starting from D-cysteine and

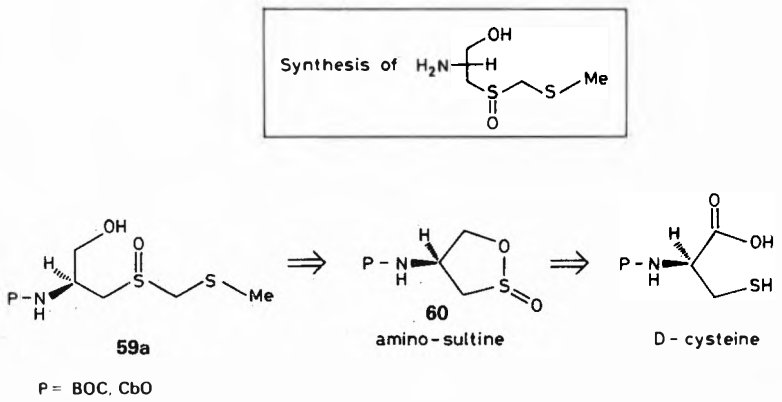
Scheme 14



Scheme 15



Scheme 16



have studied their ring opening reactions with nucleophiles.

The *N*-protected D-cystinol derivative **62**, prepared from **61** by $LiBH_4$-reduction followed by I_2-oxidation, was treated with three equivalents of NCS in AcOH to afford **60a** and **60b** (1:1 ratio) in 87% yield[21] (Scheme 17). The structures – including the absolute configuration of the sulfur atoms – were assigned by CD spectroscopy and X-ray crystallographic analysis[21,22].

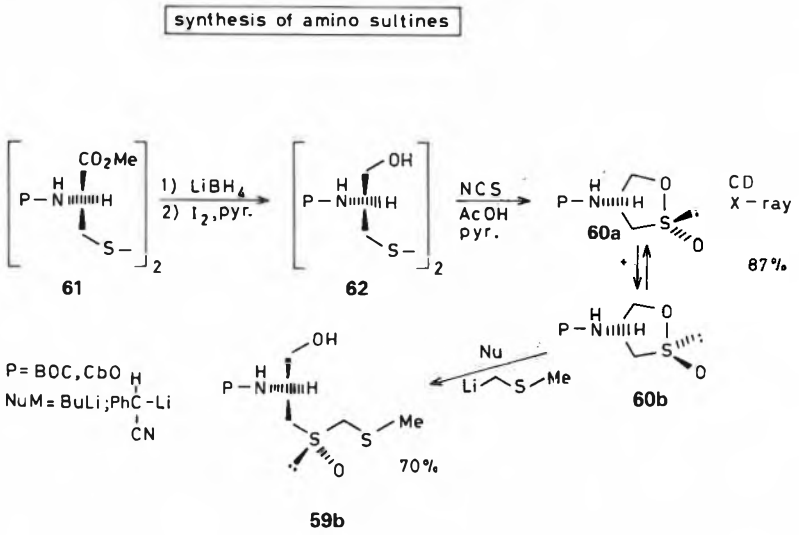
We were pleased to find that the sultines underwent nucleophilic ring opening smoothly and with inversion at sulfur: e.g. treatment of **60b** with $MeSCH_2Li$ gave **59b** in 70% yield. The amino alcohol **63b** (Scheme 18) was prepared quantitatively by treatment of **59b** (P = *tert*-butoxycarbonyl) with CF_3CO_2H at 0°C and subsequent deprotonation with an ion-exchange resin. Coupling of **58** with **63b** was achieved by means of dicyclohexylcarbodiimide (DCC) and hydroxybenzotriazole to yield a compound that was identical with sparsomycin in all respects. From this we concluded that sparsomycin's sulfoxide sulfur atom must have the *R*-chirality.

Sparsomycin's three stereoisomers **64**–**66** were prepared using the same approach. Finally, the synthesis of sparsomycin was optimized by making use of the finding[23] that sultines **60a** and **60b** undergo a clean epimerization at the sulfur atom when heated at 120–130°C. Consequently, the «wrong» isomer **60b** was recycled by conversion into a 1:1 mixture of **60a** and **60b**, out of which **60b** was readily isolated by flash column chromatography.

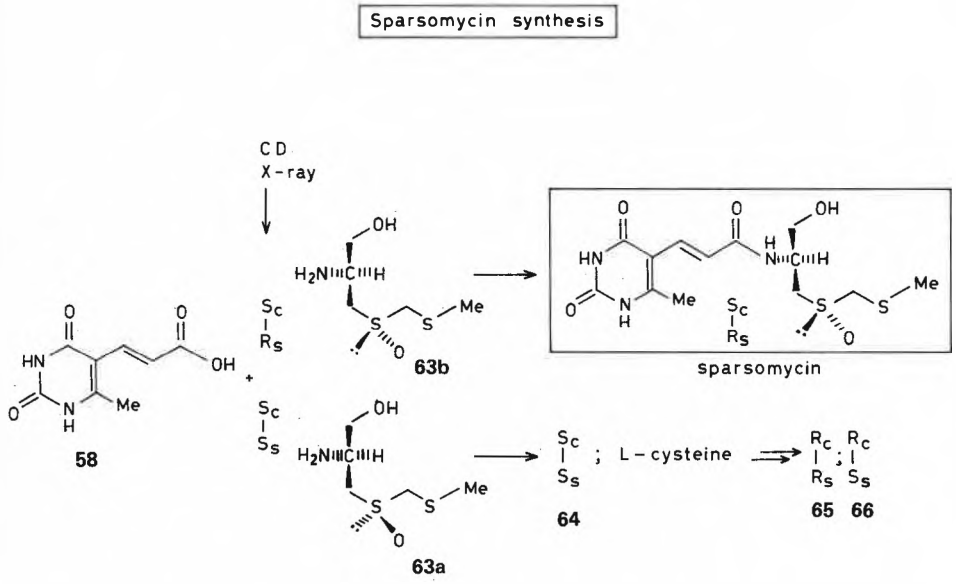
In the course of our synthetic work on **60**, we found[18,21] that selective cleavage of the S–OR bond in sultines can be achieved by a wide variety of nucleophiles (n-$Bu^{\ominus}$, $^{\ominus}CH_2CN$, $^{\ominus}CH_2CO_2tBu$, $^{\ominus}CH_2P(O)(OEt)_2$, $^{\ominus}CH(C_6H_5)CN$, and $^{\ominus}C(CH_3)(C_6H_5)CN$). With the last two nucleophiles asymmetric induction on the prochiral carbon atom was observed. In order to understand this induction we studied[24] the conformation of **60a** and **60b** in solution by ^{1}H-NMR spectroscopy. We also studied[23] the thermolytic SO_2-extrusion of the functionalized sultines. Flash vacuum thermolysis yielded a *Z*/*E*-mixture of the corresponding *N*-protected enamides, a reaction we explained by a novel migration of the benzamide group involving a Claisen-type rearrangement.

The aforementioned approach to **9** and another one we had published previously[19] was flexible enough to allow the synthesis of sixteen structural analogues of sparsomycin. Sparsomycin and these analogues were tested in cell-free systems for their ability to inhibit the protein synthesis[18] as well as in an in vitro clonogenic assay for their antitumor activity[25]. The results of these assays indicate that sparsomycin's antitumor activity is primarily due to an inhibition of the protein biosynthesis. In addition, we were able to draw conclusions about the structural features that are required for sparsomycin's antitumor ac-

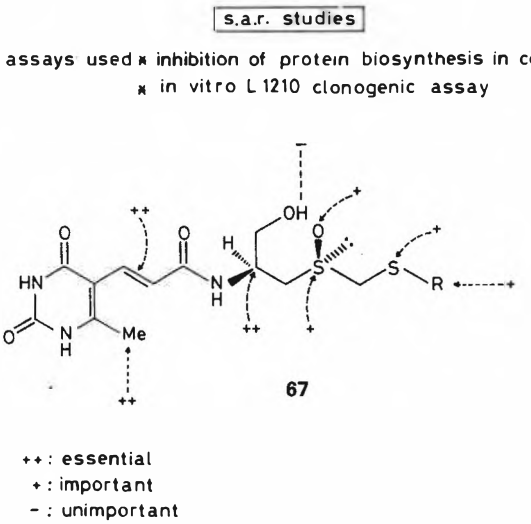
Scheme 17



Scheme 18



Scheme 19



tivity. These conclusions are summarized in Scheme 19.

These results lend support[26] to a mechanistic rationale for the activity of sparsomycin on a molecular basis: it has been proposed[27] that the inhibitory activity might be due to a Pummerer-like reaction involving sparsomycin's sulfoxide moiety causing an irreversible blocking of the growing peptide chain.

Finally, our approach resulted – for the time being – in the synthesis of two analogues – benzylsparsomycin[18] (**67**, $R = CH_2C_6H_5$) and octylsparsomycin[25] (**67**, $R = nC_8H_{17}$) – that have a higher cytostatic activity and a lower toxicity than sparsomycin (**67**, $R = CH_3$). These results indicate among others that the ribosomal binding site for sparsomycin has a lipophilic character. Work is in progress to obtain additional arguments for the introduction of the most promising analogue of sparsomycin into the clinic as an antitumor agent.

In conclusion, all these results underline our view that a flexible synthesis of a biologically active compound is a prerequisite for an analysis of the structural features that are essential for an optimal activity as well as for a study of the mode of action of that compound.

Acknowledgements: I wish to thank my coworkers for their patience and dedication in collaborating in the work I have described. I am particularly indebted to Dr. J.D.M. Herscheid, Dr. R.M.J. Liskamp, and to Mr. R. Plate. We are grateful to Prof. R.J.F. Nivard for his support.

Received: November 26, 1984 [FR 6]

[1] a) For a recent review on amino acids see: I. Wagner, H. Musso, *Angew. Chem. 95* (1983) 827; *Angew. Chem. Int. Ed. Engl. 22* (1983) 816; b) The available data on α,β-dehydroamino acids and α-functionalized amino acids have been the subject of a survey by U. Schmidt, J. Hausler, E. Oehler, H. Poisel in W. Herz, H. Grisebach, G.W. Kirby: *Progress in the Chemistry of Natural Products*, Springer-Verlag, Berlin 1979, Vol. *37*, p. 251.
[2] F.R. Quigley, H.G. Floss, *J. Org. Chem. 46* (1981) 464.
[3] For a review see B.L. Moller in B. Vennesland, E.E. Conn, C.J. Knowles, J. Westley: *Cyanide in Biology*, Academic Press, New York 1981, p. 179.
[4] H.C.J. Ottenheijm, J.D.M. Herscheid, G.P.C. Kerkhoff, T.F. Spande, *J. Org. Chem. 41* (1976) 3433.
[5] H.C.J. Ottenheijm, M.W. Tijhuis, *Org. Synth. 61* (1983) 1.
[6] See references cited in [7].
[7] N. Neuss, R. Nagarajan, B.B. Mallory, L.L. Huckstep, *Tetrahedron Lett. 9* (1968) 4467.
[8] J.D.M. Herscheid, R.J.F. Nivard, M.W. Tijhuis, H.C.J. Ottenheijm, *J. Org. Chem. 45* (1980) 1885.
[9] M.W. Tijhuis, J.D.M. Herscheid, H.C.J. Ottenheijm, *Synthesis* (1980) 890.
[10] J.D.M. Herscheid, R.J.F. Nivard, M.W. Tijhuis, H.P.H. Scholten, H.C.J. Ottenheijm, *J. Org. Chem. 45* (1980) 1980.
[11] J.D.M. Herscheid, H.P.H. Scholten, M.W. Tijhuis, H.C.J. Ottenheijm, *Recl. Trav. Chim. Pays-Bas 100* (1981) 73.
[12] J.D.M. Herscheid, J.H. Colstee, H.C.J. Ottenheijm, *J. Org. Chem. 46* (1981) 3346.
[13] H.C.J. Ottenheijm, R. Plate, J.H. Noordik, J.D.M. Herscheid, *J. Org. Chem. 47* (1982) 2147.
[14] The versatile cycloaddition of a nitroso olefin to a carbon-carbon double bond has been described for the first time by T.L. Gilchrist, D.A. Longham, T.G. Roberts, *J. Chem. Soc. Chem. Commun.* (1979) 1089.
[15] M. Nakagawa, S. Kato, S. Kataoka, S. Kodato, H. Watanabe, H. Okajima, T. Hino, B. Witkop, *Chem. Pharm. Bull. 29* (1981) 1013.

[16] J.W. Blunt, A.F. Erasmuson, R.J. Ferrier, M.H.G. Munro, *Aust. J. Chem. 32* (1979) 1013.
[17] R. Plate, H.C.J. Ottenheijm, R.J.F. Nivard, *J. Org. Chem. 49* (1984) 540.
[18] H.C.J. Ottenheijm, unpublished results.
[19] H.C.J. Ottenheijm, R.M.J. Liskamp, S.P.J.M. van Nispen, H.A. Boots, M.W. Tijhuis, *J. Org. Chem. 46* (1981) 3273 and references cited therein.
[20] P.F. Wiley, J. McKellar, *J. Org. Chem. 41* (1976) 1858.
[21] R.M.J. Liskamp, H.J.M. Zeegers, H.C.J. Ottenheijm, *J. Org. Chem. 46* (1981) 5408.
[22] H.C.J. Ottenheijm, R.M.J. Liskamp, P. Helquist, J.W. Lauher, M.S. Shekhani, *J. Am. Chem. Soc. 103* (1981) 1720.
[23] R.M.J. Liskamp, H.J. Blom, R.J.F. Nivard, H.C.J. Ottenheijm, *J. Org. Chem. 48* (1983) 2733.
[24] C.A.G. Haasnoot, R.M.J. Liskamp, P.A.W. van Dael, J.H. Noordik, H.C.J. Ottenheijm, *J. Am. Chem. Soc. 105* (1983) 5406.
[25] R.M.J. Liskamp, J.H. Colstee, H.C.J. Ottenheijm, P. Lelieveld, W. Akkerman, *J. Med. Chem. 27* (1984) 301.
[26] R.J. Ash, G.A. Flynn, R.M.J. Liskamp, H.C.J. Ottenheijm, unpublished results.
[27] G.A. Flynn, R.J. Ash, *Biochem. Biophys. Res. Commun. 114* (1983) 1.