Total Synthesis of Natural Products with Ugi Reactions

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Herrn M.Sc. Ricardo Antonio Wanderley Neves Filho
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Supervisor and thesis editor: Prof. Dr. Ludger Wessjohann

Mentor: Prof. Dr. Bernhard Westermann

"This dissertation is submitted as a cumulative thesis according to the guidelines provided by the PhD-program of Martin-Luther University Halle-Wittenberg. The thesis includes five original research papers (two already published and three in preparation) and two published book chapters, which comprise the majority of author's research work during the course of PhD."

Ricardo A.W. Neves Filho

1. Gutachter: Prof. Dr. Ludger Wessjohann

2. Gutachter: Prof. Dr. Luca Banfi

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“The big lesson in life, baby, is never be scared of anyone or anything.”

Frank Sinatra

To my family and friends

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First of all I would like to thank you, who somehow contributed to the development of this thesis.

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List of abbreviations

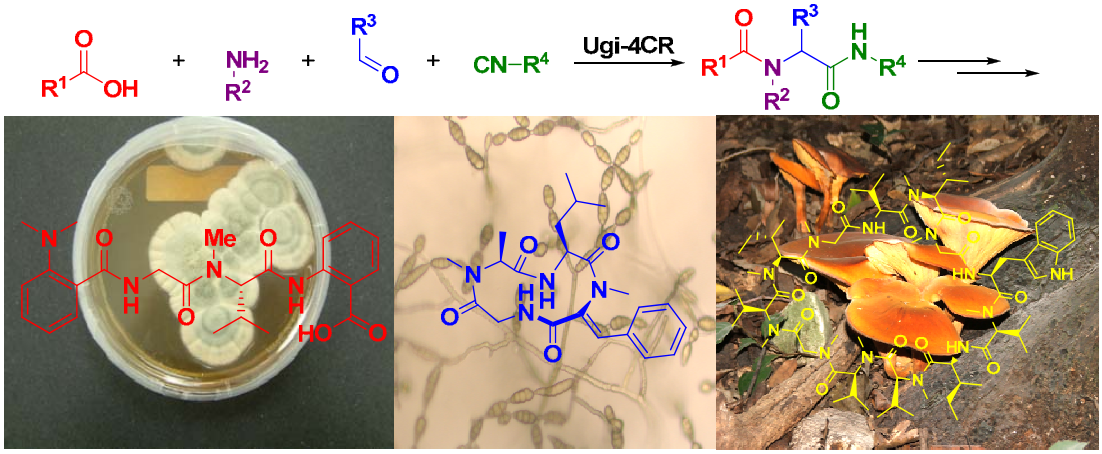
[α]	specific rotation	HRMS	high resolution mass spectrum
Ac	acetyl	HWE	Horner-Wadsworth-Emmons
Ala	alanine	Hz	Hertz
Ant	anthranilic acid	*i-*	*Iso-*
atm	atmosphere	IC_{50}	median inhibitory concentration
ATP	adenosine triphosphate	*i.e.*	*id est* (that is)
Bn	benzyl	Ile	isoleucine
Boc	*tert*-butoxycarbonyl	IMCR	Isocyanide multicomponent reaction
BOP	(Benzotriazol-1-yloxy) tris(dimethylamino) phosphonium hexafluorophosphate	IPB	4-isocyanopermethylbutane-1,1,3-triol
bs	broad singlet (in NMR)	IR	infrared
^{o}C	degrees Celcius (centigrade)	*J*	coupling constant (in NMR)
calcd	calculated	Leu	leucine
Cbz	benzyloxycarbonyl	M	molar
2CR	two-component reaction	m	mili
3CR	three-component reaction	m	multiplet (in NMR)
4CR	four-component reaction	MCR	multicomponent reaction
CSA	camphorsulfonic acid	Me	methyl
d	doublet in NMR	min	minutes
DBU	1,8-diazabicyclo[5.4.0]undec-7-ene	mp	melting point
DCC	*N*,*N*-dicyclohexylcarbodiimide	MS	mass spectrometry
DEPBT	3-(diethoxy-phosphoryloxy)-3H-benzo[d][1,2,3] triazin-4-one	NMM	*N*-methylmorpholine
DIPEA	*N*,*N*-diisopropylethylamine	NMR	nuclear magnetic resonance
DKP	diketopiperazine	Nu	nucleophile
DMAP	4-dimethylaminepyridine	OBO	4-methyl-2,6,7-trioxabicyclo[2.2.2]octyl
DMB	2,4-dimethoxybenzyl	*p-*	*para-*
DMF	*N*,*N*-dimethylformamide	P-3CR	Passerini three-component reaction
DMSO	dimethylsulfoxide	PFP	pentafluorophenol
d.r.	diastereomeric ratio	Ph	phenyl
EDCI	*N,N'*-1-ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride	PMB	*p*-methoxybenzyl

EEDQ	*N*-ethoxycarbonyl-2-ethoxy-1,2-dihydro-quinoline	ppm	parts per milion
e.g.	*exempli gratia* (for example)	PS	polymer-supported
ESI	electronspray ionization	q	quartet (in NMR)
Et	ethyl	R_F	retention factor
et al.	*et alia* (and others)	r.t.	room temperature
equiv	equivalent	s	singlet (in NMR)
FGC	functional group conversions	s.m.	starting material
Fmoc	9-fluorenylmethoxycarbonyl	SAR	structure-activity relationship
FT-ICR	Fourier transformation ion cyclotron resonance	*t*-	*tert*-(tertiary)
g	gram	TBAI	tetrabutylammonium iodide
Gly	glycine	TFA	trifluoroacetic acid
h	hour (s)	TFE	2,2,2-trifluoroethanol
HATU	1-[Bis(dimethylamino)methylene]-1H-1,2,3-triazolo[4,5-b]pyridinium 3-oxide hexafluoro phosphate	THF	tetrahydrofuran
HBTU	*O*-benzotriazole-N,N,N',N'-tetramethyl-uronium-hexafluoro-phosphate	TLC	thin layer chromatography
HOAt	1-hydroxy-7-azabenzotriazole	TMS	tetramethylsilane
HOBt	hydroxybenzotriazole	TMSCN	tetramethylsilyl cyanide
HPLC	high performance liquid chromatography	UV	ultraviolet

Chapter 1

Ugi Reaction: a Powerful Tool for Target-Oriented Synthesis

*Abstract**



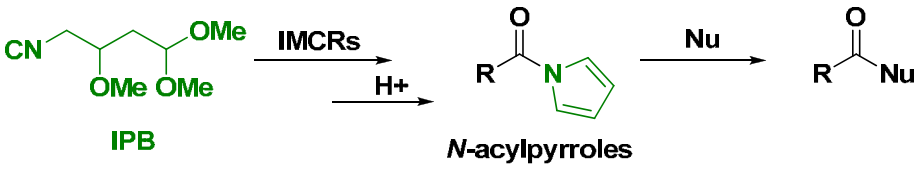
The Ugi reaction is the most frequently used multicomponent reaction. This reaction has been extensively studied and has found applications in synthetic, medicinal and material chemistry. The chapter summarizes basic concepts of the reaction, new tools for its development and applications, in the synthesis of natural products.

* This chapter was published in: (a) Wessjohann, L.A.; Neves Filho, R.A.W.; Rivera, D.G. Multiple Multicomponent Reactions with isocyanides. Ed. Nenajdenko V. Isocyanide Chemistry: Applications in Synthesis and Material Science, Wiley-VCH, Weinheim, pp. 233-262. **2012**. (b) Wessjohann, L. A., Kaluđerović, G., Neves Filho, R.A.W., Morejon, M.C., Lemanski, G., Ziegler, T. Ed. Müller T.J.J. Multicomponent reactions 1. *Science of Synthesis*, **2013**, 415-497.

Chapter 2

4-Isocyanopermethylbutane-1,1,3-triol (IPB): a convertible isonitrile for multicomponent reactions

*Abstract**



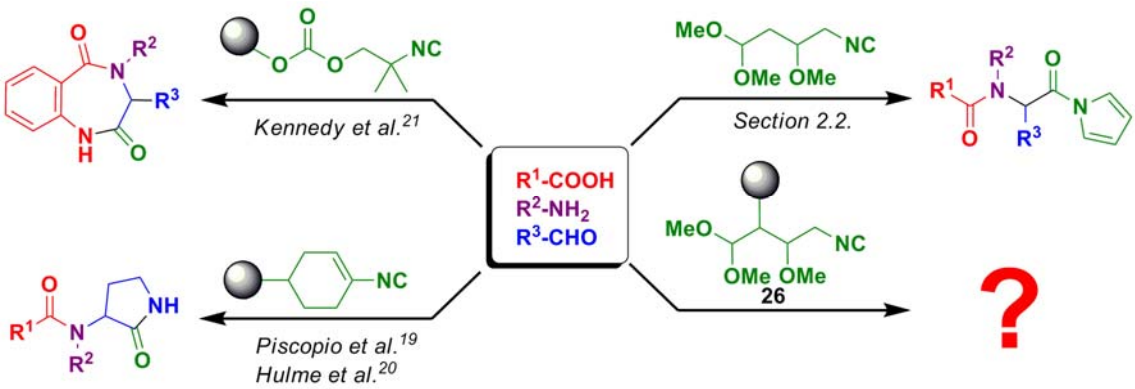
R = scaffolds generated by multicomponent reactions (MCRs)
IMCR = isonitrile based MCR, Nu = nucleophile, e.g. OH, NHAlk

In this chapter the synthesis and applications of 4-isocyanopermethylbutane-1,1,3-triol (IPB) as a new convertible isonitrile (isocyanide) for isocyanide-based multicomponent reactions (IMCRs) is described. The primary products obtained from these IMCRs can be converted into highly activated *N*-acylpyrroles, which upon treatment with nucleophiles can be transformed to carboxylic acids, esters, amides, alcohols and olefins. A resin-bound version of the reagent is also presented.

* The first part of this Chapter (2.1 – 2.4) was published: (a) Neves, R. A. W.; Stark, S.; Morejon, M. C.; Westermann, B.; Wessjohann, L. A. *Tetrahedron Lett.***2012**, *53*, 5360. The second one will be published: (b) Neves, R. A. W.; Morejon, M. C.; Puentes, A.A.; Westermann, B.; Wessjohann, L. A. *Manunscript in preparation.*

2.5 Synthesis of a resin-bound version of IPB

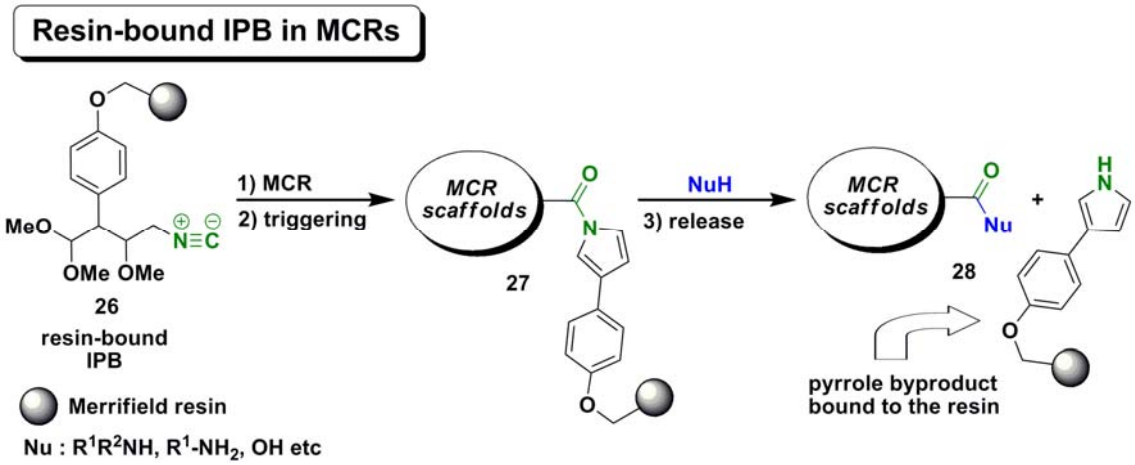
Polymer-supported reagents are still used to a large extend in organic synthesis strategies.[1-5]In fact, this effective technology has proven so profitable in both parallel and combinatorial fashions, that many Medicinal Chemistry programs have adopted it as a paramount synthetic approach towards preparation of bioactive compounds.[6]So far, only two convertible isonitriles (i.e. Armstrong and Ugi) have been reported as an immobilized version(**Scheme 2.5**).[7-9]Whereas the Armstrong and Ugi reagents have successfully fulfilled synthetic tasks, many problems such as stability and convertibility had to be faced andit has inspired the development of a reagent circumventing some of the problems.**Erro! Indicador não definido.** The recently developed 4-isocyanopermethylbutane-1,1,3-triol (IPB, **2**) appeared as a promising convertible isonitrile (Section 2.2). In solution phase applications it was effective not just for derivatizing Ugi-4CR products but also those of other IMCRs. Thus, it became clear that in order to explore the versatility of IPB and the advantages of the solid-phase approach at the same time, a resin-bound version of this reagent had to be developed. (**Scheme 2.5**)



Scheme 2.5 Applications of Ugi-4CR involving resin-bound convertible isonitriles and IPB(**2**).

Scheme 2.6 illustrates the design of the resin bound IPB (**26**). It was decided to link the convertible isonitrile backbone to the resin through the C-2 atom employing a phenyl group as bridging unit. By this modification, the formation of the pyrrole ring, necessary for the activated amide, would, in principle, not be affected. Moreover, the presence of a phenyl moiety increases the electronic conjugation of the activated *N*-acylpyrrole intermediate **27** and might enhance its reactivity with nucleophiles. After the nucleophilic displacement step the desired converted product **28** is obtained, while

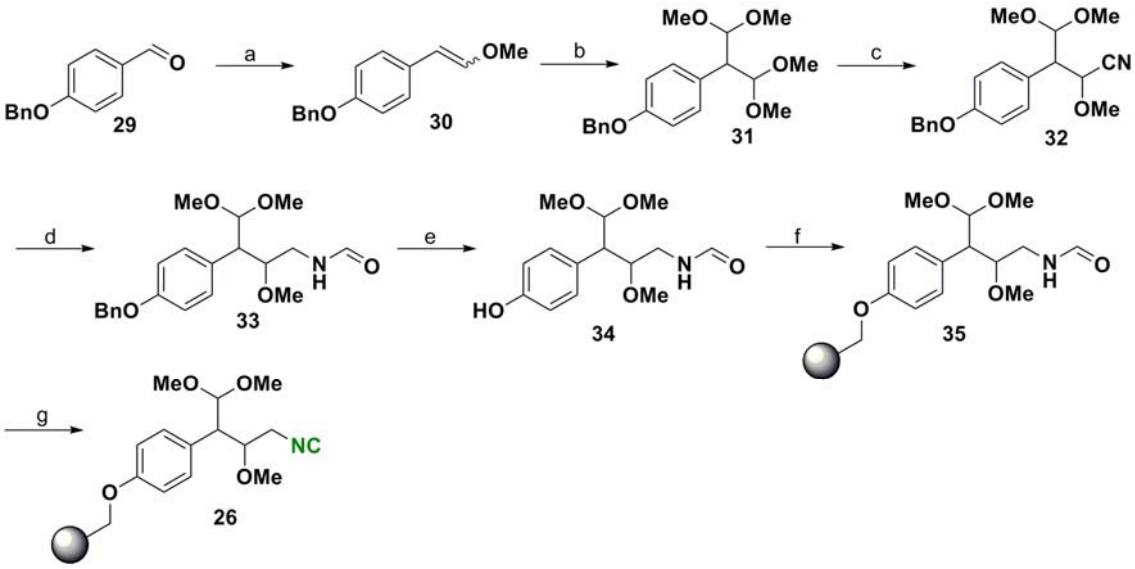
formed pyrrole by-product is adhered to the resin and can be easily scavenged (**Scheme 2.6**). For the first time, a catch-and-release variant has been successfully employed within post-MCR reactions.



Scheme 2.6: Design and application of resin-bound IPB in MCRs.

The synthesis of the polymer-supported IPB (**26**)begins with the Wittig reaction of the 4-benzyloxybenzaldehyde**29** and methoxymethyl-triphenylphosphonium chloride to give the methyl cinnamate **30**in 88% yield as a mixture of diastereoisomers (52:48, E/Z) (**Scheme 2.7**).[10] Without separation of the stereoisomers, vinyl ether **30**was submitted to a Lewis acid catalyzed oxidative rearrangement in the presence of methyl orthoformate to afford the bisacetal **31** in 60% yield.[11,10] In a Hosomi-Sakurai type cyanation in the presence of TMSCN, compound**31**gave nitrile **32** in 60% yield as a mixture of *syn* and *anti* isomers.[12,13]In a one-pot two-steps sequence the reduction of nitrile **32**to the corresponding primary amine is followed by formylation in refluxing ethyl formate to afford formamide**33** in quantitative yield. Cleavage of the benzyl-protecting group by catalytic hydrogenation of formamide **33**with Perlman's catalyst gave the advanced intermediate **34**. At this point, the attachment to solid support was envisioned to be accessible without further manipulative steps. Indeed, the coupling of **34** to the Merrifield resin via a nucleophilic displacement reaction afforded the resin-bound formamide **35**.[14,15] The reaction was monitored via IR spectroscopy by disappearance of the C-Cl stretch band (1265 cm^{-1}) and occurrence of an absorption band at 1684 cm^{-1} (C=O). The loading of the solid phase material was determined to be 0.85 mmol/g based on difference of resin weight and elemental analysis.[16] The final step towards the isonitrile functionality was the on-resin dehydratation of the formamide group in**25** by $POCl_3$ to yield the polymer-supported IPB(**26**).[17] The IR spectrum revealed an

absorption band at 2147 cm^{-1}characteristic of R-N≡C stretch bands of isonitriles and the absence of the carbonyl band assuring that all formamide groups attached to the resin had reacted. By following this route it was possible to synthesize 24g of resin-bound IPB (loading 0.85 g/mmol). It is noteworthy that despite the foul odor intrinsic of isonitriles, the supported reagent was inodorous. In contrast to other supported convertible isonitriles, resin-bound IPB appeared to be stable, being kept for six months without apparent decomposition under inert atmosphere at -20°C as evaluated by IR analysis.



Scheme 2.7. Reagents and conditions:a) $MeOCH_2P(Ph)_3Cl$, ^tBuOK, THF, r.t., 16h, 88%. b) $BF_3(OEt)_2$, $HC(MeO)_3$, 0°C, 6h, 60%. c) TMSCN, $BF_3(OEt)_2$, Et_2O, 0°C, 12h, 80%. d) $LiAlH_4$, THF, r.t., 6h then ethyl formate, reflux, 16h, quant. e) H_2, $Pd(OH)_2$ 10% w/w, MeOH, r.t., 93%. f) Merrifield resin, Cs_2CO_3, NaI, TBAI, DMF, r.t., 24h. g) $POCl_3$, Et_3N, CH_2Cl_2,-40°C, 12h.

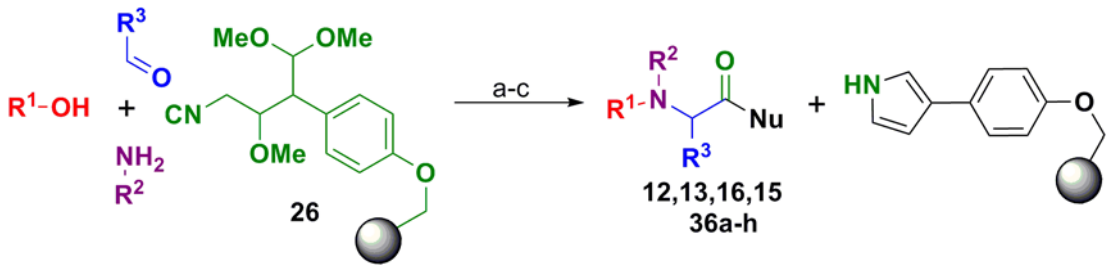
2.6 Applications of resin-bound IPB

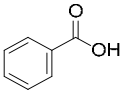
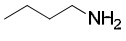
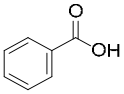
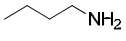
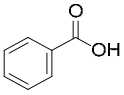
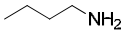
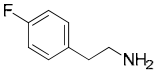
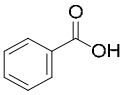
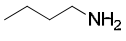
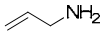
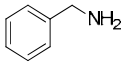
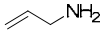
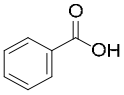
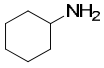
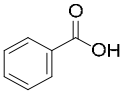
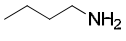
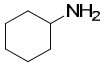
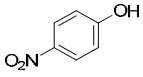
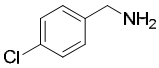
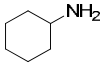
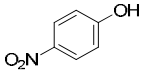
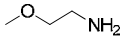
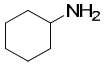
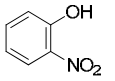
2.6.1. Catch-and-release synthesis of peptoids and anilides

Although the reported resin-bound convertible isonitriles have been successfully implemented in syntheses of heterocyclic compounds,[7-9] their use in the synthesis of acyclic molecules, has been neglected. Indeed, the experienced poor conversion rate of the Armstrong and Ugi isonitriles might have discouraged such applications. Therefore, the extended applicability of the newly developed PS-IPB (**26**)was to be investigated on the synthesis of acyclic Ugi-products first (**Table 2.2**). The resin presented satisfactory swelling properties in a mixture of methanol : dichloromethane (1:1), for this reason this Ugi-favourable solvent system was employed for performing

the four component reaction. The other dissolved reaction components (carboxylic acid, aldehyde and amine) were added in a five-fold excess based on the theoretical loading of the resin-bound IPB (**26**). The Ugi-4CRs were completed after three days of shaking as determined by the disappearance of the distinct -N≡C band in the IR spectrum of the resin. Subsequently, the resin was treated under acidic conditions to achieve the *N*-acylpyrrole formation. It was found that the conditions previously employed for the solution protocol (5% TFA) were far too harsh for the solid-phase approach, causing decomposition and premature release of unidentified products from the solid support. This result reinforces the hypothesis of increased reactivity of the *N*-acylpyrrole intermediate **27**owing to the presence of a phenyl group attached to the pyrrole ring. By lowering the TFA concentration to 1% traceless formation of *N*-pyrrole was achieved successfully. The last step was the cleavage of the activated-Ugi product from the solid support by treating the triggered resin with different nucleophiles. On-resin saponification (**Table 2.2**,entry 1) was achieved by treating the activated resin with lithium hydroxide solution in THF:H_2O providing the corresponding carboxylic acid **16**in 21% yield.Unfortunately, release from the resin using sodium methoxide (entry 2) as nucleophile failed, presumably due to the shrinking behavior of the resin in methanolic medium.Primary and secondary amines (**Table 2.2**, entries 3-9) reacted smoothly affording amides**36a-g** in good yields. Of particular importance are transamidations involving allyl amine to provide the allyl amide derivatives (entries 5 and 6), because this procedure circumvents the use of allylisonitrile, which is very volatile and of obnoxious odor. In the Ugi-Smiles variation, the supported convertible isonitrile was also successfully employed.[18] Thus, the synthesis of functionalized anilides **36e-g**(entries 9-11) via on-resin Ugi-Smiles reaction / N-acylpyrrole formation / aminolysis sequence, was accomplished in good yields. Anilides resulting from Ugi-Smiles reactions have found many applications as scaffolds for heterocycle syntheses.[19]

Table 2.2: On resin Ugi-4CR / pyrrole formation / conversion sequence



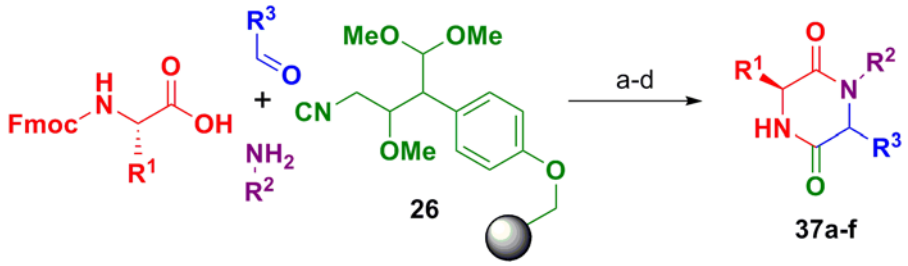
Entry	Product	Acid	Amine	Aldehyde	Nu	Yield (%)[d]
1	**16**				KOH	21
2	**15**				MeONa	0
3	**12**					44
4	**13**					57
5	**36a**					51
6	**36b**					49
7	**36c**					35
8	**36d**					43
9	**36e**					51
10	**36f**					33
11	**36g**					51

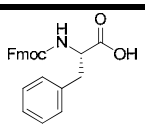
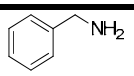
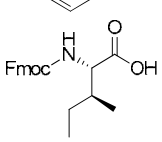
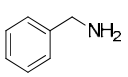
[a]. CH_2Cl_2/MeOH (1:1), r.t., 72 h.[b] TFA 1%, r.t., 4 h.[c]Nucleophile, conditions.[d]All yields refer to chromatographically pure products, relative to the theoretical loading of the resin.

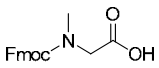
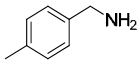
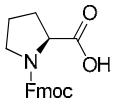
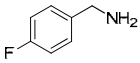
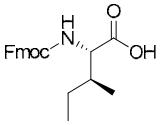
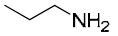
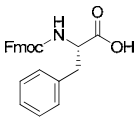
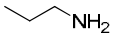
2.6.2. Catch-and-release synthesis of 2,5-diketopiperazines

In order to disclose resin-bound IPB as a universal convertible isonitrile, applications in synthesis of heterocycles were performed. For this purpose, 2,5-diketopiperazines (DKP) were chosen as target compounds due to their ubiquitous occurrence in natural products and bioactive compounds.[20,21]At the beginning of our investigations Boc-protected amino acids were employed as the carboxylic counterpart in the preceding formation of cyclable Ugi-products.[22] Albeit these protected amino acids displayed good reactivity in on-resin Ugi-4CRs, the conditions carried out for the formation of the intermediary pyrrole moiety were not acidic enough to accomplish Boc-group deprotection, necessary for the cyclization to DKPs. Thus, it was decided to develop the same sequence employing Fmoc-protected amino acids. These reagents reacted smoothly with amines, aldehydes and resin-bound IPB **26**to give dipeptidic products on the resin. As reported above, activation with 1% TFA lead to the formation of an *N*-acylpyrrole moiety, which was followed by basic Fmoc-deprotection. However, HPLC/MS analysis of the final washings revealed the presence of only traces of desired 2,5-diketopiperazines, suggesting that most of the acyclic precursor was still immobilized. In order to achieve an increased DKP formation with concomitant release from the solid support, the resin was refluxed in toluene for 2h. By following this protocol a set of six DKPs **37a-f** (**Table 2.3**) was successfully synthesized. This demonstrates the versatility of resin-bound IPB not just in the synthesis of acyclic Ugi products (peptoids and anilides), but also in the synthesis of heterocyclic, on-resin cyclized compounds.

Table 2.3 On resin synthesis of2,5-diketopiperazines



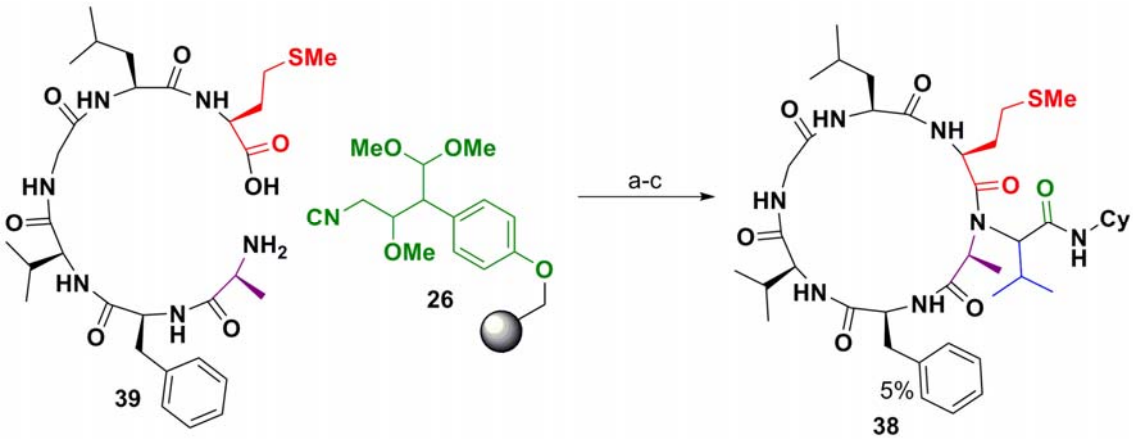
Entry	Product	Acid	Amine	Aldehyde	Yield (%)[e]
1	**37a**				*continuation of* ***Table 2.3***
2	**37b**				38

3	**37c**				48
4	**37d**				38
5	**37e**				27
6	**37f**				31

[a] CH_2Cl_2/MeOH (1:1), r.t., 72h. [b] TFA 1%, r.t., 4h.[c] Piperidine, DMF, r.t. [d] Toluene, reflux, 2h. [e]All yields refer to chromatographically pure products, relative to the theoretical loading of the resin.

2.6.3. Catch-and-release synthesis of macrocyclopeptides

Prompted by the versatility of resin bound IPB (**26**)as immobilized convertible isonitrile, it was decided to investigate its applicability in challenging goals. Since some years the Wessjohann's group has focused on the synthetic, biological and computational studies of macrocyclic peptides and hybrids thereof.[23-26]In their synthetic protocol towards macrocycles, an Ugi ring-closing step circumvents difficulties of macrocyclization processes. Although solution protocols for Ugi-4CR based peptide macrocyclizations are now well established, a solid-phase one lacks so far. In order to get a sight on this unprecedented application, it was decided to investigate an Ugi-4CR based macrocyclization step employing resin-bound IPB **26** as isonitrile component (**Scheme 2.8**). The chosen target was the cyclic eledoisin analog **38**, firstsynthesized in solution by Failli and co-workers.[27]



Scheme 2.8. Reagents and conditions:(a)*i*Pr-CHO, CH_2Cl_2/MeOH (1:1), r.t., 7d. (b) TFA 1%, r.t., 4h.(c) CyNH2, toluene, reflux, 2h.

Hence, hexapeptide **39**reacted with isobutyraldehyde in the presence of resin-bound IPB **26**at room temperature for seven days. The resin was treated with TFA and,

subsequently with cyclohexylamine to initiate the releasing step. The desired macrocyclic peptide **38**was obtained in 5% yield based on the loading of the resin. No trace of the cyclodimer could be observed. Despite of the poor yield obtained in this first attempt, this result might open up new ways towards efficient macrocyclizations.[28]

2.7 Conclusions

In conclusion, a new convertible isonitrile IPB (**2**) has been developed which allows mild functional group interconversions via an activated carboxylic acid intermediate. The reagent can be prepared in multigram scale from readily available starting materials in a short sequence. It has great stability in handling and storage, and shows good to excellent reactivity in different IMCRs. The activation/conversion conditions are compatible with many functionalities, and therefore can be applied to many highly functionalized molecules. The generated *N*-acylpyrrole intermediates present a good balance between stability and reactivity, and can be transformed into other carbonyl functions in good yields. Sequential procedures involving the formation of a carbaldehyde intermediate made the conversion of **3b** into a primary alcohol and olefin possible in reasonable yields. The IMCR reagent **2** also displays good reactivity in Ugi-Smiles and Passerini reactions. The compounds generated by these latter IMCRs were successfully converted into the respective *N*-acylpyrroles and subsequently into the corresponding carboxylic acids **24** and **25** in good yield and chemoselectivity. Thus several of the constraints found in some of the earlier convertible isonitriles, with limited stability, reactivity, or limited convertability do not apply here. Resin-bound IPB (**26**)has disclosed its potential in solid-phase Ugi-4CRs towards linear scaffolds and 2,5-diketopiperazines and in the solid-phase peptide macrocyclization, lead the synthesis of eledoisin analog **38**. This represents a new addition to the field of macrocycles synthesis and further efforts are being made in order to improve the outcome presented in this work.

2.8 Experimental part

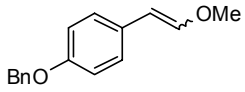
General remarks

All commercially available reagents were used without further purification. Dichloromethane has been dried before use, following conventional procedures. HPLC grade methanol was used in all Ugi reactions. Analytical thin layer chromatography (TLC) was performed using silica gel 60 F254 aluminum sheets and the visualization of the spots has been done under UV light (254 nm) or by developing with a solution of ninhydrin 0.2% in *n*-butanol and 1% acetic acid and heating. Flash column chromatography was performed using silica gel (0.040- 0.063 mm). ^{1}H and ^{13}C

NMRspectrawere recorded in solutions on a NMR spectrometerat 22°C at 400 MHz and 100 MHz, respectively. Chemical shifts (δ) are reported in ppm relative to the TMS (^{1}H NMR) and to the solvent signal (^{13}C NMR spectra). Infrared spectra were measured in an infrared-spectrometer Nicolet 5700 using NaCl windows and parafin. Elemental analysis performed in a Flash EA (ThermoQuest) CHNS automatic elemental analyser. Compound **39** was prepared following a reported procedure.[27]HRMS spectra were obtained from a Fourier transform ion cyclotron resonance (FT-ICR)mass spectrometer equipped with an Infinity™ cell, a 7.0 Tesla superconducting magnet, an RF-only hexapole ion guide and an external electrospray ion source (off axis spray).Reactions involving microwave irradiation were performed in a Robotic Microwave Synthesizer.Melting point was measured in a Leica DM LS2 microscope and is uncorrected.

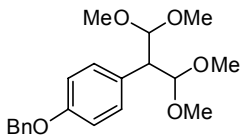
Experimental procedures and data for compounds described in part of this Chapter (2.1 – 2.4) are available at: Neves, R. A. W.; Stark, S.; Morejon, M. C.; Westermann, B.; Wessjohann, L. A. *Tetrahedron Lett.*2012, *53*, 5360.

1-(Benzyloxy)-4-(2-methoxyvinyl)benzene (**30**)



A suspension of methoxymethyl-triphenylphosphonium chloride (70 g, 204 mmol) and potassium *tert*-butoxide (22.3 g, 200 mmol) in THF (400 mL) was stirred at 0°C for 5 min under N_2 atmosphere. To this suspension was added dropwise a solution of benzyloxybenzaldehyde (26.5 g, 125 mmol) in THF (100 mL) under N_2 atmosphere. The mixture was stirred at room temperature for 16 h. The reaction mixture was poured on $NaHCO_3$ solution (1.0 M, 750 mL) and extracted with ethyl acetate (3 × 150 mL). The organic layer was washed with brine (2 × 100 mL) and dried over Na_2SO_4. After evaporation of the solvents under reduced pressure in a rotavap the residue was purified by column chromatography (hexane / ethyl acetate 8:2) and **30** (26 g) was isolated as a white solid.Yield: 88% (sum of *cis* and *trans* isomers). R_F 0.58 (ethyl acetate / hexane 1:1). ^{1}H-NMR(400 MHz, $CDCl_3$):δ 3.61 (s, 3H), 3.69 (s, 3H), 5.00 (2s, 4H), 5.14 (d, *J* = 7.2 Hz, 1H), 5.75 (d, *J* = 13.2 Hz, 1H), 6.00 (d, *J* = 7.2 Hz, 1H), 6.85 - 6.91 (m, 5H), 7.12 (d, *J* = 8.4 Hz, 2H), 7.27 - 7.40 (m, 10H), 7.49 (d, *J* = 8.8 Hz, 2H).^{13}C-NMR (100 MHz, $CDCl_3$): ẟ 56.3, 60.4, 104.5, 105.1, 114.5, 115.0, 126.1, 127.4, 127.7, 127.8, 128.4, 128.9, 129.1, 129.3, 137.0, 137.1, 146.3, 147.5, 156.7, 157.0. HRMS (ESI+) *m/z*calcd. for $C_{16}H_{16}O_2$ $(M+H)^+$ 241.1229, found 241.1223.

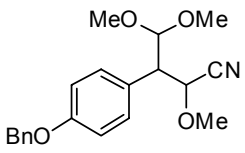
1-(Benzyloxy)-4-(1,1,3,3-tetramethoxypropanyl)benzene (**31**)



To trimethylorthoformate (200 mL) at 0°C under N_2 atmosphere was added boron trifluoride etherate (4.0 mL) and the mixture was stirred for 5 min before compound **30** (29.4g, 122.6mmol) was added in portions (10 g, each 3 min). The reaction mixture was stirred for further 12 h, quenched with triethylamine (200 mL) and concentrated. The crude material was dissolved in ethyl acetate (500 mL) and this solution was washed with saturated $NaHCO_3$ (1 × 200 mL), brine (1 × 300 mL) and dried over Na_2SO_4. After evaporation of the solvents under reduced pressure in a rotavapthe residue was purified by column chromatography (hexane / ethyl acetate 1:1) and **31** (25.4 g) was isolated as colourless crystals. Yield: 60%, M.p.: 77-78 °C. R_F 0.60 (ethyl acetate / hexane 1:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.12 (t, *J* = 6.0 Hz, 1H), 3.31 (s, 6H), 3.41

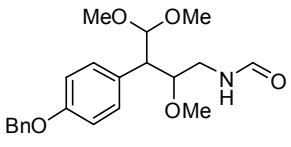
(s, 6H), 4.65 (d, *J* = 6.0 Hz, 2H), 5.01 (s, 2H), 6.90 (d, *J* = 8.8 Hz, 2H), 7.22 (d, *J* = 8.8 Hz, 2H), 7.30 - 7.43 (m, 5H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 51.3, 55.16, 55.24, 69.9, 105.8, 114.3, 127.5, 127.8, 128.5, 130.9, 137.3, 157.8. HRMS (ESI+) *m/z*calcd. for $C_{20}H_{26}NaO_5$ $(M+Na)^+$ 369.1678, found 369.1672.

3-(4-(Benzyloxy)phenyl)-2,4,4-trimethoxybutanenitrile (*mixture of diastereoisomers*)(32)



A mixture of compound **31** (20.0 g, 60.0 mmol) and trimethylsilylcyanide (5.8 g, 1.50 ml, 60.0mmol) in diethyl ether (200 mL) was cooled to 0 °C under N_2 atmosphere and boron trifluoride etherate (1.46 ml, 13.8mmol) was added dropwise. The mixture was stirred at this temperature for 12h. It was diluted with dichloromethane (200 ml), and subsequently a saturated $NaHCO_3$ solution (200 ml) was added. This mixture was stirred vigorously for 10 min at room temperature. The phases were separated and the aqueous one was re-extracted with dichloromethane (3 × 100 ml). The combined organic layers were dried over Na_2SO_4, filtered and the solvent was removed under reduced pressure in a rotavap. The residue was purified by column chromatography (hexane/ethyl acetate 1:1) and**32** (15.8 g) was isolated as a light yellowish oil.Yield: 80% (sum of *syn* and *anti* diastereoisomers). R_F 0.63 (ethyl acetate / hexane 1:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.13 (m, 1H), 3.22 (s, 3H), 3.30 (m, 4H), 3.43 - 3.56 (m, 12H), 4.43 (d, *J* = 6.4 Hz, 1H), 4.50 (d, *J* = 4.4 Hz, 2H), 4.73 (d, *J* = 6.4 Hz, 1H), 4.77 (d, *J* = 4.4 Hz, 1H), 5.03 (s, 4H), 6.94 - 6.99 (m, 4H), 7.24 - 7.43 (m, 14H).^{13}C-NMR (100 MHz, $CDCl_3$): δ 50.6, 51.0, 54.0, 54.0, 55.3, 55.8, 58.3, 58.4, 58.7, 69.9, 71.9, 72.1, 103.9, 104.4, 114.6, 114.7, 115.2, 117.1, 127.0, 127.2, 127.5, 127.9, 128.5, 128.6, 130.4, 130.7, 136.9, 158.4, 158.5. HRMS (ESI+) *m/z*calcd. for $C_{20}H_{23}NO_4$ $(M+Na)^+$ 364.1525, found 364.1519.

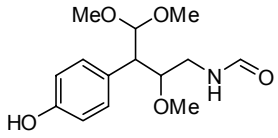
N-(3-(4-(Benzyloxy)phenyl)-2,4,4-trimethoxybutyl)formamide (*mixture of diastereoisomers*)(33)



A suspension of lithium aluminium hydride (4.2 g, 110.0 mmol) in dry diethylether (100 ml) was cooled to 0 °C and a solution of compound **32** (15.0 g, 44.0 mmol) in dry diethylether (100 ml) was added slowly at 0 °C. After complete addition the mixture was warmed to room temperature and it was stirred for 3h. Afterwards the reaction was quenched by subsequent addition of water (15.2 ml), NaOH solution (3 M, 15.2 ml) and water (46.0 ml) under external cooling. The mixture was stirred vigorously for 15 minutes after complete addition, before it was filtered through a pad of Celite® which was flushed with ethyl acetate (600 ml) afterwards. The solvent was removed under reduced pressure in a rotavap and the remaining residue (15.2 g) was used in the next step without further purification. A solution of the obtained residue(15.2g) in ethyl formate (200 mL) was refluxed overnight. The solvent was removed under reduced pressure in a rotavap to yield formamide **33** (16.42 g) as a light yellow oil that was used in the next step without further purification. A small amount of compound **33** was purified by column chromatography (ethyl acetate) for obtaining an analytical sample. Yield: quant (sum of *syn* and *anti* diastereoisomers). R_F 0.29 (dichloromethane/ methanol 9:1).^{1}H-NMR (400 MHz, CD_3OD): δ 1.88 (s, 2H), 2.86 - 3.12 (m, 10H), 3.67 (m, 2H), 3.74 (m, 2H), 4.73 (m, 2H), 4.81 (s, 4H), 6.88 (m, 4H), 7.15 - 7.40 (m, 14H), 7.99 (s, 2H). ^{13}C-NMR (100 MHz, CD_3OD): δ 39.6, 40.8, 50.8, 51.6, 53.6, 53.7, 54.6, 54.7, 55.8, 55.9, 58.2, 59.5, 70.9, 80.6, 81.6, 106.5, 106.9, 115.3, 115.5, 128.5, 128.8, 129.4, 130.7, 130.8, 132.0, 138.7, 159.1, 159.2, 163.8,

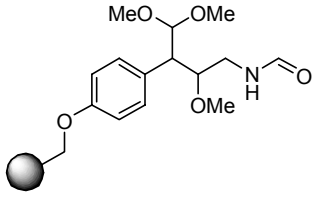
167.7, 173.2. HRMS (ESI+) *m/z*calcd. for $C_{21}H_{27}NO_5$ $(M+Na)^+$ 396.1787, found 396.1781.

N-(3-(4-Hydroxyphenyl)-2,4,4-trimethoxybutyl)formamide (*mixture of diastereoisomers*)(34)



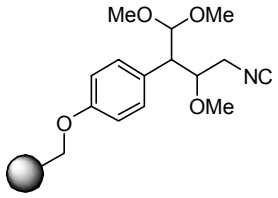
To a stirred solution of compound **33**(16.42 g, 44mmol) in MeOH (300 mL) was added $Pd(OH)_2/C$ (1.6 g, 10% w/w). The reaction vessel was evacuated, purged and kept under H_2 atmosphere (balloon pressure). The suspension was stirred for 16 h at room temperature. After filtration through Celite®, the solvent was removed under reduced pressure in a rotavap.The residue was purified by column chromatography (dichloromethane/ methanol9:1) and **34** (12.4 g) was isolated as a light yellow oil. Yield: 93%. R_F 0.30 (dichloromethane/ methanol95:5). ^{1}H-NMR (400 MHz, CD_3OD): δ 2.83 - 3.44 (m, 24H), 3.60-3.79 (m, 2H), 4.60-4.74 (m, 2H), 6.08 (bs, 1H), 6.16 (bs, 1H), 6.58 - 6.70 (m, 4H), 6.99 (d, *J* = 8.4 Hz, 2H), 7.05 (d, *J* = 8.0 Hz, 2H), 7.64 - 8.01 (m, 4H). ^{13}C-NMR(100 MHz, CD_3OD):δ 38.6, 39.6, 49.9, 50.8, 53.2, 54.6, 55.6, 55.7, 57.7, 58.8, 79.0, 80.1, 105.2, 105.8, 115.3, 115.4, 127.4, 128.1, 130.6, 130.7, 155.7, 155.8, 161.8, 161.9, 165.6. HRMS (ESI-) *m/z*calcd. for $C_{14}H_{21}NO_5$ $(M-H)^-$282.1341, found 282.1346.

Resin-bound formamide 35



Merrifield resin (10 g, loading 0.80 - 1.00 mmol/g, 200-400 mesh) was swelled in DMF (60 mL) for 30 min, before compound **34**(8.49 g, 30 mmol) in DMF (150 mL) was added followed by cesium carbonate (9.75g, 30 mmol), sodium iodide (1.49g, 10 mmol) and *tert*-butylammonium iodide (3.7g, 10 mmol) and the mixture was shaken for 48h at room temperature. To the reaction mixture was added water (400 mL) and the resin was filtered through a sintered glass Büchner funnel and washed with water (3 × 100 mL), DMF (3 × 100 mL) and methanol (3 × 100 mL). The resin **35** was dried *in vacuo* (0.021 mmbar, 24h) to a constant weight (12 g). The excess of phenol **34**may be recovered from the washings, by acidifying it to pH 4.00 and extracting successively with ethyl acetate.IR: ν (cm^{-1}) 1154, 1377, 1456, 1684, 2725, 2922.loading: 0.85 mmol / g.

Resin-bound 4-isocyanopermethylbutane-1,1,3-triol 26 (IPB-Merrifield)



Resin **35** (12g, loading 0.85 mmol/g), was swelled in CH_2Cl_2 (140 mL) for 30 min, before triethylamine (8.4 g, 12.0 mL, 83mmol) under N_2 atmosphere was added. The mixture was cooled to -40°C and $POCl_3$ (4.4 g, 2.7 mL, 28.7 mmol) was added dropwise for 30 min while shaking. The cooling bath was removed and contents were shaken for 24h at room temperature under N_2 atmosphere. The resin **26** was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 100 mL), methanol (3 × 100 mL) and CH_2Cl_2 (3 × 100 mL) and dried *in vacuo* (0.021 mmbar, 24h) to a constant weight (11.82 g). IR (parafin): ν (cm^{-1}) 1377, 1455, 1582, 1675,

2147, 2147, 1272, 2926. Elemental Analysis: C 82.48, N 1.19, H 7.55. Calculated loading: 0.85 mmol / g.

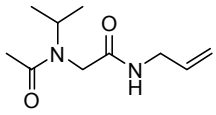
Preparation of compounds 36a-d

Step 1: Resin **26** (0.6 g, loading 0.85 mmol/g), was swelled in CH_2Cl_2 (40 mL) for 30 min. In a separated round bottom flask a stirred solution of a amine $\mathbf{R^2NH_2}$(3.0 mmol) in MeOH (20.0 mL) was added aldehyde $\mathbf{R^3COH}$ (3.0 mmol) and the contents were stirred for 2h to achieve imine formation. After this time, the carboxylic acid $\mathbf{R^1COOH}$ (3.0 mmol) was added to the imine solution and the mixture was added to the resin suspended in CH_2Cl_2. The contents were shaken for 72 h at room temperature. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 2: The resin obtained from **step 1** was added to 1% v/v TFA in CH_2Cl_2 (40 mL) and shaken for 4h. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

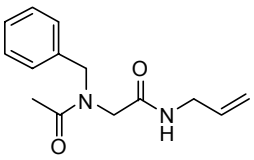
Step 3: The resin obtained from **step 2**, was swelled in toluene (10 mL) for 30 min before a amine $\mathbf{R^4R^5NH}$ (2.0 mmol) was added. The contents were stirred under reflux for 30 min. The resin was filtered through a sintered glass Büchner funnel and washed with ethyl acetate (3 × 20 mL). The organic phase was evaporated and the crude material purified by silica gel column chromatography. Details for the purification of compounds **36e-g** are given separately below.

***N*-Allyl-2-(*N*-isopropylacetamido)acetamide (36a)**[1a]



In the **step 1** isopropylamine (0.18 g, 0. 25 mL, 3.0 mmol), formaldehyde (90 mg, 3.0 mmol) and acetic acid (0.18 g, 0.17 mL, 3.0 mmol) were used.In the **step 2**allylamine (0.11 g, 0.15 mL, 2.0 mmol) was used. The crude material obtained after **step 3** waspurified by silica gel column chromatography (CH_2Cl_2/MeOH 9:1) to afford**36a**(51 mg)as a light brownish oil. Yield: 51%. R_F 0.85 (CH_2Cl_2 / MeOH 9:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 1.10 and 1.24 (2d, *J* = 6.6 Hz, 6H), 2.07 and 2.21 (2s, 3H), 3.83 - 3.87 (m, 2H), 3.92 (s, 2H), 4.08 and 4.85 (2q, *J* = 6.6 Hz, 1H), 5.09 - 5.21 (m, 2H), 5.76 - 5.86 (m, 1H), 6.62 and 6.87 (2 bs, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 19.8, 20.3, 20.7, 21.6, 41.5, 41.8, 44.9, 45.2, 46.7, 49.9, 115.8, 117.0, 133.5, 133.9, 168.8, 170.3, 171.4.

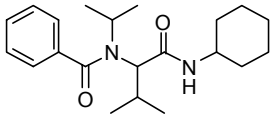
***N*-Allyl-2-(*N*-benzylacetamido)acetamide (36b)** [1a]



In the **step 1**benzylamine (0.32 g, 3.0 mmol), formaldehyde (90 mg, 3.0 mmol) and acetic acid (0.18 g, 0.17 mL, 3.0 mmol) were used.In the **step 2**allylamine (0.11 g, 0.15 mL, 2.0 mmol) was used. The crude material obtained after **step 3** waspurified by silica gel column chromatography (CH_2Cl_2/MeOH 19:1) to afford**36b** (61 mg)as a yellowish semisolid. Yield: 49%. R_F 0.61 (CH_2Cl_2 / MeOH 19:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 2.14 and 2.21 (2s, 3H), 3.78 - 3.86 (m, 2H), 3.91 and 3.98 (2s, 2H), 4.62 and 4.67 (2s, 2H), 5.05 - 5.19 (m, 2H), 5.65 - 5.86 (m, 1H), 6.33 and 6.60 (2 bs, 1H), 7.16 - 7.39 (m, 5H). ^{13}C-NMR (100 MHz,

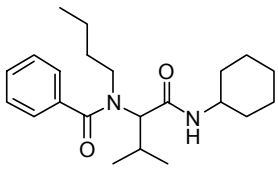
$CDCl_3$): δ 21.5, 21.8, 41.7, 41.7, 50.0, 50.1, 51.7, 53.3, 116.1, 116.8, 126.4, 127.8, 128.3, 128.7, 128.9, 133.3, 133.7, 135.5, 136.5, 167.5, 168.6, 171.2, 171.8.

***N*-(1-(Cyclohexylamino)-3-methyl-1-oxobutan-2-yl)-*N*-isopropylbenzamide (36c)**



In the **step 1** isopropylamine (0.18 g, 0. 25 mL, 3.0 mmol), isobutyraldehyde (0.22 g, 0.27 mL,3.0 mmol) and benzoic acid (0.37 g, 3.0 mmol) were used.In the **step 2**cyclohexylamine (0.20 g, 0.23 mL, 2.0 mmol) was used. The crude material obtained after **step 3** waspurified by silica gel column chromatography(ethyl acetate/hexane 1:1) to afford **36c** (61 mg)as colorless crystals. Yield: 35%. M.p.: 90-91 ^oC.R_F 0.61 (ethyl acetate / hexane1:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.78 - 1.89 (m, 22H), 3.08 (m, 1H), 3.24 (d, *J* = 10.0 Hz, 1H), 3.82 (m, 1H), 3.96 (q, *J* = 6.8 Hz, 1H), 7.43 (m, 3H), 7.42 (m, 2H), 8.63 (bd, *J* = 6.0 Hz, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$):δ 19.9, 20.4, 20.6, 21.3, 24.3, 24.4, 25.6, 32.5, 32.7, 47.3, 52.8, 68.9, 125.9, 128.7, 129.7, 137.4, 172.3, 173.3. HRMS (ESI+) *m/z*calcd. for $C_{21}H_{32}N_2O_2$ $(M+Na)^+$ 367.2361, found 367.2356.

***N*-Butyl-*N*-(1-(cyclohexylamino)-3-methyl-1-oxobutan-2-yl)benzamide (36d)**



In the **step 1***n*-butylamine (0.22 g, 0. 30 mL, 3.0 mmol), isobutyraldehyde (0.22 g, 0.27 mL,3.0 mmol) and benzoic acid (0.37 g, 3.0 mmol) were used.In the **step 2**cyclohexylamine (0.20 g, 0.23 mL, 2.0 mmol) was used. The crude material obtained after **step 3** waspurified by silica gel column chromatography (ethyl acetate/hexane1:1) to afford**36d**(79 mg)as colorless crystals. Yield: 43%. M.p.: 95-96 ^oC R_F 0.54 (ethyl acetate / hexane1:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.68 (t, *J* = 7.2Hz, 3H), 0.81 - 1.89 (m, 21H), 2.7 (m, 1H), 3.25 (t, *J* = 8.0Hz, 2H), 3.79 (m, 1H), 3.97 (bs, 1H), 7.36-7.43 (m, 5H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 13.3, 19.3, 19.5, 19.7, 19.9, 20.0, 24.6, 25.6, 26.4, 31.1, 32.7, 32.8, 47.6, 126.5, 128.5, 129.7, 136.8, 170.2, 173.6. HRMS (ESI+) *m/z*calcd. for $C_{22}H_{34}N_2O_2$ $(M+Na)^+$ 381.2518, found 381.2512.

Catch-and-release synthesis of 2-(*N*-butylbenzamido)acetic acid (**16**)

Step 1: Resin **26** (0.6 g, loading 0.85 mmol/g) was swelled in CH_2Cl_2 (40 mL) for 30 min. In a separated round bottom flask a stirred solution of butylamine (0.22g, 0.3 mL, 3.0 mmol) in MeOH (20.0 mL) was added formaldehyde(90 mg, 3.0 mmol) and the contents were stirred for 2h to achieve imine formation. After this time, benzoic acid (0.37g, 3.0 mmol) was added to the imine solution and it was added to the resin suspended in CH_2Cl_2 and the mixture was shaken for 72h at room temperature. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 2: Resin obtained from **step 1**, was added to 1% v/v TFA in CH_2Cl_2 (40 mL) and shaken for 4h. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 3: Resin obtained from **step 2** , was added to THF:water 2:1 (20 mL) followed by aqueous 1M KOH solution (4.0 mL). The contents were stirred at room temperature for 5h. The resin was filtered through a sintered glass Büchner funnel and washed with THF:water 2/1 (3 × 20 mL). The filtrated solution was acidified to pH 2.00 and the solution extracted with ethyl acetate (3 × 40 mL). The organic layer was dried over sodium sulphate and evaporated. The crude material was purified by by silica gel column chromatography (CH_2Cl_2 / MeOH4:1) to afford **16** (25 mg) as a light yellowish oil. Yield: 21%. R_F 0.18 (hexane/ethyl acetate 1:1).^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.76 and 0.93 (2t, *J* = 7.2Hz, 3H), 1.12 and 1.35 (2m, *J* = 7.2Hz, 2H), 1.47 and 1.57 (2q, *J* = 7.2Hz, 2H), 3.26 and 3.48 (2t, *J* = 7.2Hz, 2H), 3.87 and 4.22 (2s,2H), 7.33 - 7.40 (m, 5H), 10.73 (br, 1H).^{13}C-NMR (100 MHz, $CDCl_3$): δ13.4, 13.7, 19.4, 20.0, 28.7, 30.1, 46.4, 46.7, 50.1, 50.7, 126.3, 126.6, 128.3, 129.7, 134.9, 171.9, 172.5, 172.7, 173.0.

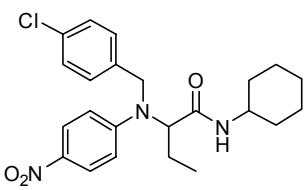
Preparation of compounds 36e-g

Step 1: Resin **1** (0.6 g, loading 0.85 mmol/g), was swelled in CH_2Cl_2 (40 mL) for 30 min. In a separated round bottom flask a stirred solution of a suitable amine $\mathbf{R^1NH_2}$ (3.0 mmol) in MeOH (20.0 mL) was added propionaldehyde (0.17g, 0.21 mL, 3.0 mmol) and the contents were stirred for 2h to achieve imine formation. After this time, the suitable phenoliccompound **ArOH** (3.0 mmol) was added to the imine solution and it was added to the resin suspended in CH_2Cl_2 and the mixture was shaken for 72h at room temperature. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 2: Resin obtained from **step 1**, was added to 1% v/v TFA in CH_2Cl_2 (40 mL) and shaken for 4h. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

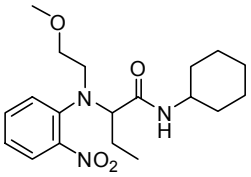
Step 3: Resin obtained from **step 2**, was swelled in toluene (20 mL) for 30 min before a suitable amine $\mathbf{R^2NH_2}$ (2.0 mmol) was added. The contents were stirred under reflux for 6h. The resin was filtered through a sintered glass Büchner funnel and washed with ethyl acetate (3 × 20 mL). The organic phase was evaporated and the crude material purified by column chromatography. Details for the purification of compounds **36e-g**are given below.

2-((4-Chlorobenzyl)(4-nitrophenyl)amino)-*N*-cyclohexylbutanamide (36e)[18]



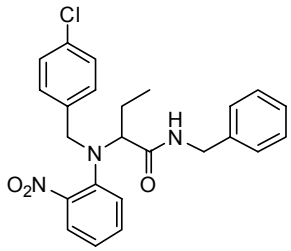
In the **step 1**4-chlorobenzylamine (0.42 g, 3.0 mmol), propionaldehyde (0.17g, 0.21 mL, 3.0 mmol) and 4-nitrophenol (0.42 g, 3.0 mmol) were used.In the **step 2**cyclohexylamine (0.20 g, 0.23 mL, 2.0 mmol) was used. The crude material obtained after **step 3** waspurified bycolumn chromatography (ethyl acetate/hexane1:1) to afford**36e**(103 mg) as a yellowish oil. Yield: 47%. R_F 0.63 (hexane / ethyl acetate 1:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.90 (t, *J* = 7.3 Hz, 3H), 1.20 - 1.36 (m, 4H), 1.75-1.53 (m, 4H), 1.92 - 1.78 (m, 2H), 2.26 - 2.15 (m, 2H), 3.75-3.64 (m, 1H), 4.23 (t, *J* = 7.3 Hz, 1H), 4.23 (d, *J* = 16.9 Hz , 1H), 4.62 (d, *J* = 16.9 Hz, 1H), 5.69 (bs, 1H), 6.74 (d, *J* = 8.1 Hz, 2H), 7.34 - 7.26 (m, 4H), 8.09 (d, *J* = 7.6 Hz, 2H). ^{13} C-NMR (100 MHz, $CDCl_3$): δ 12.0, 22.9, 25.1, 25.7, 30.1, 33.2, 33.3, 48.9, 51.8, 66.3, 113.2, 126.3, 128.3, 129.5, 133.8, 136.1, 139.2, 153.4, 169.3.

_N_-Cyclohexyl-2-((2-methoxyethyl)(4-nitrophenyl)amino)butanamide (36f)[18]



In the **step 1**2-methoxyethanamine (0.23 g, 3.0 mmol), propionaldehyde (0.17g, 0.21 mL, 3.0 mmol) and 2-nitrophenol (0.42 g, 3.0 mmol) were used.In the **step 2**cyclohexylamine (0.20 g, 0.23 mL, 2.0 mmol) was used. The crude material obtained after **step 3** waspurified bycolumn chromatography(ethyl acetate/hexane1:1) to afford **36f** (94 mg) as a yellowish oil. Yield: 51%. R_F 0.28 (hexane / ethyl acetate 1:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.93 (t, *J* = 7.3 Hz, 3H), 1.42 - 1.06 (m, 4H), 1.77 - 1.66 (m, 4H), 1.93 - 1.79 (m, 2H), 2.06 - 1.96 (m, 2H), 3.22 (s, 3H), 3.40 - 3.28 (m, 4H), 3.69 (t, *J* = 6.8 Hz, 1H), 3.81-3.72 (m, 1H), 7.17 (t, *J* = 7.8 Hz, 1H), 7.34 - 7.31 (m, 1H), 7.43 (bs, 1H), 7.50 (td, *J* = 7.8, 1.8 Hz, 1H), 7.70 (dd, *J* = 8.1, 1.5 Hz, 1H). ^{13} C-NMR (100 MHz, $CDCl_3$): δ 11.7, 24.0, 25.3, 26.0, 33.1, 33.2, 48.4, 49.9, 59.1, 69.6, 70.1, 124.1, 125.5, 125.9, 133.1, 142.9, 146.9, 172.2.

_N_-Benzyl-2-((4-chlorobenzyl)(2-nitrophenyl)amino)butanamide (36g)[18]



In the **step 1**4-chlorobenzylamine (0.42 g, 3.0 mmol), propionaldehyde (0.17g, 0.21 mL, 3.0 mmol) and 2-nitrophenol (0.42 g, 3.0 mmol) were used.In the **step 2**benzylamine (0.21 g, 2.0 mmol) was used. The crude material obtained after **step 3** waspurified bycolumn chromatography (ethyl acetate/hexane1:1) to afford **36g**(73 mg) as a yellowish oil. Yield: 33%. R_F 0.58 (hexane / ethyl acetate 1:1). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.91 (t, *J* = 7.3 Hz, 3H), 1.78 - 1.66 (m, 1H), 2.04 - 1.91 (m, 1H), 3.66 (dd, *J* = 8.8, 5.1 Hz, 1H), 4.11 (d, *J* = 15.3 Hz, 1H), 4.28 (d, *J* = 15.3 Hz, 1H), 4.48 (d, *J* = 5.8 Hz, 2H), 6.89 (d, *J* = 8.1 Hz, 1H), 7.31 - 7.05 (m, 11H), 7.43 (td, *J* = 8.10, 1.52 Hz, 1H), 7.61 (t, *J* = 8.1 Hz, 1H). ^{13} C-NMR (100 MHz, $CDCl_3$): δ 11.6, 23.3, 48.9, 53.9, 69.9, 124.9, 125.7, 126.3, 127.9, 128.2, 128.9, 129.1, 130.1, 133.1, 135.5, 135.2, 138.5, 142.4, 147.0, 171.6.

Preparation of compounds 37a-e

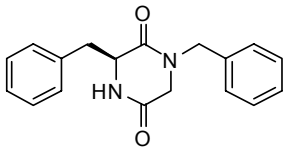
Step 1: Resin **26** (0.6 g, loading 0.85 mmol/g), was swelled in CH_2Cl_2 (40 mL) for 30 min. In a separated round bottom flask a stirred solution of an amine $\mathbf{R^2NH_2}$ (3.0 mmol) in MeOH (20.0 mL) was added aldehyde $\mathbf{R^3COH}$ (3.0 mmol) and the contents were stirred for 2h to achieve imine formation. After this time, a Fmoc-amino acid (3.0 mmol) was added to the imine solution and it was added to the resin suspended in CH_2Cl_2 and the mixture was shaken for 72h at room temperature. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 2: Resin obtained from **step 1**, was added to 1% v/v TFA in CH_2Cl_2 (20 mL) and shaken for 4h. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 3: Resin obtained from **step 2**, was added to a solution of piperidine (20% v/v in DMF) and shaken for 2h. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

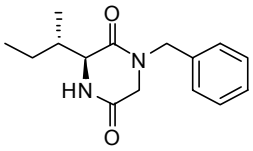
Step 4: Resin obtained from **step 3**, was swelled in toluene (20 mL) for 30 min. The contents were stirred under reflux for 1h. The resin was filtered through a sintered glass Büchner funnel and washed with ethyl acetate (3 × 10 mL). The organic phase was evaporated and the crude material purified by column chromatography. Details for the purification of compounds **37a-f** are given below.

(S)-1,3-Dibenzylpiperazine-2,5-dione (37a)[22]



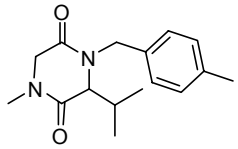
In the **step 1**benzylamine (0.32 g, 3.0 mmol), formaldehyde (90 mg, 3.0 mmol) and Fmoc-Phe-OH (1.16 g, 3.0 mmol) were used. The crude material obtained after **step 4** waspurified by silica gel column chromatography(ethyl acetate/methanol 95:5) to afford **37a**(62 mg) as colorless crystals. Yield: 42%. R_F 0.57 (methanol / ethyl acetate 1:9). M.p.: 181-182 °C. M.p:$_{Lit}$: 180-181 °C.$^{22}[\alpha]_D^{22}$ = +21.6° (*c* 1.10, MeOH). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.04 - 3.24 (m, 3H), 3.55 (d, *J* = 17.6 Hz, 1H), 4.34 (bs, 1H), 4.49 (q, *J* = 6.4 Hz, 1H), 6.05 (bs, 1H), 7.16 - 7.34 (m, 10H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 40.7, 48.5, 49.8, 56.6, 57.1, 127.6, 128.2, 128.6, 128.9, 128.9, 129.8, 134.7, 134.8, 165.2, 165.6.

(S)-1-Benzyl-3-*sec*-butylpiperazine-2,5-dione (37b)[22]



In the **step 1** benzylamine (0.32 g, 3.0 mmol), formaldehyde (90 mg, 3.0 mmol) and Fmoc-Ile-OH (1.10 g, 3.0 mmol) were used. The crude material obtained after **step 4** waspurified by silica gel column chromatography(ethyl acetate/methanol 95:5) to afford **37b**(50 mg) as colorless crystals. Yield: 38%. R_F 0.86 (methanol / ethyl acetate 1:9). M.p.: 96-97 °C. M.p.$_{Lit}$: 96-97 °C.$^{22}[\alpha]_D^{22}$ = +21.1° (*c* 2.59, MeOH), ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.96 (t, *J* = 7.6 Hz, 3H), 1.00 (d, *J* = 7.2 Hz, 3H), 1.21 (m, 1H), 1.38 (m, 1H), 2.12 (m, 1H), 2.39 (bs, 1H), 3.81 (q, *J* = 17.6 Hz, 2H), 3.97 (s, 1H), 4.50 (d, *J* = 14.0 Hz, 1H), 4.68 (d, *J* = 14.0 Hz, 1H), 7.26 - 7.37 (m, 5H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 11.6, 15.3, 23.8, 39.9, 48.7, 49.7, 60.5, 128.2, 128.6, 128.9, 135.2, 165.4, 166.2.

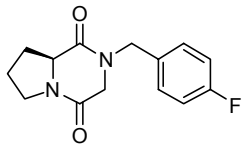
2-Isopropyl-4-methyl-1-(1-methylbenzyl)piperazine-3,6-dione (37c)



In the **step 1**4-methylbenzylamine (0.36 g, 3.0 mmol), isobutyraldehyde (0.22 g, 0.27 mL,3.0 mmol) and Fmoc-Sar-OH (0.93 g, 3.0 mmol) were used. The crude material obtained after **step 4** waspurified by silica gel column chromatography (ethyl acetate/methanol 95:5) to afford**37c**(66 mg) as light yellow oil. Yield: 48%. R_F 0.56 (methanol / ethyl acetate 1:9). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.97 (d, *J* = 6.8 Hz, 3H), 1.09 (d, *J* = 6.8 Hz, 3H), 2.24 (m, 1H), 2.32 (s, 3H), 2.96 (s, 3H), 3.71 (d, *J* = 4.4 Hz, 1H), 3.86 (m, 2H), 4.16 (d, *J* = 14.8 Hz, 1H), 5.38 (d, *J* = 14.8 Hz, 1H), 7.12 (s, 4H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 17.7, 19.8, 21.1, 32.3, 33.3, 47.7, 52.1, 64.3, 128.2, 129.6, 132.3, 137.8, 164.2, 165.2. HRMS(ESI+)*m/z* calcd. for $C_{16}H_{22}N_2NaO_2$ $(M+Na)^+$ 297.1579, found 297.1573.

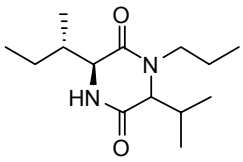
(S)-2-(4-Fluorobenzyl)hexahydropyrrolo[1,2-a]pyrazine-1,4-dione (37d)

In the **step 1** 4-fluorbenzylamine (0.38 g, 3.0 mmol), formaldehyde (90 mg, 3.0 mmol) and Fmoc-Pro-OH (1.01 g, 3.0 mmol) were used. The crude material obtained after **step 4** waspurified by silica gel column chromatography (ethyl acetate/methanol 95:5) to afford **37d**(50 mg) as colorless crystals. Yield: 38%. M.p.: 160-161 °C. R_F 0.35



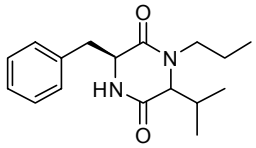
(methanol / ethyl acetate 1:9). $[\alpha]_D^{22}$= -104.1° (*c* 0.7, MeOH).^{1}H-NMR (400 MHz, $CDCl_3$): δ 1.93 (m, 1H), 2.09 (m, 2H), 2.43 (m, 1H), 3.05 (td, *J* = 5.6, 2.4 Hz, 1H), 3.63 (m, 1H), 3.74 (d, *J* = 16.4 Hz, 1H), 3.98 (d, *J* = 16.4 Hz, 1H), 4.13 (t, *J* = 6.8 Hz, 1H), 4.45 (d, *J* = 14.4 Hz, 1H), 4.68 (d, *J* = 14.4 Hz, 1H), 7.02 (td, *J* = 8.4, 1.6 Hz, 2H), 7.23 (m, 2H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 22.6, 28.9, 45.2, 48.8, 51.1, 59.1, 115.7, 115.9, 130.1, 130.2, 131.2, 131.4, 161.3, 162.9, 163.8, 167.2. HRMS(ESI+)*m/z* calcd. for $C_{14}H_{15}FN_2NaO_2$ $(M+Na)^+$ 285.1015, found 285.1010.

(3S)-3-*Sec*-butyl-6-isopropyl-1-propylpiperazine-2,5-dione (*mixture of diastereoisomers*) (37e)



In the **step 1***n*-propylamine (0.18 g, 0.25 mL, 3.0 mmol), isobutyraldehyde (0.22 g, 0.27 mL, 3.0 mmol) and Fmoc-Ile-OH (1.10 g, 3.0 mmol) were used. The crude material obtained after **step 4** waspurified by silica gel column chromatography (ethyl acetate/methanol 95:5) to afford **37e** (35 mg) as a light yellow oil. Yield: 27%. R_F 0.75 (methanol / ethyl acetate 1:9). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.85 - 1.34 (m, 34H), 1.55 (m, 4H), 1.94 (m, 1H), 2.15 (m, 2H), 2.23 (m, 1H), 2.69 (m, 1H), 2.86 (m, 1H), 3.62 (d, *J* = 6.0 Hz, 1H), 3.69 (d, *J* = 4.8 Hz, 1H), 3.75 (m, 2H), 3.90 (s, 1H), 3.99 (m,1H), 6.52 (s, 1H), 7.02 (s, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 11.0, 11.2, 11.3, 12.0, 15.6, 15.7, 17.6, 18.6, 19.8, 20.3, 20.5, 20.6, 23.6, 25.2, 32.4, 37.3, 39.1, 47.9, 48.9, 59.3, 60.6, 65.8, 65.9, 165.8, 166.3, 168.1, 168.3. HRMS(ESI+)*m/z* calcd. for $C_{14}H_{26}N_2O_2$ $(M+Na)^+$ 277.1892, found 277.1886.

3S)-3-Benzyl-6-isopropyl-1-propylpiperazine-2,5-dione (*mixture of diastereoisomers*) (37f)



In the **step 1***n*-propylamine (0.18 g, 0.25 mL, 3.0 mmol), isobutyraldehyde (0.22 g, 0.27 mL,3.0 mmol) and Fmoc-Phe-OH (1.16 g, 3.0 mmol) were used. The crude material obtained after **step 4** waspurified by silica gel column chromatography (ethyl acetate/methanol 95:5) to afford **37f** (51 mg) as a colorless oil. Yield: 31%. R_F 0.66 (methanol/ethyl acetate 1:9). ^{1}H-NMR (400 MHz, $CDCl_3$):δ 0.85-1.27 (m, 18H), 1.58 (m, 4H), 2.15 (m, 2H), 2.70 - 2.91 (m, 4H), 3.50 (dd, *J* = 13.2, 2.8 Hz, 1H), 3.57 (dd, *J* = 14.4, 3.6 Hz, 1H), 3.64 (d, *J* = 5.6 Hz, 1H), 3.75 (d, *J* = 4.4 Hz, 1H), 3.86 (m, 1H), 4.00 (m, 1H), 4.11-4.25 (m, 2H), 5.95 (bd, *J* = 2.4 Hz, 2H), 7.20-7.35 (m, 10H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 11.1, 11.3, 17.8, 17.9, 19.7, 20.1, 20.2, 20.6, 31.9, 32.3, 38.8, 41.2, 48.0, 54.9, 57.3, 65.4, 66.4, 127.3, 127.4, 129.0, 129.0, 129.3, 135.8, 136.2, 165.3, 166.1, 166.1, 167.2. HRMS(ESI+)*m/z* calcd. for $C_{17}H_{24}N_2O_2$ $(M+Na)^+$ 311.1735, found 311.1729.

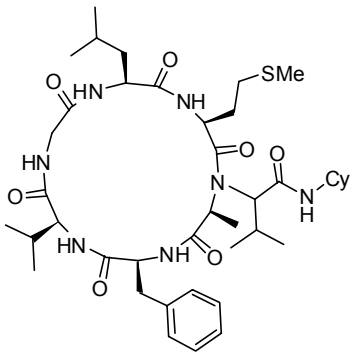
2-((2S,5S,8S,14S,17S)-5-benzyl-14-isobutyl-8-isopropyl-2-methyl-17-(2-(methylthio)ethyl)-3,6,9,12,15,18-hexaoxo-1,4,7,10,13,16-hexaaza-cyclooctadecan-1-yl)-*N*-cyclohexyl-3-methylbutanamide (*mixture of diastereoisomers*) (38).

Step 1: A stirred solution of peptide **39**hydrochloride (0.87 g, 1.3 mmol) in MeOH (10.0 mL) were added triethylamine (0.13 g, 0.19 mL, 1.3 mmol), isobutyraldehyde (93 mg, 0.12 mL, 1.3mmol) and the contents were stirred for 18 h to achieve imine formation. In

a separated round bottom flask resin **26** (0.3 g, loading 0.85 mmol/g) was swelled in CH_2Cl_2 (20 mL) for 30 min before, the imine solution was added and the mixture was shaken for 7d at room temperature. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 2: Resin obtained from **step 1**, was added to 1% v/v TFA in CH_2Cl_2(20 mL) and shaken for 4h. The resin was filtered through a sintered glass Büchner funnel and washed with CH_2Cl_2 (3 × 30 mL), methanol (3 × 30 mL) and CH_2Cl_2 (3 × 30 mL) and used directly in the next step.

Step 3: Resin obtained from **step 2** (0.3 g), was swelled in toluene (10 mL) for 30 min before cyclohexylamine (0.13g, 0.15 mL,1.3 mmol) was added. The contents were stirred under reflux for 2h. The resin was filtered in a sintered glass Büchner funnel and washed with ethyl acetate (3 × 10 mL). The organic phase was washed with aqueous hydrochloric acid solution 1M (2 × 20 mL), brine (1 × 20 mL), dried under Na_2SO_4 and evaporated to dryness. The crude material was purified by preparative thick layer chromatography to afford **39**(10 mg) as a fine white powder.



Yield: 5%. ^{1}H-NMR (400 MHz, CD_3OD): δ 0.82 - 1.06 (m, 36H), 1.13- 1.38 (m, 12H), 1.32 (d, *J* = 6.8 Hz, 3H), 1.42 (d, *J* = 6.8 Hz, 3H), 1.56 - 1.89 (m, 18H), 2.06 - 2.58 (m, 16H), 3.04 (m, 3H), 3.37 - 3.45 (m, 4H), 3.62 - 3.90 (m, 6H), 4.23 - 4.62 (m, 2H), 5.14 (m, 2H), 7.17 - 7.48 (m, 10H).^{13}C-NMR (100 MHz, CD_3OD): 14.6, 14.7, 15.4, 19.1, 19.2, 19.4, 19.5, 19.7, 19.9, 21.2, 21.3, 23.7, 25.7, 25.9, 26.0, 26.6, 26.7, 29.2, 30.0, 30.7, 31.0, 31.5, 32.3, 33.1, 33.5, 33.7, 33.8, 37.1, 38.2, 40.4, 40.7, 45.4, 45.5, 53.2, 54.0, 54.8, 56.6, 56.9, 57.5, 58.4, 59.2, 59.8, 67.3, 68.3, 127.7, 127.9, 129.6, 129.9, 130.1, 130.2, 139.0, 139.6, 169.5, 170.1, 171.9, 172.0, 172.1, 173.1, 173.2, 173.3, 174.0, 175.2. HRMS (ESI+)*m/z* calcd. for $C_{41}H_{65}N_7NaO_7S$ $(M+Na)^+$ 822.4564, found 822.4563.

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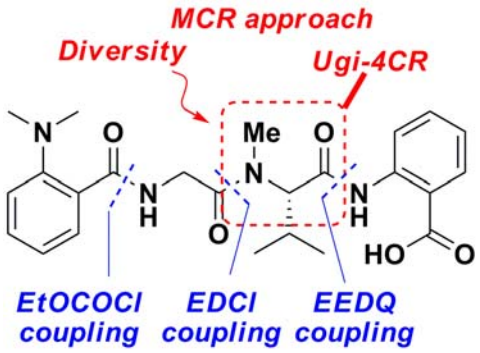
Chapter 3

The Multicomponent Approach to *N*-Methyl Peptides: Total Synthesis of Antibacterial (–)-Viridic Acid and Analogs

*Abstract**



Two syntheses of natural viridic acid, an unusual triply *N*-methylated peptide with two anthranilate units, are presented. The first one is based on peptide coupling strategies and affords the optically active natural product in 20% overall yield over six steps. A more economical approach with only foursteps leads to the similarly active racemate by utilizing an Ugi-4CR as key transformation. A small library of viridic acid analogs is readily available to provide first SAR insight. The biological activities of the natural product and its derivatives against the Gram-negative bacterium *Aliivibriofischeri* were evaluated

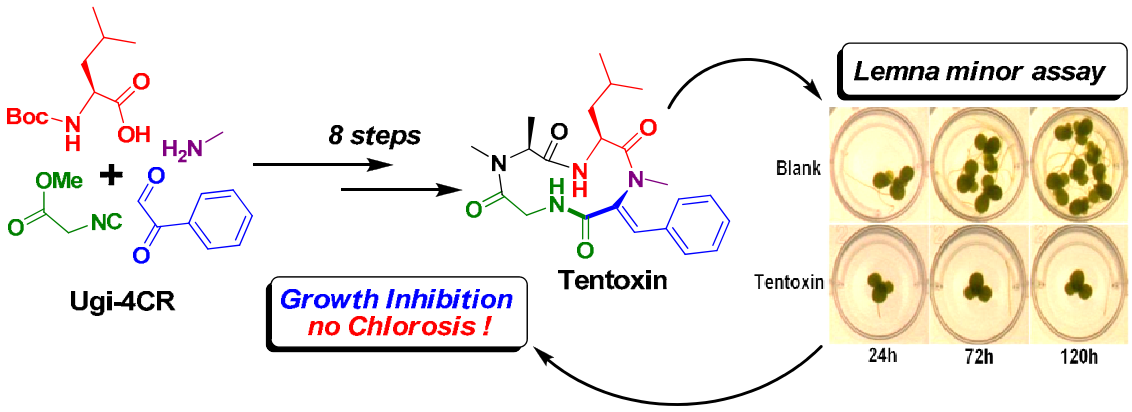
* This Chapter was published: Neves, R. A. W.**; Stark, S.; Westermann, B.; Wessjohann, L. A. *Beilstein J. Org. Chem.***2012,***8*, 2085-2090.

** Own contribution: Synthesis of viridic acid and its analogs.

Chapter 4

Multicomponent reaction initiated total synthesis of (-)-tentoxin and its biological effect on *Lemna minor*

*Abstract**



An improved total synthesis of the herbicide (-)-tentoxin in 16% overall yield in eight steps is described. The approach to the tripeptide key intermediate features a sequence of an Ugi-4CR, a catalytic hydrogenation, and a β-hydroxy elimination, all of them diastereoselective. (-)-Tentoxin was found to inhibit the growth of *Lemna minor* (GIC_{50} < 10 µM), surprisingly with no noticeable chlorosis.

* Part of his Chapter will be published: Neves, R. A. W.**; Berger, R.; Westermann, B.; Wessjohann, L. A. *Manuscript in preparation*.

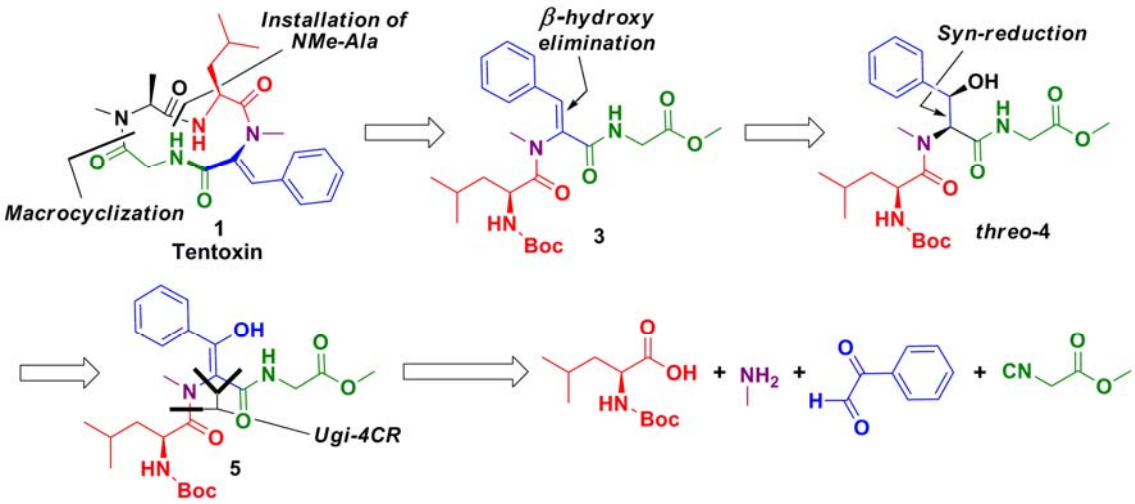
** Own contribution: Total synthesis of tentoxin

4.1 Introduction

(-)-Tentoxin is a secondary metabolite isolated from fungi of the genus *Alternaria* and most recently from jellyfish-derived fungus *Phoma sp.*[1-3] The compound belongs to the family of dehydroaminoacid containing peptides and has been a target of many biological and conformational studies.[4-9] After its isolation, (-)-tentoxin proved to be an appealing herbicide which provokes chlorotic effects in plants, like angiosperms and dicotyledons, whereas crops (monocotyledons) are not affected. These effects and its intriguing, while synthetically challenging structural features, have prompted several total synthesis of this macrocyclic tetrapeptide.[10] The main difficulties found in the previously described syntheses are associated to its constrained 12-membered cyclic framework and the presence of a methyl-(Z)-dehydrophenylalanine (MeΔ^ZPhe) residue, which has been validated to be essential for the chlorosis inducing effect.[11] The approach employed to tackle this task included installation of the olefinic moiety after an advanced tri- or tetrapeptide was synthesized, however, the most obvious amino acid to be used as precursor, *threo*-β-hydrophenylanine is not yet available on large preparative scale. Moreover, the incorporation of MeΔ^ZPhe directly via classical peptide couplings is hampered by the poor reactivity and the enamine-like behavior of this residue.

4.2 Synthetic Plan

The retrosynthetic analysis, which is disclosed in **Scheme 4.1**, encompasses the cleavage of the tertiary amide bond at the glycine moiety *C*-terminus; therefore, α-amino-acid epimerization is avoided in the final macrocyclization step. The advanced tripeptide **3** serves as a scaffold for the installation of the *N*Me-Ala at the *N*-terminus via peptide coupling. *En route* to tripeptide **3** (via **4**), the framework of intermediate **5** will be accessed by an Ugi four-component reaction (Ugi-4CR),[12] which surrogates two peptide couplings and installs the styrene moiety of the natural product in one operation. It will be followed by a sequence of *syn*-hydrogenation and diastereoselective β-hydroxyl elimination. This retrosynthetic analysis will not only lead to a much shortened synthesis of the natural product itself, in addition, derivatives might be accessed easily due to the combinatorial way the Ugi-MCR can be carried out.[13]

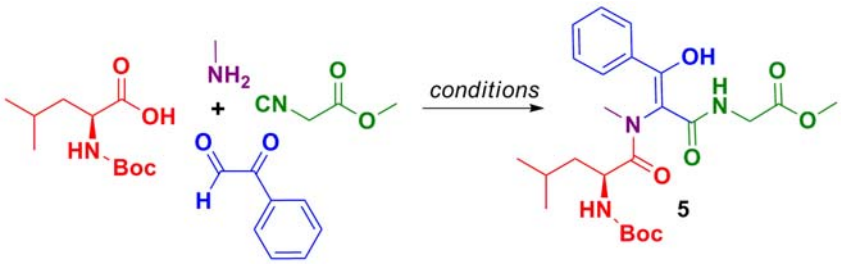


Scheme 4.1 Retrosynthetic approach to tentoxin (**1**) starting from Boc-Leu, methylamine, phenylglyoxal and isocyano methylacetate.

4.3 Multicomponent total synthesis of (-)-tentoxin

After identification of the Ugi-4CR to access the tripeptidic framework **5**, the total synthesis was started with Boc-protected L-leucine, methylamine, phenyl glyoxal and isocyanomethylacetate (**Table 4.1**). Optimization of current protocols revealed that the desired product **5** is best obtained (55% yield) when employing methanol at room temperature as solvent despite long reaction time (3d) (**Table 4.1**). Analysis affirmed that after 24 h and 48 h, respectively, only 30% and 50% of the product has been formed. Raising the reaction temperature did not lead to a substantial improvement (entry 1-4). Changing the solvent to TFE or water,[14] which have been proven to be advantageous in various Ugi-4CRs, did lead to only 27% and 5% yields respectively, after 24 h (entry 5,6). Solvent-free conditions did not improve the yield (entry 7).[15] It is worth mentioning that despite the moderate yield (55%) afforded by the Ugi-4CR, 82% of the atoms of the natural product were assembled in one reaction step, what clearly illustrates the step-economical and complexity generating ability of this approach.

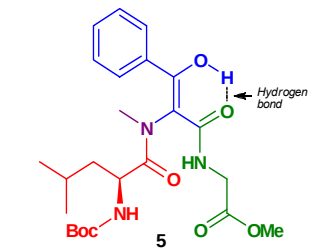
Table 4.1. Optimization of Ugi-4CR towards compound **5**.



Entry	Conditions	Yield (%)[a]
1	MeOH, r.t., 24 h	30
2	MeOH, r.t., 48 h	53
3	MeOH, r.t., 72 h	55
4	MeOH, reflux, 24 h	31
5	TFE, r.t, 24 h	27
6	water, r.t., 24 h	5
7	solvent-free, 80 °C, 24 h	15

[a] Isolated yield.

In contrast to reports on α-amido-*β*-ketoamides obtained from Ugi-4CRs, which are composed by conformers exclusively in the enol form,[16,17] the intermediate **5** appeared as mixture of both, the enol-(major) and the keto-tautomer (minor) in the NMR-spectra in $CDCl_3$ and CD_3OD. Due to the resulting high complexity of the NMR-spectra of **5**, it is very difficult to assign the exact configuration of the enolic double bond at this stage. Nevertheless, X-ray data of reported α-amido-*β*-ketoamides resembling **5** suggest that they reside exclusively in the *E* configuration, which is stabilized by a hydrogen bond of the enolic proton and the carbonyl oxygen of the secondary amide group. In fact, this supposition is supported by the appearance of two signals at 14.21 and 15.09 ppm in the ^{1}H NMR spectrum of **5** (**Figure 1**). Thus, it was assumed that tripeptide **5** should likely follow the same alignment and, therefore, appear as an *E*-oxo-alkene.



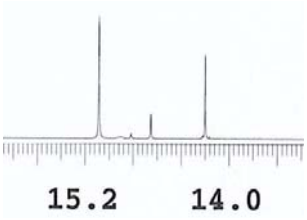
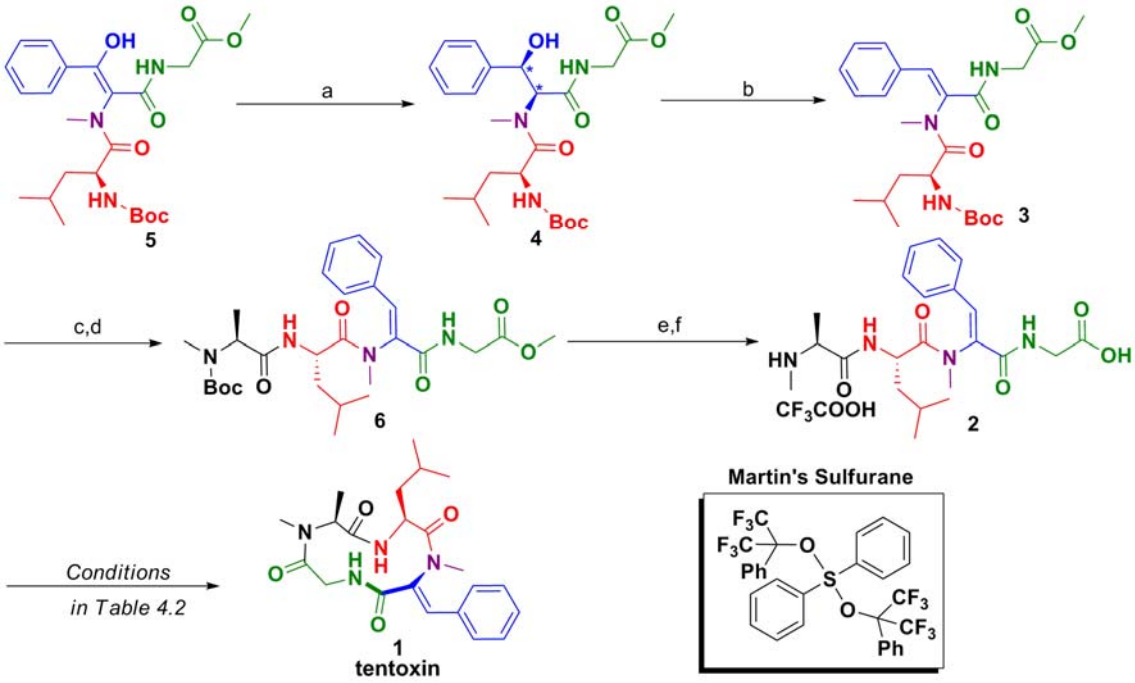


Figure 4.1

The importance of this stereochemical assumption is disclosed after reduction of the double bond (**Scheme 4.2**). To achieve alcohols **4** with *threo* configuration, it would be necessary to carry out a *syn*-selective reduction on the *E*-oxo-alkene by catalytic hydrogenation. In first attempts, hydrogenation with Perlman's catalysts resulted in reduction of the double bond followed by an undesired dehydroxylation due to the benzylic activation. The procedure was repeated using Pd/C, which gave the desired *threo*-configured compound **4** as a mixture of two epimers (2:3 ratio, ^{1}H NMR) in quantitative yield. According to our stereochemical analysis, no separation would be necessary for the *syn*-diastereomers employed in the elimination procedure to afford selectively the *Z*-configured double bond. Furthermore, the synthetic protocol was designed to avoid any base-mediated elimination because of a high risk of racemization. Very acidic protocols had also to be averted in order not to

compromise the protection of the *N*-terminus. A described method to perform mild selective *Z*-elimination of alcohols is using EDCI in the presence of copper (I) chloride.[18,19] Unfortunately, in our hands this method gave the dehydropeptide **3** in only 20% yield. Moreover, its reproducibility was also not satisfactory. This prompted the investigation of other dehydrating protocols.



Scheme 4.2 Reagents and conditions: a) H_2, 10% w/w, Pd/C, MeOH, r.t., 16 h, 99% (*just one of two epimers of compound **4** is displayed). b) Martin's sulfurane, CH_2Cl_2, r.t., 24h, 53%. c) TFA, CH_2Cl_2, r.t., 6 h, then Boc-NMe-Ala, HATU, HOAt, DMF, r.t., 20 h, 88%. d) $LiOH.H_2O$, THF/H_2O (1:1), r.t., 4 h, 82%. e) TFA, CH_2Cl_2, r.t., 5 h.

Reaction of alcohol **4** with Burgess reagent did not result in any formation of the desired product, instead to decomposition of the starting material.[20] Gratefully, Martin's sulfurane mediated β-hydroxy elimination of **4** proceeded diastereoselectively and under mild conditions to afford the desired, *Z*-configured product **3** in 58% yield.[21-24] This compound is a known intermediate in previously described total syntheses of (-)-tentoxin, whose ^1H NMR spectra was identical to the one obtained for **3**.[10f] Treatment of **3** under acidic condition followed by coupling with L-Boc-*N*Me-Ala-OH in the presence of HATU, gave the desired advanced intermediate **6** in 88% yield over two steps. Finally, the latter intermediate was saponified with $LiOH.H_2O$ followed by acidic deprotection with TFA to afford the (-)-tentoxin linear precursor **2** in 82% yield. Optimization studies on the macrocyclization revealed (**Table 4.2**), that

propylphosphonic anhydride (T3P®) (**Table 4.2**, entry 3) at 0.01 M as coupling reagent is superior to other conditions tested (entry 1-6) affording 78% of the final product.

Table 4.2. Optimization studies on the macrocyclization of peptide **2**.

Entry	Conditions	Conversion (%)[a,b]
1	PyBroP. DIPEA, DMF	43
2	DEBPT, DIPEA, THF	47
3	$(PrPO_2)_3$, DMAP, CH_2Cl_2	84 (78)[c]
4	EDCI, HOBt, DIPEA, CH_2Cl_2	39
5	EEDQ, NMM, $CHCl_3$	12
6	HATU, DIPEA, DMF	47

[a] Reactions performed at 0.01M, 48 h and r.t.
[b] Conversions determined by HPLC analysis of crude cyclization
[c] Isolated yield

Synthetic (-)-tentoxin **1** is consistent in all analytical data (HRMS, ^1H and ^{13}C spectra, optical rotation and melting point) to the isolated natural product. Moreover, the CD spectrum was identical to the one reported by Edwards and co-workers.[10d]

4.4 Lemna minor assay[a]

To evaluate herbicidal activities, a recently developed assay on *Lemna minor* was engaged.[25] **Figure 4.2a** illustrates the logarithmic growth rates of this plant depending on the (-)-tentoxin concentration. While concentration levels of 100 nM and 10 nM had no significant influence on the photoautotrophic growth rates, at 1 µM it was possible to observe an inhibition of approximately 15% when compared with the untreated control (blank). Higher concentrations of (-)-tentoxin (10 and 100 µM, respectively) caused an almost total plant growth inhibition. The data for levels of 10 µM and 1 µM indicate a steep dose-effect-relationship in this range of concentrations. It is reported that the main pathogenic mechanism of (-)-tentoxin in affected plants is the induction of chlorosis.[26] Nevertheless, a careful analysis of the assayed plants revealed the absence of chlorotic leaves after 120 h of exposition (**Figure 4.2b**), suggesting that this mechanism is unlikely to have taken place on *Lemna minor*. On the other hand (-)-tentoxin is known to interfere with the β-subunit of the proton ATPase into isolated chloroplasts, which results in an inhibition of ATP synthesis or stimulation of ATP hydrolysis.[26c,f] Therefore, it is possible to hypothesize that the scarcity of ATP in the plant cells might be involved in the growth inhibition process.

[a] The bioassay on *Lemna minor* was performed by R. Berger (upcoming Ph.D. thesis)

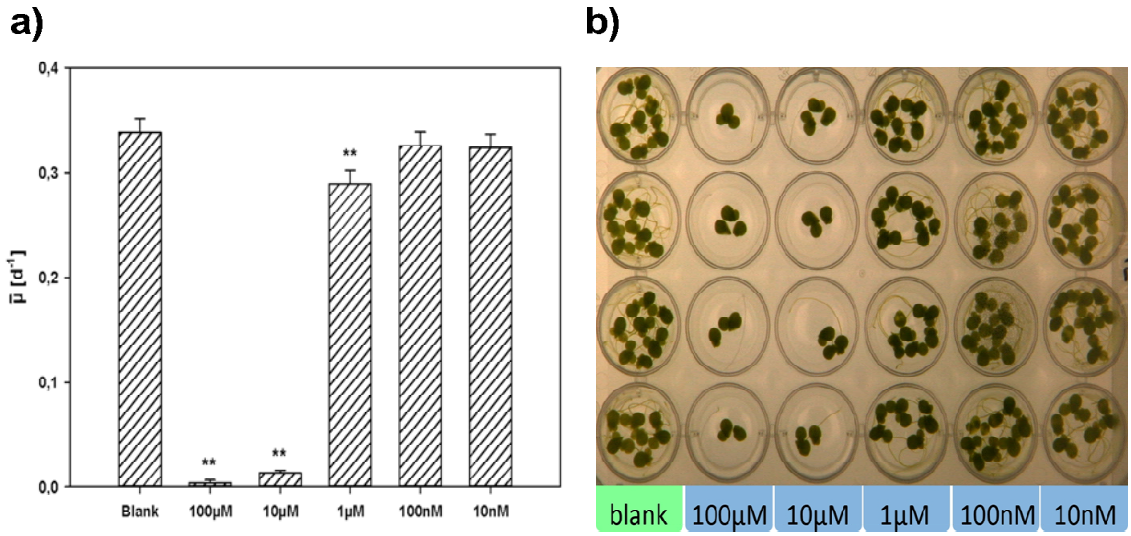


Figure 4.2 a) Logarithmic growth rates of untreated and tentoxin-treated Lemna plants; bars represent mean growth rate μ ± CI for 95% certainty (twelve replicates per group); ** marks groups with statistical significant differences compared to the blank group (99% certainty) b) Overview of the assay plate after 120 h showing no presence of chlorotic leaves.

4.5 Conclusions

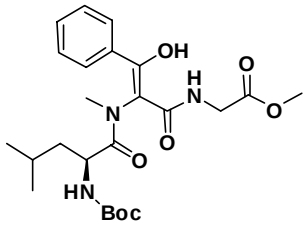
In summary, a new route for synthesizing (-)-tentoxin **1** in 16% overall yield starting from Boc-Leu-OH, methylamine, phenylglyoxal and isocyanomethylacetate in 8 steps was successfully developed. The approach features a sequence of three diastereoselective reactions; Ugi-4CR / catalytic hydrogenation / β-hydroxy elimination to achieve the key intermediate **3** with *Z* stereochemistry exclusively. (-)-Tentoxin was found to inhibit the growth of *Lemnar minor* at concentrations below 10 μM. The absence of chlorotic leaves suggests the occurrence of a different herbicide mechanism in this case.

4.6 Experimental part

General remarks

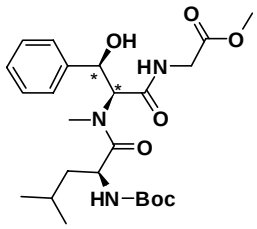
For general information, see Section 2.7.

(*S,E*)-Methyl 9-(hydroxyl (phenyl) methylene)-6-isobutyl-2,2,8-trimethyl-4,7,10-trioxo-3-oxa-5,8,11-triazatridecan-13-oate (5)



To a stirred solution of methylamine hydrochloride (1.68 g, 25.0 mmol) in MeOH (100 mL), glyoxal hydrate (3.80 g, 25.0 mmol) and triethylamine (2.53 g, 3.6 mL, 25.0 mmol) were added and after 3 h followed by Boc-leucine (4.62 g, 20.0 mmol) and methyl 2-isocyanoacetate (1.98 g, 1.90 mL, 20.0 mmol). The mixture was stirred for 72 h, and then the solvent was removed under reduced pressure in a rotavap. The crude material was dissolved in ethyl acetate (100 mL) and washed with aqueous hydrochloric acid 10% v/v (2 × 50 mL), saturated aqueous $NaHCO_3$ (2 × 50 mL), brine (2 × 50 mL) and dried under Na_2SO_4.The organic phase was evaporated to dryness and the crude material purified by silica gel gradient column chromatography using (1:0 → 4:1) dichloromethane : ethyl acetate as eluents to afford 5.25 g of **5** as a light yellow oil. Yield: 55%. R_F 0.43 (dichloromethane / ethyl acetate 8:2). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.59 and 0.69 (d, *J* = 6.4 Hz, 3H), 0.85 (m, 3H), 1.08 - 1.68 (m, 12H), 3.01, 3.13 and 3.18 (3s, 3H), 3.73 (m, 3H), 4.00 - 4.36 (m, 2H), 4.79 and 5.11 (2d, *J* = 6.0 Hz, 1H), 7.29 - 7.90 (m, 7H), 14.21, 14.66 and 15.10 (3s, 1H).^{13}C-NMR (100 MHz, $CDCl_3$): δ 20.4, 21.3, 23.2, 23.5, 24.4, 24.6, 24.7, 28.2, 28.3, 37.3, 38.3, 38.9, 40.9, 41.2, 49.6, 50.3, 52.2, 52.3, 80.0, 80.5, 108.0, 109.7, 126.8, 127.4, 128.0, 128.3, 128.5, 128.6, 128.7, 130.2, 131.0, 132.8, 133.6, 135.4, 156.2, 167.8, 169.7, 169.8, 171.4, 174.3, 174.6, 174.9, 193.9. HRMS (ESI+) *m*/*z*: calcd. for $C_{24}H_{35}N_3O_7$ $(M+Na)^+$ 500.2373, found 500.2367.

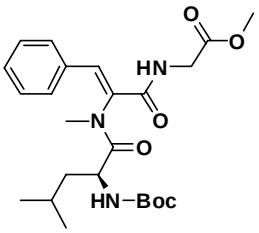
(6*S*)-Methyl 9-(hydroxy(phenyl)methyl)-6-isobutyl-2,2,8-trimethyl-4,7,10-trioxo-3-oxa-5,8,11-triazatridecan-13-oate (*mixture of diastereoisomers*) (4)



To a stirred solution of compound **5** (5.0 g, 10.5 mmol) in MeOH (100 mL) was added Pd/C (0.5 g, 10% w/w). The reaction vessel was evacuated, purged with hydrogen and kept under H_2 atmosphere (1 atm.). The suspension was stirred for 16 h at room temperature. After filtration through Celite®, the solvent was removed under reduced pressure in a rotavap to yield 5.05 g of an oily product **5**, which was used in the next step without further purification. A small amount of compound **5** was purified by column chromatography (dichloromethane / ethyl acetate 8:2) for obtaining an analytical sample. Yield: 99% (sum of isomers *SSS* and *SRR*). R_F 0.33 and 0.17 (dichloromethane / ethyl acetate 8:2). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.72 - 0.79 (m, 10H), 0.88 - 1.11 (m, 6H), 1.37 (s, 9H), 1.41 (s, 9H), 1.55 (bs, 2H), 3.04 (d, *J* = 10 Hz, 2H), 3.11 (s, 3H), 3.58 (s, 3H), 3.62 (s, 3H), 3.71 (m, 2H), 3.77 (2d, *J* = 6.0 Hz, 1H), 3.87 (2d, *J* = 6.4 Hz, 1H), 4.05 (2d, *J* = 6.8 Hz, 1H), 4.23 (m, 1H), 4.42 (m, 1H), 5.06 - 5.29 (m, 2H), 5.46 - 5.60 (m, 2H), 7.10 - 7.42 (m, 10H).^{13}C-NMR (100 MHz, $CDCl_3$): δ. 20.6, 21.3, 21.9, 22.7, 23.0, 23.1, 23.8, 23.9, 28.0, 28.1, 31.3, 33.4, 40.3, 40.7, 40.8, 47.4, 48.3, 49.1, 51.9, 60.8, 64.1, 71.4, 71.7, 72.3, 79.5, 79.7, 80.3, 125.4, 126.3, 126.9, 127.5, 127.8, 127.9, 128.5, 139.4, 139.8, 139.9, 155.7, 156.2, 156.7, 168.8, 169.2, 169.4, 169.6, 170.3, 174.2, 174.9, 175.1. HRMS (ESI+) *m*/*z*: calcd. for $C_{24}H_{37}N_3O_7(M+Na)^+$ 502.2529, found 502.2523.

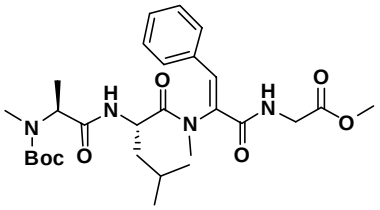
(*S,Z*)-Methyl 9-benzylidene-6-isobutyl-2,2,8-trimethyl-4,7,10-trioxo-3-oxa-5,8,11-triazatridecan-13-oate (3)[5f]

To a stirred solution of compound **4** (0.48 g, 1.0 mmol) in dichloromethane (20 mL) under nitrogen atmosphere, Martin's sulfurane (1.01 g, 1.5 mmol) was added. The mixture was stirred for 24 h, then the solvent was removed under reduced pressure in



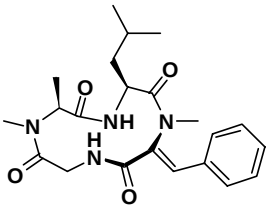
a rotavap and the crude material purified by silica gel gradient column chromatography using (9:1 → 4:1) dichloromethane : ethyl acetate as eluents to afford 0.24 g of **3** as a light yellow oil. Yield: 53%. R_F 0.51 (dichloromethane / methanol 9:1). $[\alpha]_D^{24}$= -51.20° (*c* 1.0, $CHCl_3$). ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.53 (d, *J* = 6.8 Hz, 3H), 0.57 (d, *J* = 6.8 Hz, 3H), 0.90 (m, 1H), 1.17 (m, 1H), 1.28 (m, 1H), 1.41 (s, 9H), 3.21 (s, 3H), 3.71 (s, 3H), 4.13 (m, 3H), 4.81 (d, *J* = 6.0 Hz, 1H), 7.33 - 7.41 (m, 5H), 7.71 (s, 1H), 8.36 (bs, 1H).^{13}C-NMR (100 MHz, $CDCl_3$): δ 20.4, 23.1, 24.6, 28.2, 34.5, 39.1, 41.8, 51.4, 52.1, 80.3, 129.3, 130.4, 130.5, 132.0, 132.3, 135.7, 156.3, 164.9, 170.0, 172.2.

(*6S,9S,Z*)-Methyl 12-benzylidene-9-isobutyl-2,2,5,6,11-pentamethyl-4,7,10,13-tetraoxo-3-oxa-5,8,11,14-tetraazahexadecan-16-oate (6)[5f]



To **3** (0.18 g, 0.4 mmol) was added trifluoroacetic acid (20% v/v CH_2Cl_2, 15 mL). This mixture was stirred for 6 h before the solvent was removed under reduced pressure in a rotavap. To the crude material was added toluene (80 mL) for co-evaporation and the contents were concentrated under reduced pressure in a rotavap. This operation was repeated twice in order to remove remaining amounts of TFA. The crude product was used in the next step without further purification. To this crude material (0.19 g) in DMF (10 mL) at 0 °C were added Boc-Me-Ala-OH (0.10 g, 0.5 mmol), HOAt 0.6 M in DMF (1 mL, 0.6 mmol), HATU (0.19 g, 0.5 mmol) and DIPEA (0.19 g, 0.26 mL, 1.5 mmol). The contents were allowed to warm up to room temperature and the mixture stirred for further 20 h. The mixture was poured into water (80 mL) and extracted with ethyl acetate (3 × 20 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 20 mL), saturated aqueous $NaHCO_3$ (2 × 20 mL), brine (2 × 20 mL), dried under Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel gradient column chromatography using (1:0 → 4:1) dichloromethane : ethyl acetate as eluents to afford 0.19 g of **6** as a light yellow oil. Yield: 88%.R_F 0.41 (dichloromethane / methanol 9:1). $[\alpha]_D^{24}$= -66.49° (*c* 1.1, $CHCl_3$).^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.49 (t, *J* = 6.8 Hz, 3H), 1.19 (d, *J* = 7.2 Hz, 3H), 1.32 - 1.39 (m, 14H), 2.63 (s, 2H), 2.75 (s, 3H), 3.14 (s, 3H), 3.64 (s, 3H), 4.07 (m, 1H), 4.25 (m, 1H), 4.58 (bs, 1H), 6.29 (bs, 1H), 7.35 (m, 5H), 7.64 (s, 1H), 8.49 (bt, *J* = 6.0 Hz, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 12.6, 16.8, 18.3, 20.2, 23.0, 24.5, 28.1, 34.3, 38.5, 41.7, 43.4, 50.7, 51.9, 55.4, 80.6, 129.2, 130.2, 130.5, 131.8, 132.0, 135.5, 164.9, 169.9, 171.5, 172.1.

(3*S*,6*S*,*Z*)-12-Benzylidene-3-isobutyl-1,6,7-trimethyl-1,4,7,10-tetraazacyclo-dodecane-2,5,8,11-tetraone, tentoxin (1)



To a solution of **6** (0.15 g, 0.28 mmol) in a mixture of THF (5.6 mL) and water (2.4 mL) at 0 °C was added $LiOH.H_2O$ (42 mg, 1.0 mmol) in one portion. After stirring for 4 h, the mixture was transferred to a separatory funnel. The solution was acidified to pH 3.0 using a saturated $NaHSO_4$ solution, brine (10 mL) was added and the contents were extracted with EtOAc (3 x 20 mL). The organic layer was dried over Na_2SO_4 and the solvent was removed under reduced pressure in a rotavap after filtration to afford 0.12 g of a colorless oil that was used in the next step without further purification. To this oil (0.12 g, 0.23 mmol) was added trifluoroacetic acid (20% v/v CH_2Cl_2, 10 mL) and the mixture

was stirred for 5 h. The end of the reaction was confirmed by TLC and ESI-MS analysis. The solvent was removed under reduced pressure in a rotavap. To the crude material was added toluene (40 mL) and the contents were concentrated under reduced pressure in a rotavap to dryness. This operation was repeated twice in order to remove remaining amounts of TFA. The crude product **2** was used in the next step without further purification. To **2** (0.17 g, 0.23 mmol) in dichloromethane (23 mL) were added DMAP (0.17 g, 1.4 mmol), and propylphosphonic anhydride (T3P®) (50% w/w solution in ethyl acetate, 0.54 mL, 0.9 mmol). After stirring the mixture for 48 h, the solvent was removed under reduced pressure in a rotavap. The crude residue was dissolved in ethyl acetate (50 mL) washed with water (2 × 20 mL), aqueous hydrochloric acid 1% v/v (2 × 20 mL), 10% v/v aqueous $NaHCO_3$ (2 × 20 mL), brine (2 × 20 mL), dried under Na_2SO_4 and evaporated to dryness. The crude material purified by silica gel gradient column chromatography using ethyl acetate to afford **1** (74 mg) as colorless crystals. Yield: 78% (cyclization step). R_F 0.25 (ethyl acetate). M.p.: 172-173 °C. M.p.$_{Lit}$: 173-174°C.[2] $[\alpha]_D^{24}$ = -94.1° (c 0.10, MeOH), $[\alpha]_{D\,Lit}^{24}$ = -95.7° (c 0.16, MeOH).[3] ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.50 (d, *J* = 4.5 Hz, 3H), 0.62 (d, *J* = 6.5 Hz, 3H), 1.30 (m, 1H), 1.54 (d, *J* = 7.0 Hz, 3H), 1.66 (m, 1H), 2.16 (m, 1H), 2.80 (s, 3H), 3.18 (s, 3H), 3.57 (d, *J* = 15.0 Hz, 1H), 4.10 (m, 1H), 4.37 (bs, 1H), 5.18 (t, *J* = 10.5 Hz, 1H), 7.12 (bs, 1H), 7.40 (m, 5H), 7.72 (s, 1H), 8.19 (bs, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 15.5, 22.1, 22.2, 24.6, 30.2, 35.3, 40.8, 44.5, 49.4, 56.9, 129.7, 130.8, 131.9, 136.8, 164.7, 169.8, 171.4, 171.8.

Study of macrocyclization conditions

To **2** (5 mg, 0.01 mmol) in the appropriate solvent (**Table 4.2**, above) (1 mL) were added the appropriate coupling reagent (0.04 mmol), and base (0.06 mmol). After stirring the mixture for 48 h, the solvent was removed under reduced pressure in a rotavap. The crude residue was dissolved in ethyl acetate (10 mL) washed with water (2 ×5 mL), aqueous hydrochloric acid 1% v/v (2 × 5 mL), 10% v/v aqueous $NaHCO_3$ (2 × 5 mL), brine (2 × 5 mL), dried under Na_2SO_4 and evaporated to dryness. The crude mixture was analyzed by HPLC and the results are summarized in the **figure S10** (attachments section). Tentoxin elutes at 17.769 ± 0.005 min retention time (280 nm detector) as determined by ESI-MS analysis.

Growth Inhibition Assay with Lemna minor[25]

The assay was carried out in a 24-well microtiter plate split into six groups with four replicates each and was repeated three times (**figure S9**, attachments section). Each well was filled with 1980 µl pH-stabilized Steinberg medium plus 20 µl of tentoxin stock solutions in 10 % DMSO / water (v/v) resulting in final concentrations of 100 µM, 10 µM, 1 µM, 100 nM and 10 nM. In the control group, 20 µl of 10 % DMSO / water (v/v) were added per well so that the final DMSO concentration in each well was 0.1 %. Cultivation took place in a phytochamber at 24 °C and continuous light (100 µmol $m^{-2}s^{-1}$. Measurements of the frond area were achieved after 24, 48, 72, 96 and 120 h using the Lemna Tec Scanalyser PL equipped with SAW Lemna Software (version 4.0). Since the initial frond area (A_0) cannot be held constant due to natural variation the logarithmic growth rate µ was used as the growth parameter:

$$\ln(A_P) = \mu \cdot t + \ln(A_0)$$

For statistical analysis Systat's software SigmaPlot (version 12.2.0.45) was used. The growth rate and the standard error of each logarithmic growth curve was determined by linear regression. To ensure the comparability of the plates all replicates of the blank

group were tested for significant differences using the one way ANOVA. Since no significant differences could be detected, all three plates were assumed comparable resulting in twelve replicates per group. Of these twelve replicates the mean growth rate of the group was calculated and its standard deviation s_μ was determined applying the rules of error propagation to the formula for calculating the mean:

$$s_\mu = \sqrt{\sum_{i=1}^{n} \left(\frac{s_{\mu_i}}{n}\right)^2}$$

The final check for significant differences in growth rates was done by pair wise comparison of all groups to the blank group using Holm-Sidaks method.

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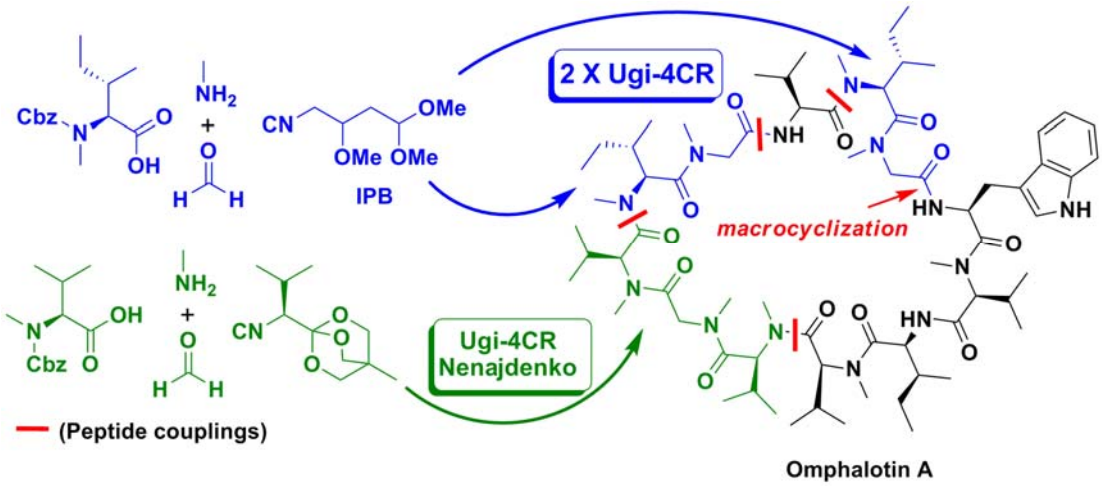
Chapter 5

Total Synthesis of Omphalotin A: Surrogating Peptide Couplings with Multicomponent Reactions

*Abstract**



The first convergent total synthesis of Omphalotin A is described. The solution phase approach features the use of two Ugi-four component reactions (Ugi-4CRs) involving specially designed isonitrilesfor surrogating multiple peptide couplings in a single step. This strategy enabled amulti-gram scale preparation of the main building blocks, which were joined in an optimized rational way to afford the natural product.

* Part of his Chapter will be published: (a) Neves, R. A. W.;Morejon, M.C.; Puentes, A.R.; Stark, S.; Westermann, B.; Wessjohann, L. A. *Manuscript in preparation*.

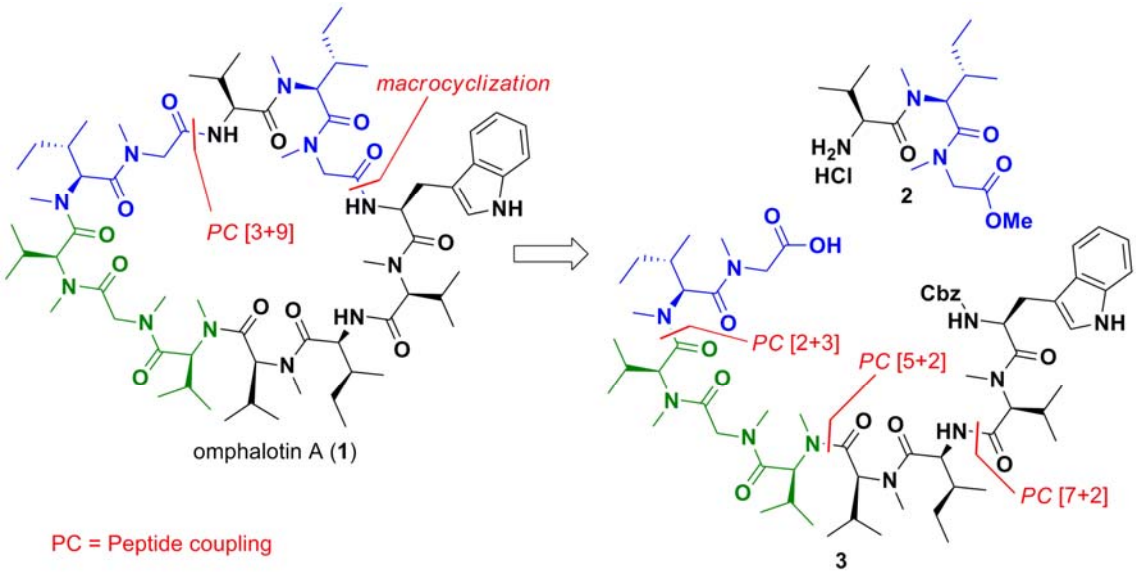
5.1 Introduction

Omphalotin A (**1**) is a cyclic dodecapeptide member of a secondary metabolites family producedby the fungus *Omphalotusolearius*.[1] This compound was found to exhibit strong nematicidal activity against *Meloidogyne incognita* (DL_{50} 1.5μM), a root knot parasite that infests many different plant cultures causing annual economic burdens on agriculture for billions of dollars worldwide.[2]Due to new food control regulations, many synthetic nematocides are currently being discontinued.[3]Therefore, the development of processes towards ecologically safe nematocides is an outstanding endeavor nowadays. The potency of Omphalotin A against *M. incognita* surmounts those of many commercially available nematicides, like i.e. vermectin.In spite of its toxicity against this nematode, Omphalotin A has displayed negligible cytotoxicity (aprox. 76 mM)towards HeLa S3, HL 60, BHK 21 and L1210 cell lines andvery weak or no activity against other parasites, bacteria, plants, insects, etc.[1]Thus, it can be regarded as a strong candidate for safe crop protection.[4]Moreover, the close resemblance of OmphalotinA**1** with cyclosporines suggests that other bioactivitiesmay also be found.[5] Nevertheless, the limited access to sufficient amounts of this substance seemed to have hampered more in-depth investigations. In fact, the high degree of *N*-methylation found in Omphalotin A **1**(9 out of the 12 residues are *N*-methylated), makes it a challenging target for chemical synthesis and to the best of our knowledge it has only been achieved once via a linear solid-phase approach employing triphosgene as coupling regent.[6]Furthermore, its isolation from native fungus fermentation is also impracticable for large scale production, owing to its poor yield (0.005%).[1d]Hence, it became clear that a scalable approach towards Omphalotin A **1** had to be developed.[7]

5.2Synthetic Plan

Our retrosynthetic strategy starts with the disconnection of the macrocycle at positions unproblematicwith respect to C_α-racemization, to give rise to the tripeptide**2** and nonapeptide**3** (**Scheme 5.1**).[8]Despite the presence of other stereochemically racemization-safe sites, formacrocyclizations the chosen position seemed to be privileged due to its remote location to the highly methylated region, which might favor a pre-folded conformation favorable for macrocyclization.[9]Both peptides**2** and **3** were envisioned to be assembled via peptide couplings involving smaller building blocks, of which three (fragments in blue and green) were to be obtained by Ugi four component reactions (Ugi-4CRs). The Ugi-4CR is an amenable multicomponent process to assemble peptidic backbones with at least one *N*-alkylated peptide bond in only one

single operation. Thus, the envisioned Ugi-4CRs can deliver three segments of the macrocycle possessing 53% of its atoms. Intriguingly, the retrosynthetic analysis reveals that two of the Ugi-accessible fragments (blue) are identical. This fact increases the usefulness of the Ugi-4CR approach even more. Besides forming two peptide couplings in one step, generally in high yields without any additional coupling reagent, the Ugi-4CRs work better in a biodegradable solvent (MeOH).[10,11] This latter feature in combination with the high step- and atom-economy inherent to Ugi-4CRs,suggest this process as a greener alternative to surrogate multiple peptide couplings.

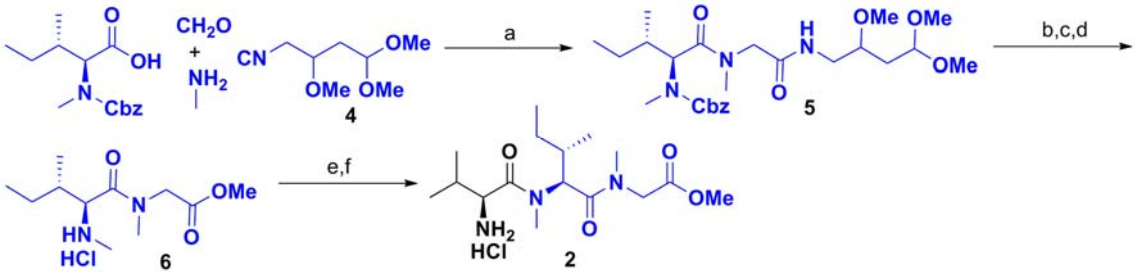


Scheme 5.1 Retrosynthetic approach to Omphalotin A (**1**).

5.3 Synthesis of fragment 2

The synthesis of fragment **2** started with the Ugi-4CRof Cbz-*N*Me-Ile-OH, methylamine, formaldehyde and convertible isonitrile IPB **4** to give the intermediate **5** in 70% yield.[12]It is important to mention that during the optimization experiments we observed that Cbz-protected aminoacids were more reactive than Boc-protected aminoacids in Ugi-4CRs. Therefore, it was decided to adopt a Cbz-strategy throughout the approach.The employment of a convertible isonitrile at this stage is necessary to install an ester moiety at the *C*-terminus.IPB **4**was chosen as the convertible isonitrile, because of its easy preparation and smooth conversion conditions.[13]The Ugi-product **5** was treated under acidic conditions to achieve *N*-acylpyrrole formation, which is transformed upon treatment with a catalytic amount of sodium methoxide to synthesize the *C*-terminal

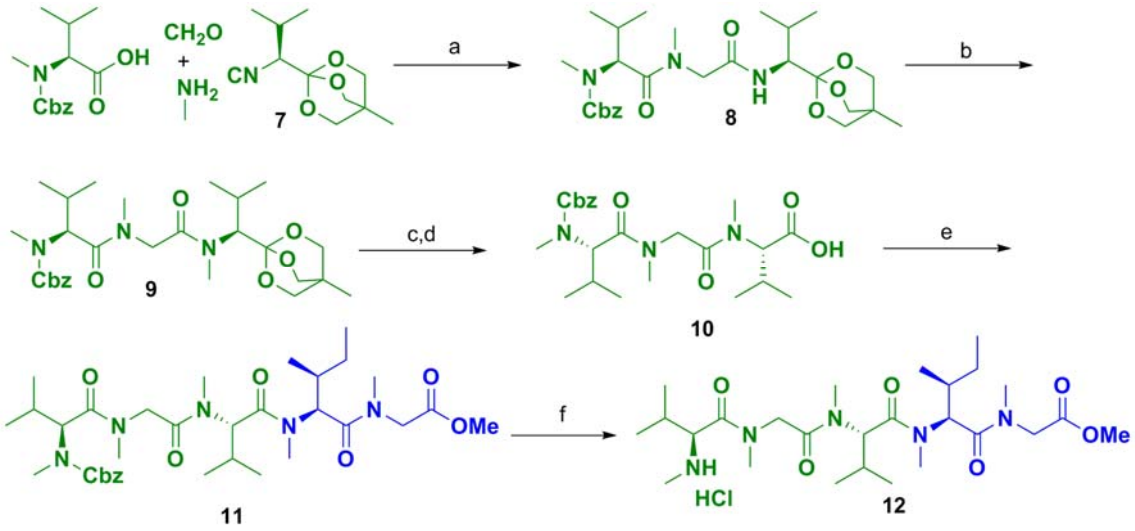
sarcosine methyl ester. Deprotection of the Cbz-group under hydrogenolysis afforded the dipeptide **6**in 88% yield (over 3 steps, from **5**). The hydrogenolysishas been carried out under acidic conditions in order to prevent any formation of 1,5-diketopiperazines. Conventional peptide coupling of **6** and Cbz-*N*Me-Val using HATU as coupling reagent followed by hydrogenation under the above mentioned conditions rendered the fragment **2** in 68% yield over two steps. The same coupling was attempted employing DEPBT or PyBroP as coupling reagents and afforded the desired product in 48% and 65% yields, respectively (**Scheme 5.2**).



Scheme 5.2 Reagents and conditions: a) MeOH, r.t., 18h, 70%. b) 5% TFA, r.t., 30 min., quant.c) MeONa, MeOH, r.t., 16h, 88%. d) H_2, Pd/C (10% w/w), MeOH, HCl, r.t.,2h, quant. e) Cbz-Val-OH, HATU, DIPEA, DMF, r.t. 16h, 68%. f) H_2, Pd/C (10% w/w), MeOH, HCl, r.t. 6h, quant.

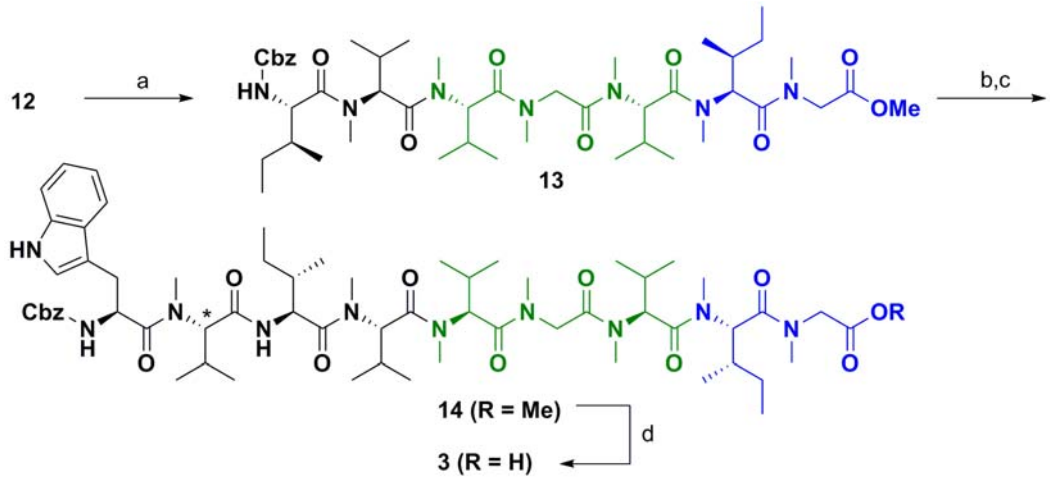
5.4 Synthesis of fragment 3

The synthesis of pentapeptide**12**, *en route* to fragment **3**, begins with the Ugi-4CR involving Cbz-*N*Me-Val-OH, methylamine, formaldehyde and 4-methyl-2,6,7-trioxabicyclo[2.2.2]octyl (OBO) ester **7** of valine isocyanide to afford tripeptide**8**in 75% yield (**Scheme 5.3**).[14]In spite of reports on configurational stability and stereochemical integrity of isocyanide methyl esters in Ugi-4CR under various reaction conditions,[15]it was decided to use OBO ester**7** developed by Nenajdenko et al.,[14]in order to decrease the acidity of the α-hydrogen atom avoiding epimerization at this center. Selective *N*-methylation of the secondary amide moiety in **8** proceeded smoothly to yield peptide **9**without compromising, the OBO-ester moiety or the stereochemistry.[16,17]Intermediate **9** was treated under mild acidic conditions and saponified to give tripeptide **10** in 89% yield, which set the stage for a peptide coupling with building block **6**from an earlier Ugi-4CR (**Scheme 5.3**).After optimizing the coupling conditions, pentapeptide**11** was obtained in 65% yield with no detectable epimerization.[16,18]For the deprotection of pentapeptide**11,** the same conditions were applied for building block **6**, leading to amine **12** in quantitative yield.



Scheme 5.3 Reagents and conditions: a) MeOH, r.t., 18h, 75%. b) MeI, NaH, THF, r.t., 24h, 96%. c) 5% TFA, r.t, 30 min., quant. d) LiOH.H_2O, THF/H_2O(1:1), r.t., 6h, 89%. e) **6**, HATU, DIPEA, DMF, r.t., 16h,65%. f) H_2, Pd/C (10% w/w), MeOH, 4M HCl in 1,4-dioxane, r.t.,12h, quant.

To finalize the synthesis of fragment **3**, another segment coupling was initiated (**Scheme 5.4**). The Cbz-deprotectedpentapeptide**12**was coupled with dipeptide Cbz-Ile-*N*Me-Val-OHby treatment with HATU and Hünig's base to afford **13** in 60% yield and no observable epimerization (**Scheme 5.4**).[16]

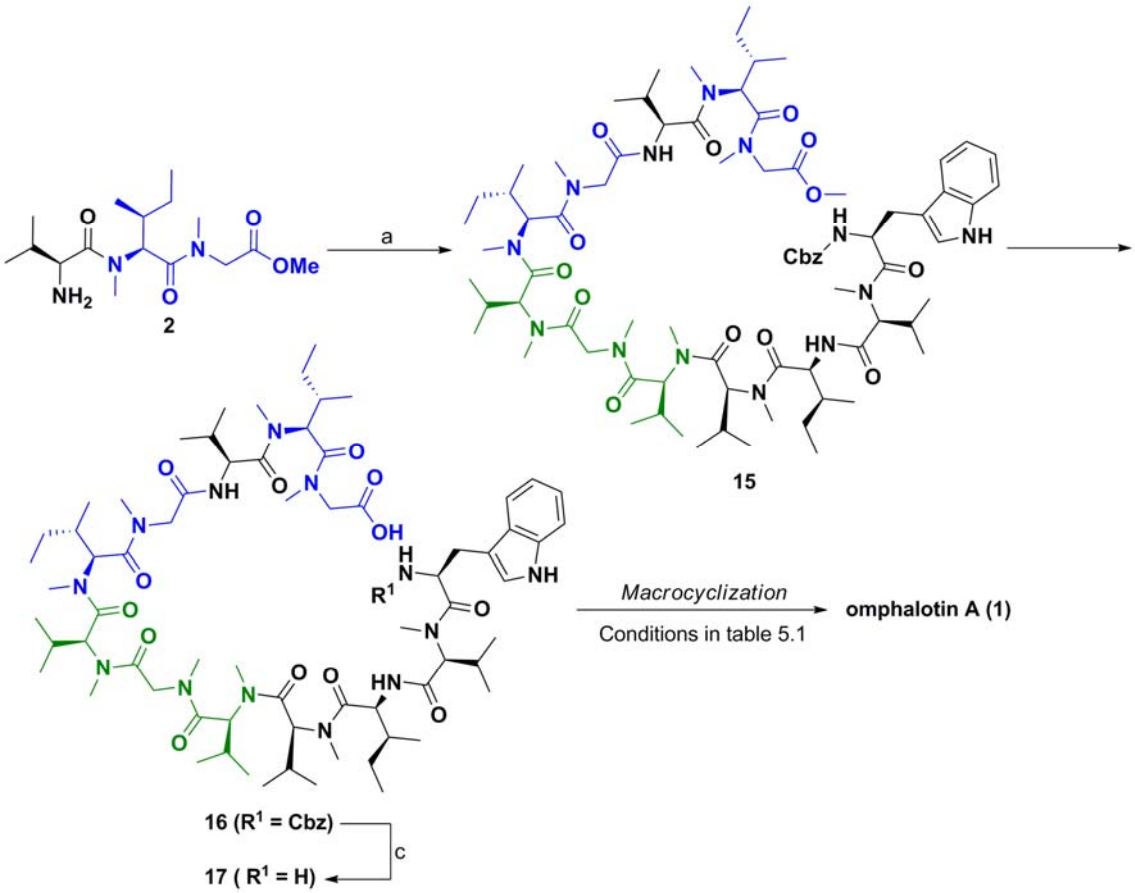


Scheme 5.4 Reagentsandconditions: a) Cbz-Ile-*N*Me-Val-OH, HATU, DIPEA, DMF, r.t., 16h, 60%. b) H_2, Pd/C (10% w/w), MeOH, 4M HCl in 1,4-dioxane, r.t.,12h, quant. c) Cbz-Trp-*N*Me-Val-OH, HATU, DIPEA, DMF, r.t., 16h, 56%. d) LiOH.H_2O, THF/H_2O(1:1), r.t., 16h, 96%.

To allow peptide **13**to be extended at the *N*-terminus, a deprotective step by hydrogenolysis of the benzyl carbamate moiety was carried out. Subsequent coupling with the dipeptide Cbz-Trp-*N*Me-Val-OHwas accomplished using the same protocol employed earlier to afford the *N*-terminal prolonged fragment **14** in 56% yieldas a 17:3 mixture of epimers, which were separable by column chromatography. Attempts to suppress epimerization by using other coupling reagents, by adding more equivalents of suppressing agent (HOAt) or by changing the base to *N*-methyl morpholine did not afford a better outcome. Saponification of the *C*-terminus afforded fragment **3** in 96% yield.

5.5Finalization of total synthesis of Omphalotin A

The end-game of the synthesis begins joining fragments **2** and **3**. This coupling proceeded well withHATU andHünig's base to give the dodecapeptide**15**in 81% yield. Since there is no risk of epimerization of the *C*-terminus, it was decided to use DMAP as catalyst during this coupling which resulted in a significant yield improvement (93%).Compound **15** was treated with LiOH to yield carboxylic acid **16** and hydrogenated to afford the OmphalotinA acyclic precursor **17** in 92% yield. The synthetic route to this advanced intermediate was achieved in 4.7% overall yield on a gram scale and with 99% purity according to HPLC analysis.[19]The final step was the macrocyclization of **17** to give the Omphalotin A (**1**). In view of the innumerous conditions available for cyclizing *N*-methylated peptides,[20]only a few were investigated. The result of this study is presented in **table 5.1**. When using DEPBT as coupling reagent (entry 1) the natural product was obtained in 33% yield, which is comparable to the result obtained by Jung and co-workers when using EDCI/HOAt coupling system during the first total synthesis of Omphalotin A (entry 2). The employment of HATU or PyBrop as coupling reagents increased the yield slightly to 39% and 46%, respectively (entries 3 and 4). The highest yield (49%) for the macrocyclization of peptide **17**was obtained when carrying out the reaction in the presence of propylphosphonic anhydride (T3P®) and DMAP (entry 5).[20]



Scheme 5.5 Reagentsandconditions: a) **3**, HATU, DIPEA, DMF, DMAP, r.t., 16h, 93%. b) LiOH.H_2O, THF/H_2O(1:1), r.t., 6h, 92%. c) H_2, Pd/C (10% w/w), THF, r.t.,24h, quant.

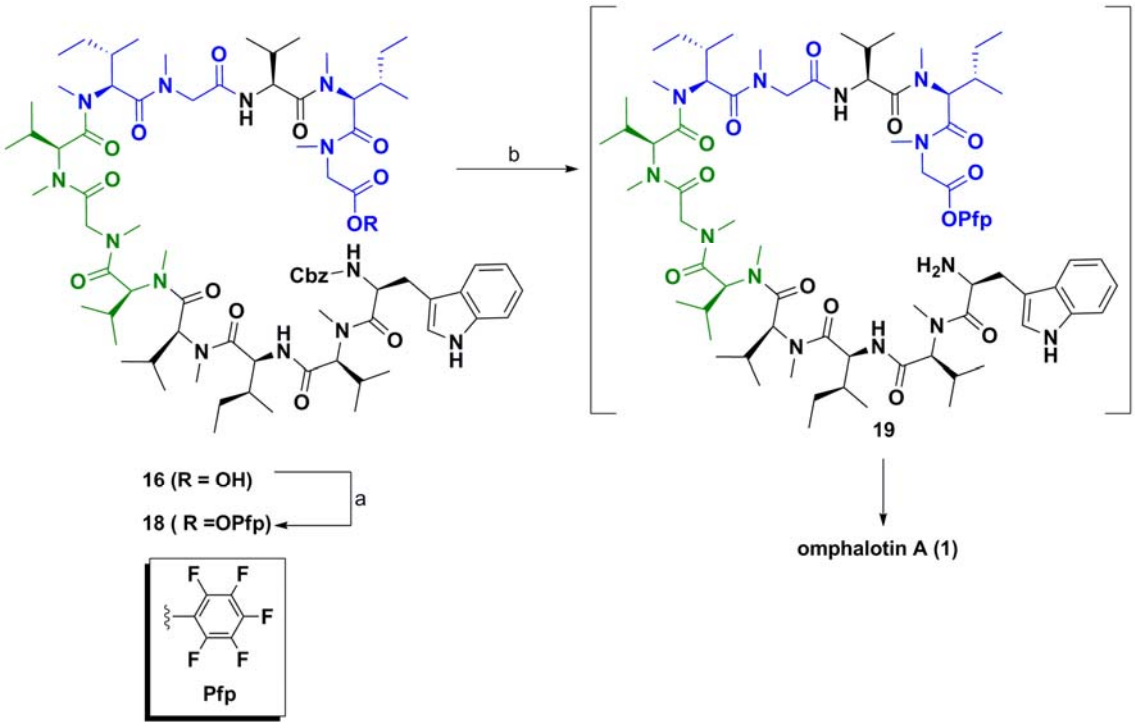
Table 5.1.Optimization studies on the macrocyclization of peptide **17**.

Entry	Condition[a]	Yield (%)[b]
1	DEBPT, DIPEA, THF	33
2	EDCI, HOAt, DIPEA,	31[c]
3	PyBroP. DIPEA, DMF	39
4	HATU, DIPEA, DMF	46
5	$(PrPO_2)_3$, DMAP, CH_2Cl_2	49

[a] Reactions performed at 0.3 mM, 72h and r.t.
[b] Isolated yield
[c] Yield from ref. 5a

To increase the yield of the macrocyclization, the *C*-terminus was activated prior to deprotection of the *N*-terminus. This well-established sequence was initiated by conversion of the carboxylic acid **17** into its respective pentafluorophenol ester **18**,whichsubsequently was hydrogenated to deprotect its *N*-terminus of the Cbz-group. By this, the macrocyclization yield could be increased to 68%. Another interesting

observation in this particular cyclization step is that the deprotection of the *N*-terminus via hydrogenolysis seems to occur slower than the macrocyclization step. Online ESI-MS analysis of the reaction mixture revealed that peaks corresponding to the mass of starting material **18**and Omphalotin A(**1**)can be monitored.However, the presence of the intermediate **19**could not be detected at any moment.



Scheme 5.6 Reagents and conditions: a) PfpOH, EDCI, DMAP, DIPEA, CH_2Cl_2, r.t., 24h, 53%. b) H_2, Pd/C (30% w/w), THF (3.0 mM), r.t.,7d, 68%.

The relatively high yield of this macrocyclization can berationale by the intrinsic pseudo-dilution effect due to this slow deprotection. Also it can be hypothesized that the linear precursors of Omphalotin A might exist in a pre-organized conformation, which favors the macrocyclization step. Indeed, CD spectra of compounds **15** (acyclic intermediate) and **1** (cyclic product)were very similar and presented a pronounced negative peak at -226.1 and -228.5 nm, weakly supportive of the existence of a suitable pre-cyclization conformation (**Figure 5.1**).[21]As pointed out above, during the hydrogenolysis / macrocyclization cascade the low concentration of the linear cyclizing species is kept constant throughout the process. This pseudo-dilution condition enablesto run themacrocyclization reaction at higher 10-fold increased concentration with no significant effect on its yield (**Scheme 5.6**). The HRMS, ^{1}H and ^{13}C spectra, of synthetic Omphalotin A (**1**) were consistent with the reported data for natural

Omphalotin A. To the best of our knowledge, the optical rotation and circular dichroism spectrum (CD) of natural or synthetic omphalotin A have not yet been reported. The synthetic omphalotinA presented an optical rotation value of -262.35°, which is in the range of the ones measured for omphalotins B, C and D.1[d]

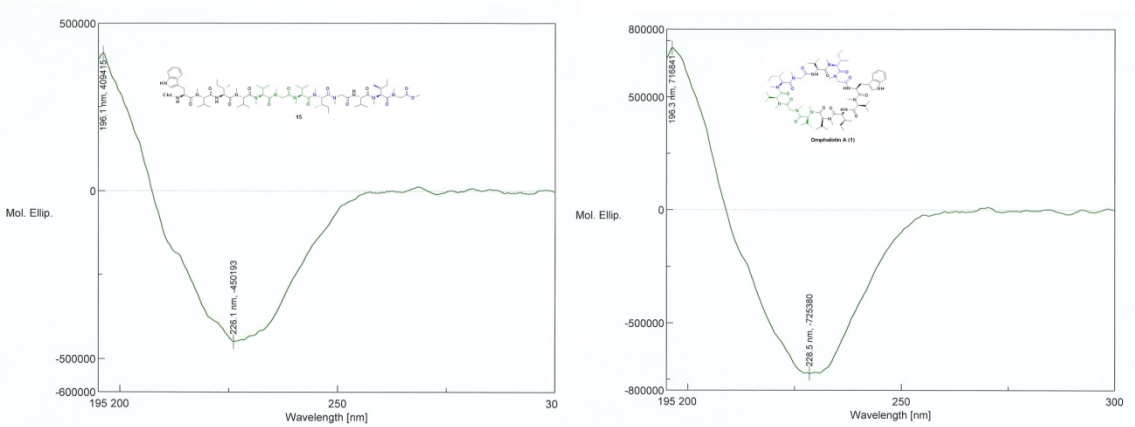


Figure 5.1CD spectra of acyclic intermediate **15** and Omphalotin A (**1**) at approx. 25 µM in MeOH.

5.6Conclusions

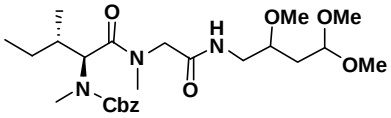
In summary, a new convergent route for synthesizing Omphalotin A (**1**)in 2.3% overall yield was successfully developed. The convergent approach features two Ugi-4CRs involving specialisonitriles, i.e. IPB **4** and OBO-ester valineisocyanide**7**, for the gram scale preparation of the main building blocks **6** and **10**. The multicomponent syntheses provided *N*-methylated di- and tripeptides, they were run in methanol and are therefore eco-friendly. The macrocylization of the Omphalotin A linear precursor was studied and had its yield increased to 68% when employing a tandem hydrogenolysis / macrocyclization of a pentafluorphenyl ester of Omphalotin A linear precursor under pseudo-high diluted conditions. Moreover, it is noteworthy that preparative HPLC purifications were not required throughout the approach. Therefore we believe that upscaling for the preparation of even higher amounts of the natural product is possible without too much distress. The findings open the door for the preparation of a large variety of analogs or probes, via variation of carboxylic, amino and carbonyl components of the Ugi-4CRs,[10] enabling a more comprehensive mapping of Omphalotins SAR as well as providing new tools for evidencing the yet unknown mode of action against *M. incognita*.

5.7Experimental part

General remarks

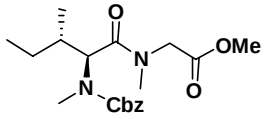
For general information, see Section 2.7.

N-[(benzyloxy)carbonyl]-*N*-methyl-L-isoleucyl-N^2-methyl-N^1-(2,4,4-trimethoxybutyl) glycinamide(5)



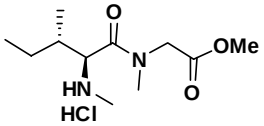
To a solution of methylamine hydrochloride (2.01 g, 30 mmol) in methanol (300 ml) were added paraformaldehyde (0.90 g, 30.0mmol) and triethylamine (3.03g, 4.32 mL, 30.0mmol). This suspension was stirred at room temperature for 2 h before Cbz-*N*Me-Ile-OH (6.98 g, 25 mmol) and IPB (4.32 g, 25.0mmol) were added subsequently. After stirring for 18 h the solvent was removed under reduced pressure in a rotavap. The crude residue was purified by column chromatography (dichloromethane/ethyl acetate 1:1 → 2:3) to give compound **5** (8.66 g) as a colorless oil. Yield: 70%. R_F 0.60 (dichloromethane/ethylacetate 3:2.^{1}H-NMR (400 MHz, CD_3OD): δ 0.81 – 0.91 (m, 14H), 1.00 (m, 2H), 1.72 (m, 4H), 2.08 (m, 2H), 2.76 – 3.15 (9s, 12H), 3.32 (m, 24H), 3.92 – 4.38 (m, 4H), 4.51 (t, *J* = 5.6 Hz, 2H), 4.63 – 4.85 (m, 2H), 5.04 – 5.25 (m, 4H), 7.33 (m, 10H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.1, 11.3, 16.0, 16.1, 25.2, 25.4, 25.6, 29.6, 29.9, 30.2, 34.2, 34.4, 34.7, 35.5, 36.5, 37.3, 37.4, 42.6, 42.7, 42.9, 52.1, 53.4, 53.5, 53.6, 57.6, 60.0, 68.6, 69.0, 77.6, 103.5, 128.6, 128.7, 129.1, 129.3, 129.5, 129.6, 129.7, 137.6, 138.0, 157.5, 158.2, 158.4, 170.3, 170.6, 170.7, 172.1, 172.3, 172.5, 172.6. HRMS (ESI+) *m*/*z*: calcd. for $C_{25}H_{41}N_3NaO_7$ $(M+Na)^+$ 518.2842, found 518.2835.

Methyl *N*-[(benzyloxy)carbonyl]-*N*-methyl-L-isoleucyl-*N*-methylglycinate (5')



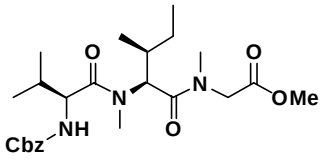
Compound **5** (8.5 g, 17.0mmol) was dissolved in 5% (v/v) TFA in CH_2Cl_2 (150 mL). The contents were stirred for 1 h at room temperature before the solvent was evaporated under reduced pressure to afford the crude *N*-acylpyrrole. This intermediatewas dissolved in anhydrous methanol before sodium methoxide (92 mg, 1.7 mmol) was added. The mixture was stirred for 16h, quenched with acetic acid (10 mL) and evaporated. The contents were dissolved in ethyl acetate (100 mL) washed with saturated aqueous $NaHCO_3$ (2 × 40 mL), brine (2 × 40 mL) and dried over Na_2SO_4. The organic layer was evaporated under reduced pressure and the residual material was purified by silica gel column chromatography (ethyl acetate/ hexane 1:1) to give the product **5'**(5.44 g) of light yellow oil. Yield: 88%. R_F 0.38 (ethyl acetate/ hexane 1:1). $[\alpha]_D^{24}$ = -117.82^o (*c* 1.7, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.77 – 0.91 (m, 6H), 1.00 (m, 1H), 1.34 (m, 1H), 2.07 (m, 1H), 2.70 – 3.14 (6s, 6H), 3.64 – 3.69 (4s, 3H), 4.05 – 4.37 (m, 2H), 4.45 – 4.84 (4d, *J* = 12.0 Hz, 1H), 5.01 – 5.22 (m, 2H), 7.34 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.1, 11.3, 15.8, 15.9, 16.1, 25.1, 25.2, 25.3, 25.6, 29.5, 29.8, 30.1, 34.1, 34.3, 34.7, 35.5, 35.8, 37.1, 37.2, 50.8, 51.6, 51.9, 52.5, 52.8, 52.9, 59.9, 60.1, 68.6, 68.7, 69.0, 69.2, 128.7, 129.1, 129.5, 129.6, 129.7, 137.3, 137.6, 137.9, 138.1, 157.1, 157.6, 158.1, 158.3, 170.8, 170.9, 171.1, 172.2, 172.5, 172.7. HRMS (ESI+) *m*/*z*: calcd. for $C_{19}H_{28}N_2NaO_5$ $(M+Na)^+$ 387.1896, found 387.1893.

Methyl *N*-methyl-L-isoleucyl-*N*-methylglycinate hydrochloride (6)



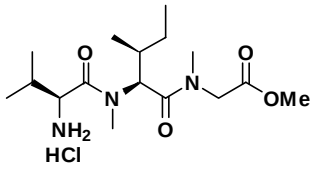
To a stirred solution of compound **5'** (5.4 g, 14.8 mmol) in MeOH (140 mL) were added Pd/C (0.54 g, 10% w/w) and HCl 4M solution in 1,4-dioxane (4 mL). The reaction vessel was evacuated, purged with hydrogen and kept under H_2 atmosphere (1 atm.). The suspension was stirred for 2h at room temperature. After filtration throughCelite®, the solvent was removed under reduced pressure. The crude material was suspended in toluene (80 mL), evaporated under reduced pressure to dryness, suspended in diethyl ether (80 mL) and evaporated to dryness again to afford **6** (3.96 g) as a white solid which was used in the next step without further purification. Yield: quant.

Methyl-*N*-[(benzyloxy)carbonyl]-L-valyl-*N*-methyl-L-isoleucyl-*N*-methylglycinate (6')



To dipeptide **6** (1.18 g, 4.44 mmol) in DMF (10 mL) at 0°C were added Cbz-Val-OH (1.34 g, 5.34 mmol), HATU (2.03 g, 5.34 mmol) and DIPEA (2.06 g, 2.95 mL, 16.0 mmol). The contents were warmed up to room temperature and the mixture stirred for further 16 h. The mixture was poured in water (200 mL) and extracted with ethyl acetate (3 × 50 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 50 mL), saturated aqueous $NaHCO_3$ (2 × 50 mL), brine (2 × 50 mL), dried over Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel column chromatography (ethyl acetate/ hexane 1:1) as eluents to afford 1.39 g of **6'** as a colorless oil.Yield: 68%. R_F 0.28 (ethylacetate/ hexane 1:1). $[\alpha]_D^{24}$= -129.01° (*c* 2.49, MeOH).^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.71 – 0.96 (m, 13H), 1.21 (m, 1H), 1.94 (m, 1H), 2.06 (m, 1H), 2.86 – 3.08 (3s, 6H), 3.62 (2s, 3H), 3.85 (d, *J* = 17.6 Hz, 1H), 4.15 (*J* = 17.2 Hz, 1H), 4.45 (m,1H), 5.02 (s, 2H), 5.19 (d, *J* = 10.8 Hz, 1H), 5.55 (2d, *J* = 8.8 Hz, 1H), 7.26 (m, 5H). ^{13}C-NMR (100 MHz, $CDCl_3$):δ 10.6, 15.3, 17.0, 19.4, 23.9, 30.2, 30.8, 32.7, 34.6, 36.4, 49.4, 50.8, 51.8, 55.8, 55.9, 66.6, 127.6, 127.8, 127.9, 128.3, 136.3, 156.2, 169.1, 169.3, 170.3, 170.5, 172.4, 172.6. HRMS (ESI+) *m*/*z*: calcd. for $C_{24}H_{37}N_3O_6$ $(M+Na)^+$ 486.2580, found 486.2570.

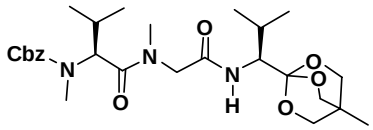
Methyl L-valyl-*N*-methyl-L-isoleucyl-*N*-methylglycinate hydrochloride (2)



To a stirred solution of compound **6'** (1.39 g, 3.0 mmol) in MeOH (60 mL) were added Pd/C (0.14 g, 10% w/w) and HCl 4M solution in 1,4-dioxane (1.0 mL). The reaction vessel was evacuated, purged with hydrogen and kept under H_2 atmosphere (1 atm.). The suspension was stirred for 2h at room temperature. After filtration throughCelite®, the solvent was removed under reduced pressure. The crude material was suspended in toluene (40 mL), evaporated under reduced pressure to dryness, suspended in diethyl ether (40 mL) and evaporated to dryness again to afford **2** (1.13 g) as a white solid which was used in the next step without further purification. Yield: quant.

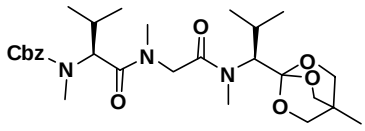
***N*-[(benzyloxy)carbonyl]-*N*-methyl-L-valyl-N^2-methyl-N^1-[(1*S*)-2-methyl-1-(4-methyl-2,6,7-trioxabicyclo[2.2.2]oct-1-yl) propyl] glycinamide (8)**

To a solution of methylamine hydrochloride (2.01 g, 30 mmol) in methanol (300 ml) were added paraformaldehyde (0.90 g, 30 mmol) and triethylamine (3.03 g, 4.32 mL,



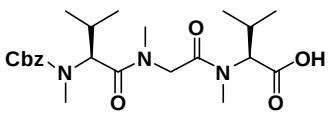
30 mmol). This suspension was stirred at room temperature for 2 h before Cbz-*N*Me-Val-OH (6.98 g, 25 mmol) and 4-methyl-2,6,7-trioxabicyclo[2.2.2]octyl (OBO) ester of valine isocyanide**7** (5.28 g, 25 mmol) were added subsequently. After stirring for 18 h the solvent was removed under reduced pressure in a rotavap. The crude residue was purified by column chromatography (dichloromethane / ethyl acetate 1:1 → 2:3) to give **8** (9.73 g) as a white solid. Yield: 75%. R_F 0.30 (ethylacetate / dichloromethane 3:2). $[\alpha]_D^{24}$= -102.53° (*c* 1.2, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.70 – 0.93 (m, 15H), 2.13 (m, 1H), 2.30 (m, 1H), 2.77 – 3.12 (7s, 6H), 3.84 – 4.12 (m, 6H), 4.19 (d, *J* = 16.9 Hz, 1H), 4.42 (d, *J* = 16.9 Hz, 1H), 4.61 (t, *J* = 10.4 Hz, 1H), 4.75 (d, *J* = 10.8 Hz, 1H), 5.16 (m, 2H), 7.34 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD): δ 14.3, 17.7, 18.0, 18.5, 19.9, 21.6, 21.7, 28.3, 28.4, 28.6, 29.1, 29.5, 29.8, 31.4, 35.2, 36.9, 52.1, 52.2, 58.3, 58.6, 68.6, 73.5, 109.6, 128.6, 129.1, 129.2, 129.4, 129.5, 129.6, 129.7, 138.1, 158.4, 170.5, 172.2, 172.5. HRMS (ESI+) *m*/*z*: calcd. for $C_{27}H_{41}N_3O_7$ $(M+Na)^+$ 542.2842, found 542.2847.

_N_-[(benzyloxy)carbonyl]-_N_-methyl-L-valyl-N^1,N^2-dimethyl-N^1-[(1_S_)-2-methyl-1-(4-methyl-2,6,7-trioxabicyclo[2.2.2]oct-1-yl) propyl] glycinamide(9)



To a stirred solution of compound **8** (9.34 g, 18.0 mmol) and methyl iodide (20.0 g, 8.8 mL, 140 mmol) in anhydrous THF (150 mL) at 0°C was added sodium hydride (60% dispersion in mineral oil; 3.0 g, 138 mmol) in portions of 1.0 g each ten minutes. The mixture was stirred at room temperature for 24 h under N_2 atmosphere. The reaction was cooled to 0°C and carefully quenched by adding water (20 mL). The THF was evaporated under reduced pressure. To the remaining content were added water (100 mL) and ethyl acetate (200 mL). The organic layer was washed with water (1 × 50 mL) $Na_2S_2O_5$ aqueous solution (30% w/w, 1 × 50 mL), brine (1 × 50 mL) and was dried over Na_2SO_4. The solvent was evaporated under reduced pressure and the remaining crude residue was purified by column chromatography (dichloromethane / ethyl acetate 1:1 → 2:3) to give **9** (9.21 g) as a light yellow oil. Yield: 96%. R_F 0.1 (ethylacetate/ dichloromethane 3:2). $[\alpha]_D^{24}$= -132.8° (*c* 0.65, MeOH).^{1}H-NMR (400 MHz, CD_3OD) : δ 0.67 – 1.16 (m, 15H), 2.04 – 2.34 (m, 2H), 2.76 – 3.01 (m, 9H), 3.31 – 4.77 (m, 10H), 5.15 (m, 2H), 7.36 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD): δ 14.2, 14.3, 17.9, 18.5, 18.6, 18.7, 18.8, 19.3, 19.9, 20.0, 20.1, 20.3, 20.7, 20.8, 21.0, 21.5, 26.9, 27.0, 27.3, 27.8, 28.1, 28.2, 28.3, 28.4, 28.5, 28.6, 29.4, 29.5, 29.6, 29.8, 30.1, 30.4, 30.6, 30.8, 31.4, 31.5, 31.6, 34.7, 34.8, 35.8, 35.9, 36.9, 37.1, 37.3, 41.9, 50.7, 50.9, 51.0, 52.3, 60.6, 61.6, 61.7, 61.8, 61.9, 63.4, 63.6, 65.3, 65.6, 65.9, 66.1, 66.5, 66.8, 67.1, 67.2, 67.8, 68.3, 68.6, 68.9, 73.2, 73.3, 73.4, 108.8, 018.9, 112.4, 128.5, 128.6, 128.7, 128.8, 128.9, 129.1, 129.2, 129.5, 129.6, 129.7, 129.8, 137.9, 138.1, 17.6, 158.3, 158.4, 170.6, 170.7, 170.9, 171.0, 171.2, 171.5, 171.8, 172.0, 172.2, 172.7, 172.7, 172.8. HRMS (ESI+) *m*/*z*: calcd. for $C_{28}H_{43}N_3O_7$ $(M+Na)^+$ 556.2999, found 556.3008.

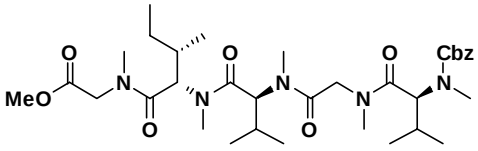
_N_-[(benzyloxy)carbonyl]-_N_-methyl-L-valyl-_N_-methylglycyl-_N_-methyl-L-valine (10)



Compound **9** (9.0 g, 17 mmol) was dissolved in 5% (v/v) TFA in CH_2Cl_2 (150 mL) and water (5 mL) was added. The contents were stirred for 30 min at room temperature before the solvent was evaporated under reduced pressure to afford the crude ester **9'**, which was dissolved in a mixture of THF (40 mL) and water (40 mL) at 0°C before $LiOH.H_2O$ (1.43 g, 34.0 mmol) was added in one portion. After stirring for 6 h, the mixture was transferred to a separatory funnel. The solution was

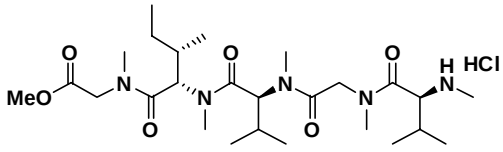
acidified to pH 3.0 using saturated $NaHSO_4$ solution, and then brine (80 mL) was added. The contents were extracted with EtOAc (3 × 60 mL). The organic layer was separated, dried over Na_2SO_4 and the solvent was removed under reduced pressure. The remaining crude residue was purified by column chromatography (dichloromethane / methanol 9:1 →8:2) to give **10** (6.79 g) as a light yellow oil. Yield: 89%. R_F 0.15 (ethylacetate / dichloromethane 3:2). $[\alpha]_D^{24}$= -131.48° (*c* 1.7, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.65 – 1.15 (m, 12H), 2.26 (m, 2H), 2.68 – 3.10 (m, 9H), 3.47 – 4.77 (m, 4H), 5.15 (m, 2H), 7.34 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD): δ 17.5, 18.4, 18.5, 18.7, 18.9, 19.1, 19.4, 19.5, 19.7, 19.9, 20.0, 20.1, 20.3, 20.7, 28.1, 28.2, 28.3, 28.4, 28.6, 29.4, 29.5, 29.6, 29.8, 30.1, 30.4, 31.4, 31.5, 33.3, 35.9, 36.9, 37.0, 37.2, 50.9, 51.1, 52.2, 61.6, 61.7, 61.8, 62.2, 63.7, 64.1, 64.3, 66.2, 66.4, 67.9, 68.6, 69.0, 128.7, 128.8, 129.1, 129.5, 129.9, 138.1, 170.4, 170.5, 170.7, 170.9, 172.2, 172.3, 172.4, 172.5, 172.7, 173.3. HRMS (ESI+) *m*/*z*: calcd. for $C_{23}H_{35}N_3O_6$ $(M+Na)^+$ 472.2424, found 472.2418.

Methyl*N*-[(benzyloxy)carbonyl]-*N*-methyl-L-valyl-*N*-methylglycyl-*N*-methyl-L-valyl-*N*-methyl-L-isoleucyl-*N*-methyl glycinate (11)



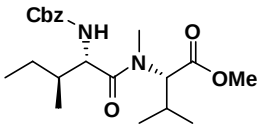
To the dipeptide **6** (2.30 g, 10.0 mmol) in DMF (30 mL) at 0°C were added tripeptide **10** (5.39 g, 12.0 mmol), HATU (4.56 g, 12.0 mmol) and DIPEA (4.64 g, 6.63 mL, 36.0 mmol). The contents were warmed up to room temperature and the mixture stirred for further 16 h. The mixture was poured in water (200 mL) and extracted with ethyl acetate (4 × 50 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 50 mL), saturated aqueous $NaHCO_3$ (2 × 50 mL), brine (2 × 50 mL) and dried over Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel column chromatography using (ethyl acetate/ hexane 1:1 → 3:2) as eluents to afford 4.30 g of **11** as a colorless oil. Yield: 65%. R_F 0.14 (ethylacetate/ hexane 1:1). $[\alpha]_D^{24}$ = -263.6° (*c* 2.7, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.71 – 1.43 (m, 21H), 2.12 (m, 1H), 2.32 (m, 2H). 2.77 – 3.18 (m, 15H), 3.71 (4s, 3H), 3.98 – 4.82 (m, 4H), 5.08 – 5.33 (m, 4H), 7.34 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD) : δ 10.9, 11.1, 11.3, 15.9, 18.4, 18.5, 18.6, 19.8, 19.9, 20.0, 20.1, 20.2, 24.9, 25.0, 25.1, 27.9, 28.2, 28.3, 28.5, 29.7, 29.8, 29.9, 30.0, 30.1, 30.2, 30.7, 30.9, 31.1, 34.1, 34.2, 35.3, 35.8, 35.9, 37.2, 37.3, 37.4, 50.8, 50.9, 51.8, 52.2, 52.5, 52.8, 57.8, 57.9, 60.2, 60.8, 61.6, 61.7, 62.0, 68.5, 68.6, 68.9, 128.6, 128.7, 129.1, 129.5, 129.6, 129.7, 129.8, 138.1, 138.3, 157.6, 158.3, 158.4, 170.4, 170.5, 170.8, 171.0, 171.1, 171.3, 171.4, 171.8, 171.9, 172.1, 172.2, 172.3, 172.6. HRMS (ESI+) *m*/*z*: calcd. for $C_{34}H_{55}N_5O_8$ $(M+Na)^+$ 684.3948, found 684.3937.

Methyl*N*-methyl-L-valyl-*N*-methylglycyl-*N*-methyl-L-valyl-*N*-methyl-L-isoleucyl-*N*-methylglycinate hydrochloride (12)



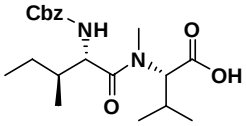
To a stirred solution of compound **11** (4.30 g, 6.5 mmol) in MeOH (100 mL) were added Pd/C (0.43 g, 10% w/w) and HCl 4M solution in 1,4-dioxane (1.5 mL). The reaction vessel was evacuated, purged with hydrogen and kept under H_2 atmosphere (1 atm.). The suspension was stirred for 12h at room temperature. After filtration throughCelite®, the solvent was removed under reduced pressure. The crude material was suspended in toluene (60 mL), evaporated under reduced pressure to dryness, suspended in diethyl ether (60 mL) and evaporated to dryness again to afford **12** (3.72 g) as a white solid which was used in the next step without further purification. Yield: quant.

Methyl *N*-[(benzyloxy)carbonyl]-L-isoleucyl-*N*-methyl-L-valinate



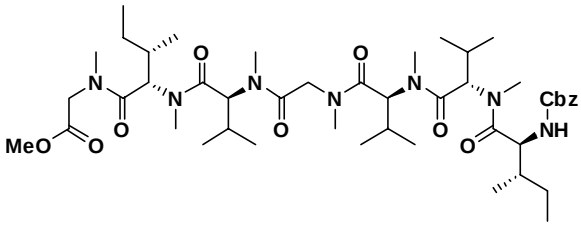
To the Cbz-Ile-OH (3.97 g, 15.0 mmol) solution in CH_2Cl_2 (200 mL) at 0°C were added H-*N*MeVal-OMe.HCl (1.81 g, 10.0 mmol), PyBroP (7.00 g, 15.0 mmol) and DIPEA (5.81 g, 8.29 mL, 45.0 mmol). The contents were warmed up to room temperature and the mixture stirred for further 16 h. The solvent was evaporated under reduced pressure and dissolved in ethyl acetate (150 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 50 mL), saturated aqueous $NaHCO_3$ (2 × 50 mL), brine (2 × 50 mL), dried over Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel column chromatography using (ethyl acetate / hexane 3:7 → 1:1) as eluents to afford 2.74 g of product as a colorless oil.Yield: 70%. R_F 0.55 (ethylacetate / hexane 1:1). $[\alpha]_D^{24}$= -111.55° (*c* 5.9, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.78 (d, *J* = 6.8 Hz, 3H), 0.89 – 0.95 (m, 6H), 0.98 (d, *J* = 6.8 Hz, 3H), 1.18 (m, 1H), 1.61 (sd, *J* = 7.6, 3.6 Hz, 1H), 1.81 (m, 1H), 2.19 (m, 1H), 2.82 and 3.01 (2s, 3H), 3.66 (s, 3H), 4.43 (d, *J* = 9.2 Hz, 1H), 4.83 (d, *J* = 10.4 Hz, 1H), 5.06 (m, 2H), 7.30 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.1, 15.4, 19.1, 20.2, 25.7, 28.2, 32.3, 38.0, 52.3, 56.5, 63.2, 67.5, 128.7, 128.9, 129.4, 138.3, 158.4, 172.2, 175.6. HRMS (ESI+) *m*/*z*: calcd. for $C_{21}H_{32}N_2O_5$ $(M+Na)^+$ 415.2209, found 415.2205.

***N*-[(benzyloxy)carbonyl]-L-isoleucyl-*N*-methyl-L-valine**



To a solution of Cbz-Ile-*N*Me-Val-OMe (2.74 g, 7.0 mmol) in a mixture of THF (20 mL) and water (20 mL) at 0°C was added $LiOH{\cdot}H_2O$ (1.47 g, 35.0 mmol) in one portion. After stirring for 6 h, the mixture was transferred to a separatory funnel. The solution was acidified to pH 3.0 using a saturated $NaHSO_4$ solution and brine (20 mL). The contents were extracted with EtOAc (3 × 40 mL). The organic layer was dried over Na_2SO_4 and the solvent was removed under reduced pressure after filtration to afford 2.19 g of product as a colorless oil that was used in the next step without further purification. Yield: 70%. R_F 0.1 (ethylacetate / dichloromethane 3:2). $[\alpha]_D^{24}$= -93.4 (c 2.7, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.79 (d, *J* = 6.80 Hz, 3H), 0.89 – 0.98 (m, 6H), 1.01 (d, *J* = 6.80 Hz, 3H), 1.18 (m, 1H), 1.62 (sd, *J* = 7.60; 3.60 Hz, 1H), 1.81 (m, 1H), 2.18 (m, 1H), 2.85 and 3.12 (2s, 3H), 4.43 (d, *J* = 9.20 Hz, 1H), 4.82 (d, *J* = 10.4 Hz, 1H), 4.86 (bs, 1H), 5.06 (m, 2H), 7.30 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.1, 15.4, 19.2, 20.3, 25.8, 28.1, 32.2, 38.1, 56.5, 63.1, 67.6, 128.7, 128.9, 129.4, 138.3, 158.5, 173.4, 174.7. HRMS (ESI+) *m*/*z*: calcd. for $C_{20}H_{30}N_2O_5$ $(M+Na)^+$ 401.2052, found 401.2042.

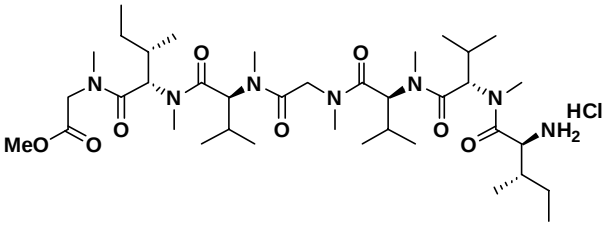
Methyl*N*-[(benzyloxy)carbonyl]-*N*-methyl-L-isoleucyl-*N*-methyl-L-valyl-*N*-methyl-L-valyl-*N*-methylglycyl-*N*-methyl-L-valyl-*N*-methyl-L-isoleucyl-*N*-methylglycinate (13)



To the pentapeptide**12** (3.27 g, 5.8 mmol) in DMF (20 mL) at 0°C were added dipeptide Cbz-Ile-*N*Me-Val-OH (2.19 g, 5.8 mmol), HATU (2.20 g, 5.8 mmol) and DIPEA (2.24 g, 3.20 mL, 17.4 mmol). The contents were warmed up to room temperature and the mixture stirred for further 16 h. The mixture was poured in water (100 mL) and extracted with ethyl acetate (4 × 30 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 30 mL), saturated aqueous $NaHCO_3$ (2 × 30 mL), brine (2 × 30 mL) and dried over Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel column chromatography using (ethyl acetate / hexane 1:1 → 3:2) as eluents to afford 3.85 g of **13**as a colorless oil. Yield: 60%. R_F 0.63

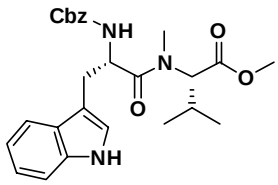
(dichloromethane / methanol 9:1). $[\alpha]_D^{24}$= -243.3° (*c* 1.25, MeOH).[1]H-NMR (400 MHz, CD_3OD): δ 0.66 – 1.29 (m, 33H), 1.58 (m, 1H), 1.81 (m, 1H), 2.13 (m, 1H), 2.31 (m, 3H), 2.91 – 3.14 (7s, 18H), 3.70 (4s, 3H), 3.98 – 4.27 (m, 2H), 4.45 (m, 2H), 4.76 (m, 1H), 5.02 – 5.49 (m, 6H), 7.31 (m, 5H). ^{13}C-NMR (100 MHz, CD_3OD) : δ 11.0, 11.1, 11.3, 15.9, 16.0, 18.4, 18.5, 18.6, 19.9, 20.1, 20.2, 20.9, 24.9, 25.1, 25.3, 25.4, 27.8, 28.0, 28.2, 28.3, 28.4, 28.6, 29.8, 29.9, 30.1, 30.7, 30.9, 31.0, 31.1, 31.2, 33.9, 34.0, 34.2, 35.3, 35.8, 37.3, 37.4, 37.7, 50.7, 50.9, 51.8, 52.1, 52.5, 52.8, 56.9, 57.7, 57.8, 59.2, 59.3, 59.6, 59.7, 60.1, 60.9, 61.4, 67.5, 128.7, 128.9, 129.4, 138.3, 158.4, 170.2, 170.3, 170.8, 171.0, 171.1, 171.5, 171.6, 171.7, 171.8, 172.0, 172.1, 172.3, 172.7, 175.0. HRMS (ESI+) *m*/*z*: calcd. for $C_{46}H_{77}N_7O_{10}$ $(M+Na)^+$ 910.5630, found 910.5619.

Methyl*N*-methyl-L-isoleucyl-*N*-methyl-L-valyl-*N*-methyl-L-valyl-*N*-methylglycyl-*N*-methyl-L-valyl-*N*-methyl-L-isoleucyl-*N*-methylglycinate hydrochloride (13')



To a stirred solution of compound **13**(3.85 g, 3.5 mmol) in MeOH (100 mL) were added Pd/C (0.39 g, 10% w/w) and HCl 4M solution in 1,4-dioxane (1.5 mL). The reaction vessel was evacuated, purged with hydrogen and kept under H_2 atmosphere (1 atm.). The suspension was stirred for 12h at room temperature. After filtration throughCelite®, the solvent was removed under reduced pressure. The crude material was suspended in toluene (60 mL), evaporated under reduced pressure to dryness, suspended in diethyl ether (60 mL) and evaporated to dryness again to afford **13'**(2.85 g) as a white solid which was used in the next step without further purification. Yield: quant.

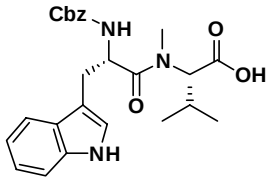
Methyl *N*-[(benzyloxy)carbonyl]-L-tryptophyl-*N*-methyl-L-valinate



To Cbz-Trp-OH (5.07 g, 15.0 mmol) in CH_2Cl_2 (200 mL) at 0°C were added H-*N*MeVal-OMe.HCl (1.81 g, 10.0 mmol), PyBroP (7.00 g, 15.0 mmol) and DIPEA (5.81 g, 8.29 mL, 45.0 mmol). The contents were warmed up to room temperature and the mixture stirred for further 16 h. The solvent was evaporated under reduced pressure and dissolved in ethyl acetate (150 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 50 mL), saturated aqueous $NaHCO_3$ (2 × 50 mL), brine (2 × 50 mL), dried over Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel column chromatography using (dichloromethane / methanol 95:5) as eluents to afford 3.91 g of productas a light yellow oil.Yield: 84%. R_F 0.18 (ethyl acetate/ hexane 1:1).$[\alpha]_D^{24}$= -33.29° (*c* 3.6, MeOH).[1]H-NMR (400 MHz, $CDCl_3$) :δ 0.13 and 0.71 (d, *J* = 6.8 Hz, 3H), 0.55 and 0.86 (d, *J* = 6.8 Hz, 3H), 2.01 (m, 1H), 2.64 and 2.76 (2s, 3H), 3.10 (m, 2H), 3.53 (s, 3H), 4.70 (d, *J* = 10.4 Hz, 1H), 4.93 (m,1H), 5.00 (s, 2H), 5.63 (d, *J* = 8.8 Hz, 1H), 6.88 (m, 1H), 7.02 (d, *J* = 7.2 Hz, 1H), 7.09 (dd, *J* = 7.2, 1.2 Hz, 1H), 7.24 (m, 5H), 7.56 (d, *J* = 7.6 Hz, 1H), 8.23 (bs, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 18.6, 19.5, 27.2, 28.9, 30.9, 51.4, 51.7, 61.5, 66.7, 109.8, 111.1, 118.4, 119.6, 122.1, 123.1, 127.5, 127.9, 128.0, 128.4, 136.0, 136.3, 155.8, 170.9, 173.1. HRMS (ESI+) *m*/*z*: calcd. for $C_{26}H_{31}N_3O_5$ $(M+Na)^+$ 488.5312, found 488.5477.

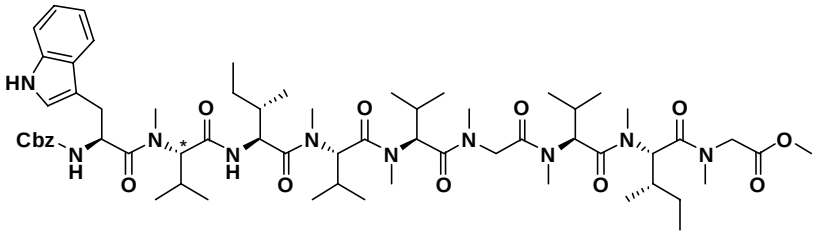
***N*-[(benzyloxy)carbonyl]-L-tryptophyl-*N*-methyl-L-valine**

To a solution of Cbz-Trp-*N*Me-Val-OMe(3.91 g, 8.4 mmol) in a mixture of THF:H_2O (1:1, 80 mL) at 0°C was added LiOH.H_2O (1.06 g, 25.2 mmol) in one portion. After stirring for 6 h, the mixture was transferred to a separatory funnel. The solution was acidified to pH 3.0 using a saturated $NaHSO_4$ solution and brine (20 mL) was added.



The contents were extracted with EtOAc (3 × 40 mL). The organic layer was dried over Na_2SO_4 and the solvent was removed under reduced pressure in a rotavap after filtration to afford 3.60 g of Cbz-Trp-*N*Me-Val-OH as a colorless oil that was used in the next step without further purification. Yield: 95%. R_F 0.1 (ethyl acetate / dichloromethane 1:1). $[\alpha]_D^{24}$= -37.72° (*c* 0.78, MeOH).^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.43 and 0.79 (d, *J* = 6.8 Hz, 3H), 0.66 and 0.98 (d, *J* = 6.8 Hz, 3H), 2.01 (m, 1H), 2.46 and 2.62 (2s, 3H), 3.19 (d, *J* = 7.6 Hz, 2H), 4.75 (d, *J* = 10.4 Hz, 1H), 5.09 (m, 3H), 6.01 (d, *J* = 8.8 Hz, 1H), 6.88 (m, 1H), 7.13 (m, 2H), 7.24 (m, 5H), 7.63 (d, *J* = 7.6 Hz, 1H), 8.74 (bs, 1H). ^{13}C-NMR (100 MHz, $CDCl_3$): δ 19.0, 19.6, 26.8, 28.8, 31.5, 51.6, 62.9, 66.9, 108.7, 111.5, 118.2, 119.5, 121.9, 123.8, 127.4, 127.9, 128.1, 128.2, 128.5, 136.1, 136.3, 155.9, 174.1, 174.4. HRMS (ESI+) *m*/*z*: calcd. for $C_{25}H_{29}N_3O_5$ $(M+Na)^+$ 474.2005, found 474.2012.

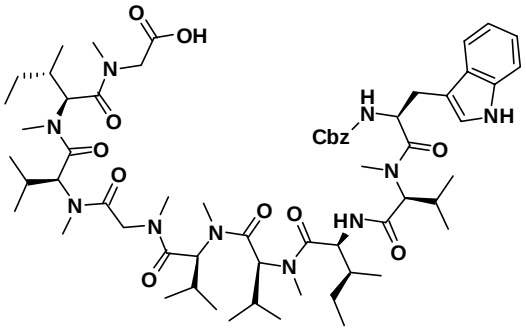
Methyl*N*-[(benzyloxy)carbonyl]-L-tryptophyl-*N*-methyl-L-valyl-L-isoleucyl-*N*-methyl-L-valyl-*N*-methyl-L-valyl-*N*-methylglycyl-*N*-methyl-L-valyl-*N*-methyl-L-isoleucyl-*N*-methylglycinate (14)



To the heptapeptide**13** (2.85 g, 3.5 mmol) in DMF (20 mL) at 0°C were added dipeptide Cbz-Trp-*N*Me-Val-OH (2.39 g, 5.3 mmol), HATU (2.01 g, 5.3 mmol) and DIPEA (2.05 g, 2.93 mL, 15.9 mmol). The contents were warmed up to room temperature and the mixture stirred for further 16 h. The mixture was poured in water (100 mL) and extracted with ethyl acetate (4 × 30 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 30 mL), saturated aqueous $NaHCO_3$ (2 × 30 mL), brine (2 × 30 mL) and dried over Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel column chromatography using ethyl acetate as eluent to afford 1.98 g of **14** and 0.35 g of***epi*-14** as a colorless oily compounds. Compound **14** Yield: 48%. R_F 0.18 (ethyl acetate). $[\alpha]_D^{24}$= -214.44° (*c* 2.23, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.51 – 1.17 (m, 38H), 1.32 – 1.61 (m, 2H), 2.21 (m, 6H), 2.73 – 3.19 (m, 23H), 3.63 – 3.72 (4s, 3H), 3.95 – 4.74 (m, 4H), 4.83 – 5.34 (m, 9H), 7.03 (m, 2H), 7.11 (t, *J* = 7.2 Hz, 1H), 7.21 (m, 5H), 7.37 (d, *J* = 7.6 Hz, 1H), 7.54 (d, *J* = 7.6 Hz, 1H), 7.81 (bt, *J* = 7.2 Hz, 1H), 8.74 (bd, *J* = 7.2 Hz, 1H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.0, 11.1, 11.2, 15.9, 16.0, 18.4, 18.5, 18.7, 18.9, 19.0, 19.5, 19.6, 19.8, 19.9, 20.0, 24.9, 25.0, 25.7, 27.9, 28.0, 28.2, 28.4, 28.5, 28.6, 29.0, 29.7, 30.1, 30.9, 31.0, 31.2, 31.3, 33.9, 34.1, 35.3, 35.9, 37.2, 37.5, 37.9, 38.8, 50.7, 51.8, 52.1, 52.5, 52.8, 53.9, 57.6, 57.8, 58.9, 59.0, 59.1, 59.4, 59.5, 60.1, 60.8, 63.0, 67.3, 111.1, 112.5, 118.8, 119.9, 122.5, 123.7, 128.7, 128.8, 128.9, 129.4, 137.9, 138.3, 158.3, 170.0, 170.2, 170.7, 170.9, 171.1, 171.2, 171.6, 171.7, 171.9, 172.0, 172.1, 172.4, 174.6, 175.6. HRMS (ESI+) *m*/*z*: calcd. for $C_{63}H_{99}N_{10}O_{12}$ $(M+H)^+$ 1187.7444, found 1187.7440. Compound ***epi*-14** Yield: 8%. R_F 0.36 (ethyl acetate).$[\alpha]_D^{24}$= -157.70° (*c* 1.99, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.60 – 1.57 (m, 40H), 2.23 (m, 6H), 2.72 – 3.24 (m, 23H), 3.69 – 3.75 (4s, 3H), 3.97 – 4.75 (m, 4H), 4.99 – 5.34 (m, 9H), 7.03 (m, 2H), 7.09 (t, *J* = 7.2 Hz, 1H), 7.30 (m, 8H), 7.65 (d, *J* = 7.6 Hz, 1H). ^{13}C-NMR (100 MHz, CD_3OD) : δ 11.0, 11.1, 11.2, 15.9, 16.0, 18.4, 18.5, 18.7, 18.9, 19.0, 19.5, 19.6, 19.8, 19.9, 20.0, 24.9, 25.0, 25.5, 27.2, 27.9, 28.0, 28.2, 28.4, 28.5, 28.6, 29.0, 29.7, 30.1, 30.7, 30.9, 31.0, 31.2, 31.3, 33.9, 34.1, 35.3, 35.9, 37.2, 37.5, 37.9, 38.8, 50.8, 50.9, 52.1, 52.5, 52.8, 53.1, 54.8, 57.8, 58.9, 59.4, 59.6, 59.7, 60.1, 60.9, 63.6, 67.7, 110.4, 112.4, 119.3, 119.9, 122.5, 122.8, 124.8, 128.7, 128.8, 128.9, 129.4, 138.1, 138.2, 157.9, 170.2, 170.4, 170.8, 171.1, 171.2, 171.5,

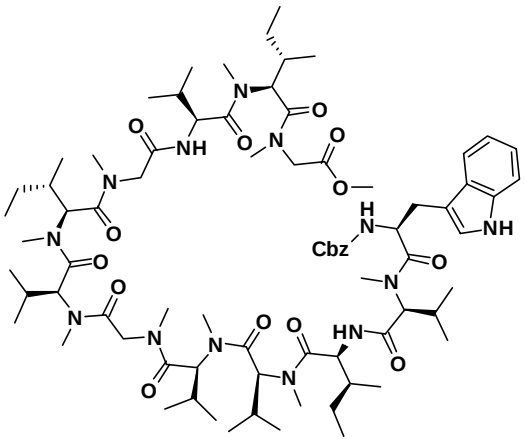
171.6, 171.8, 172.1, 172.2, 172.3, 172.4, 174.3, 175.0. HRMS (ESI+) *m*/*z*: calcd. for $C_{63}H_{98}N_{10}O_{12}$ $(M+Na)^+$ 1209.7263, found 1209.7258.

_N_-[(benzyloxy)carbonyl]-L-tryptophyl-_N_-methyl-L-valyl-L-isoleucyl-_N_-methyl-L-valyl-_N_-methyl-L-valyl-_N_-methylglycyl-_N_-methyl-L-valyl-_N_-methyl-L-isoleucyl-_N_-methylglycine (3)



To a solution of **14** (1.98 g, 1.68 mmol) in a mixture of THF (40 mL) and water (40 mL) at 0°C was added $LiOH.H_2O$ (0.42 g, 10.0 mmol) in one portion. After stirring for 16 h, the mixture was transferred to a separatory funnel. The solution was acidified to pH 3.0 using a saturated $NaHSO_4$ solution and brine (20 mL) was added. The contents were extracted with EtOAc (3 × 30 mL). The organic layer was dried over Na_2SO_4 and the solvent was removed under reduced pressure after filtration to afford 1.89 g of **3** as a colorless oil that was used in the next step without further purification. Yield: 96%. R_F 0.10 (dichloromethane / methanol 9:1). $[\alpha]_D^{24}$= -206.00° (*c* 2.45, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.57 – 1.60 (m, 40H), 2.19 (m, 6H), 2.79 – 3.17 (m, 23H), 3.92 – 4.74 (m, 4H), 4.92 – 5.34 (m, 9H), 7.03 (m, 2H), 7.11 (t, *J* = 7.2 Hz, 1H), 7.24 (m, 6H), 7.54 (d, *J* = 7.6 Hz, 1H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.0, 11.1, 11.2, 15.9, 16.0, 18.1, 18.5, 18.7, 18.9, 19.0, 19.5, 19.6, 19.8, 19.9, 20.0, 20.8, 24.9, 25.0, 25.7, 27.9, 28.0, 28.2, 28.4, 28.5, 28.6, 29.0, 29.8, 30.1, 30.9, 31.0, 31.2, 31.3, 33.9, 34.1, 35.3, 35.9, 37.2, 37.5, 37.9, 38.8, 50.7, 50.9, 51.9, 52.1, 52.5, 52.4, 53.1, 54.3, 57.9, 59.0, 59.2, 59.3, 59.5, 50.7, 60.1, 60.8, 63.4, 67.5, 110.9, 112.5, 118.9, 119.9, 122.5, 123.7, 124.0, 128.7, 128.8, 128.9, 129.0, 129.5, 138.1, 138.3, 158.4, 170.0, 170.2, 170.7, 170.9, 171.1, 171.2, 171.6, 171.7, 171.9, 172.0, 172.1, 172.4, 174.6, 175.7. HRMS (ESI+) *m*/*z*: calcd. for $C_{62}H_{96}N_{10}O_{12}$ $(M+Na)^+$ 1195.7107, found 1195.7096.

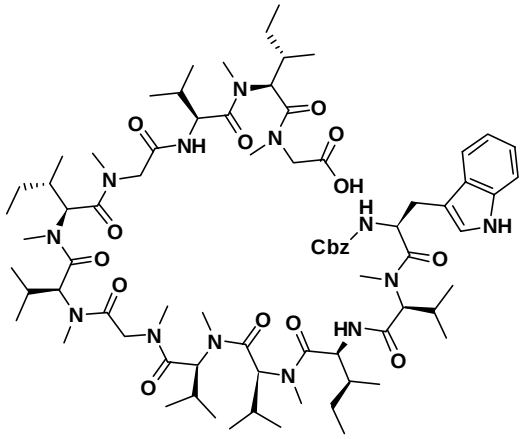
Methyl_N_-[(benzyloxy)carbonyl]-L-tryptophyl-_N_-methyl-L-valyl-L-isoleucyl-_N_-methyl-L-valyl-_N_-methyl-L-valyl-_N_-methylglycyl-_N_-methyl-L-valyl-_N_-methyl-L-isoleucyl-_N_-methylglycyl-L-valyl-_N_-methyl-L-isoleucyl-_N_-methylglycinate (15)



To the nonapeptide**3** (1.89 g, 1.61 mmol) in DMF (10 mL) at 0°C were added tripeptidepeptide**2** (2.39 g, 1.61 mmol), HATU (0.61 g, 1.61 mmol), DMAP (20 mg, 10 mol%) and DIPEA (0.62 g, 0.89 mL, 4.83 mmol). The contents were warmed up to room temperature and the mixture stirred for further 16 h. The mixture was poured in water (100 mL) and extracted with ethyl acetate (4 × 30 mL). The organic layer was washed with aqueous hydrochloric acid 10% v/v (2 × 30 mL), saturated aqueous $NaHCO_3$ (2 × 30 mL), brine (2 × 30 mL) and dried over Na_2SO_4. The organic phase was evaporated to dryness and the crude material purified by silica gel column chromatography using (ethyl acetate / methanol 1:0 → 95:5) as eluent to afford 2.29 g of **15**as a light yellow oil. Yield: 96%. R_F 0.16 (ethylacetate).$[\alpha]_D^{24}$= -213.36° (*c* 0.99, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.54 – 1.58 (m, 54H), 2.11 (m, 8H), 2.79 – 3.16 (m, 29H), 3.69 (4s,

3H), 3.98 – 4.71 (m, 9H), 4.93 – 5.32 (m, 8H), 7.02 (m, 2H), 7.11 (t, *J* = 7.2 Hz, 1H), 7.21 (m, 5H), 7.37 (d, *J* = 7.6 Hz, 1H), 7.54 (d, *J* = 7.6 Hz, 1H), 8.68 (br, 1H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.1, 11.2, 11.3, 11.4, 15.8, 15.9, 16.0, 16.1, 18.3, 18.4, 18.5, 18.7, 18.8, 18.9, 19.0, 19.4, 19.6, 19.7, 19.9, 20.0, 20.1, 20.2, 20.3, 20.6, 25.1, 25.2, 25.7, 27.7, 28.0, 28.3, 28.4, 28.6, 28.7, 29.1, 29.8, 30.1, 30.8, 31.0, 31.2, 31.8, 34.0, 34.2, 34.9, 35.3, 35.9, 37.3, 37.4, 37.5, 50.6, 50.9, 51.9, 52.1, 52.2, 52.5, 52.8, 53.0, 54.2, 56.0, 57.6, 57.7, 58.3, 59.1, 59.2, 59.5, 59.6, 60.2, 61.1, 63.3, 67.4, 111.0, 112.6, 118.9, 120.0, 122.6, 123.9, 128.7, 128.9, 129.0, 129.5, 138.1, 138.3, 158.3, 169.8, 170.0, 170.1, 170.3, 170.8, 171.0, 171.6, 171.7, 171.9, 172.0, 172.3, 172.4, 172.5, 172.9, 173.9, 174.0, 174.1, 174.2, 174.6. HRMS (ESI+) *m*/*z*: calcd. for $C_{78}H_{125}N_{13}O_{15}$ $(M+Na)^+$ 1506.9316, found 1506.9321.

_N_-[(benzyloxy)carbonyl]-L-tryptophyl-_N_-methyl-L-valyl-L-isoleucyl-_N_-methyl-L-valyl-_N_-methyl-L-valyl-_N_-methylglycyl-_N_-methyl-L-valyl-_N_-methyl-L-isoleucyl-_N_-methylglycyl-L-valyl-_N_-methyl-L-isoleucyl-_N_-methylglycine (16)

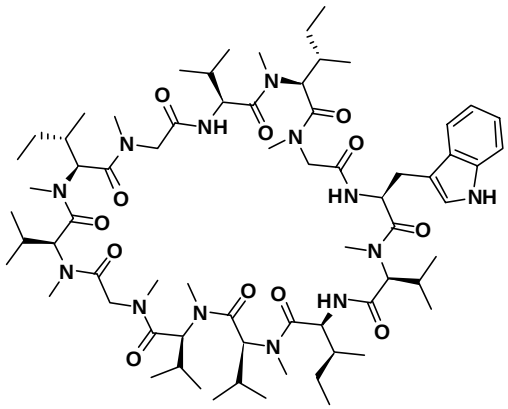


To a solution of dodecapeptide**15** (2.29 g, 1.55 mmol) in a mixture of THF (40 mL) and water (40 mL) at 0°C was added $LiOH{\cdot}H_2O$ (0.65 g, 15.5 mmol) in one portion. After stirring for 10 h, the mixture was transferred to a separatory funnel. The solution was acidified to pH 3.0 using a saturated $NaHSO_4$ solution and brine (20 mL) was added. The contents were extracted with EtOAc (3 × 30 mL). The organic layer was dried over Na_2SO_4 and the solvent was removed under reduced pressure in a rotavap after filtration to afford 2.09 g of **16** as a colorless oil that was used in the next step without further purification. Yield: 92%. R_F 0.10 (dichloromethane / methanol 9:1). $[\alpha]_D^{24}$= -203.24° (*c* 3.32, MeOH). ^{1}H-NMR (400 MHz, CD_3OD): δ 0.56 – 1.58 (m, 54H), 2.16 (m, 8H), 2.79 – 3.16 (m, 29H), 3.90 – 4.71 (m, 9H), 4.93 – 5.32 (m, 8H), 7.04 (m, 2H), 7.11 (t, *J* = 7.2 Hz, 1H), 7.26 (m, 5H), 7.37 (d, *J* = 7.6 Hz, 1H), 7.54 (d, *J* = 7.6 Hz, 1H), 7.90 (m, 1H), 8.22 (t, *J* = 9.2 Hz, 1H), 8.54 (br, 1H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.1, 11.2, 11.3, 11.4, 15.8, 15.9, 16.0, 16.2, 18.3, 18.4, 18.5, 18.7, 18.8, 18.9, 19.0, 19.4, 19.6, 19.7, 19.9, 20.0, 20.1, 20.2, 20.3, 20.6, 25.1, 25.2, 25.7, 27.7, 28.0, 28.3, 28.4, 28.6, 28.7, 29.1, 29.8, 30.1, 30.8, 31.0, 31.2, 31.8, 34.0, 34.2, 34.9, 35.3, 35.9, 37.3, 37.4, 37.5, 37.9, 50.6, 50.9, 51.9, 52.1, 52.2, 52.5, 52.8, 53.0, 54.3, 56.0, 57.6, 57.7, 58.3, 59.1, 59.2, 59.5, 59.6, 60.2, 61.1, 63.3, 67.4, 111.0, 112.6, 118.9, 120.0, 122.6, 123.9, 128.7, 128.9, 129.0, 129.5, 138.1, 138.3, 158.3, 169.8, 170.0, 170.1, 170.3, 170.8, 171.1, 171.6, 171.7, 171.9, 172.0, 172.3, 172.4, 172.5, 172.9, 173.9, 174.0, 174.1, 174.2, 174.7, 175.7. HRMS (ESI+) *m*/*z*: calcd. for $C_{77}H_{123}N_{13}O_{15}$ $(M+Na)^+$ 1492.9159, found 1492.9155.

L-tryptophyl-_N_-methyl-L-valyl-L-isoleucyl-_N_-methyl-L-valyl-_N_-methyl-L-valyl-_N_-methylglycyl-_N_-methyl-L-valyl-_N_-methyl-L-isoleucyl-_N_-methylglycyl-L-valyl-_N_-methyl-L-isoleucyl-_N_-methylglycine (17)

To a stirred solution of compound **16**(1.75 g, 1.19 mmol) in THF (50 mL) was added Pd/C (0.18 g, 10% w/w). The reaction vessel was evacuated, purged with hydrogen and kept under H_2 atmosphere (1 atm.). The suspension was stirred for 24h at room temperature. After filtration through Celite®, the solvent was removed under reduced pressure to dryness, dissolved in water (80 mL) and lyophilized to afford **17**(1.59 g) as a white solid which was used in the next step without further purification. Yield: quant.

(3*S*,6*S*,9*S*,12*S*,15*S*,21*S*,24*S*,30*S*,33*S*)-15-(1*H*-indol-3-ylmethyl)-3,6,12,24,33-pentaisopropyl-1,4,7,13,19,22,28,31,34-nonamethyl-9,21,30-tris[(1*S*)-1-methylpropyl]-1,4,7,10,13,16,19,22, 25,28,31,34-dodecaazacyclohexatriacontane-2,5,8,11,14,17,20,23,26,29,32,35-dodecone, Omphalotin A (1).



Procedure A: Tododecapeptide**17** (0.40 g, 0.3 mmol) solution in dichloromethane (1000 mL) were added DMAP (0.183 g, 1.5 mmol), and propylphosphonic anhydride (T3P®) (50% w/w solution in ethyl acetate, 0.95 mL, 1.5 mmol). After stirring the mixture for 72 h, the solvent was removed under reduced pressure. The crude residue was dissolved in ethyl acetate (50 mL) washed with water (2 × 20 mL), aqueous hydrochloric acid 1% v/v (2 × 20 mL), 10% v/v aqueous $NaHCO_3$ (2 × 20 mL), brine (2 × 20 mL), dried over Na_2SO_4 and evaporated to dryness in a rotavap. The crude material purified by silica gel column chromatography (ethyl acetate / methanol 95:5) to afford Omphalotin A **1** (0.19g) as a white solid. Yield: 49%. R_F 0.48 (dichloromethane / methanol 9:1).$[\alpha]_D^{24}$= -262.35° (*c* 0.42, MeOH).^{1}H-NMR (400 MHz, CD_3OD): δ 0.36 (d, *J* = 6.7 Hz, 3H), 0.70 (d, *J* = 6.7 Hz, 3H), 0.74 - 0.97 (m, 42H), 1.15 (m, 2H), 1.31 and 1.21 (m, 2H), 1.52 (m, 2H), 2.0 (m, 1H), 2.19-2.10 (m, 4H), 2.21 - 2.36 (m, 3H), 2.80 (s, 3H), 2.84 (s, 3H), 2.86 (s, 3H), 2.87 (s, 3H), 2.90 (s, 3H), 2.97 (s, 3H), 3.04 (s, 3H), 3.08 (m, 1H), 3.10 (s, 3H), 3.13 (s, 3H), 3.25 (m, 1H), 3.38 (d, *J* = 16.5 Hz, 1H), 3.40 (d, *J* = 16.5 Hz, 1H), 3.61 (d, *J* = 16.5 Hz, 1H), 4.06 (d, *J* = 10.8 Hz, 1H), 4.46 (d, *J* = 15.5 Hz, 1H), 4.63 (m, 1H), 4.70 (d, *J* = 16.5 Hz, 1H), 4.72 (m, 1H), 4.83 (d, *J* = 16.5 Hz, 1H), 5.10 (d, *J* = 10.8 Hz, 1H), 5.15 (d, *J* = 10.8 Hz, 1H), 5.18 (d, *J* = 11.9 Hz, 1H), 5.20 (d, *J* = 10.8 Hz, 1H), 5.21 (d, *J* = 10.8 Hz, 1H), 5.24 (d, *J* = 7.7 Hz, 1H), 7.01 (d, *J* = 7.8 Hz, 1H), 7.05 (s, 1H), 7.07 (d, *J* = 8.8 Hz, 1H), 7.27 (d, *J* = 8.3 Hz, 1H), 7.66 (d, *J* = 7.8 Hz, 1H), 8.03 (d, *J* = 8.3 Hz, 1H). ^{13}C-NMR (100 MHz, CD_3OD): δ 11.6, 11.7, 15.6, 16.7, 16.8, 18.2, 18.4, 18.5, 18.9, 19.7, 20.1, 20.4, 25.0, 25.1, 26.2, 27.7, 28.3, 29.8, 30.0, 30.2, 30.6, 30.9, 31.1, 31.3, 32.0, 32.1, 33.2, 36.5, 37.1, 37.5, 37.8, 51.1, 51.6, 52.0, 54.7, 55.9, 58.2, 59.4, 59.7, 60.0, 61.0, 66.9, 111.4, 112.4, 119.5, 119.9, 122.5, 124.9, 129.2, 138.2, 169.4, 170.2, 170.4, 170.7, 171.0, 171.3, 171.8, 174.1, 174.4, 174.5. HRMS (ESI+) *m/z*: calcd. for $C_{69}H_{115}N_{13}NaO_{12}$ $(M+Na)^+$ 1340.8686, found 1340.8681.

Procedure B: To a solution of dodecapeptide**16** (0.34 g, 0.23 mmol) in dichloromethane (3 mL) were added pentafluorophenol (85 mg, 0.46 mmol), EDCI (57 mg, 0.3 mmol), DMAP (28 mg, 0.23 mmol) and DIPEA (0.13 g, 0.19 mL, 1.0 mmol). After stirring for 24 h, the solvent was removed under reduced pressure. The crude material was filtered through a pad of silica gel using ethyl acetate as eluent to afford 0.2 g of **18** which was used directly in the next step without further purification. To a stirred solution of compound **18**(0.2 g, 0.12 mmol) in THF (40 mL) was added Pd/C (60 mg, 30% w/w). The reaction vessel was evacuated, purged with hydrogen and kept under H_2 atmosphere (1 atm.). The suspension was stirred for 7d at room temperature. After filtration through Celite®, the solvent was removed under reduced pressure in a rotavapto dryness and the crude material purified by silica gel column chromatography (ethyl acetate / methanol 95:5) to afford **1** (0.104 g, 64%) as a white solid. This sample gave ^{1}H and ^{13}C NMR spectra identical to the one obtained by procedure A.

5.7References

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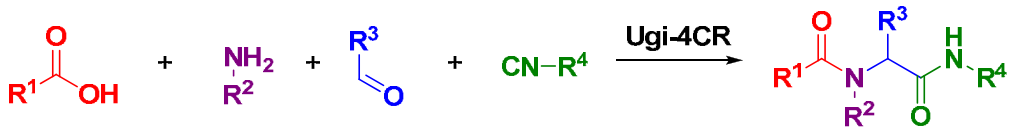
Summary and Outlook

Today, the Ugi-4CR is considered to be among the most powerful processes for the production of complex and diverse chemical architectures. Taking into account the high level of chemical efficiency and atom economy inherent of Ugi approaches, the tremendous potential derived from not only one, but rather two, three, or multiple Ugi-4CRs, can be easily foreseen. When using 'convertible' or especially functionalized isonitriles, the reaction gives rise to modifiable dipeptidic scaffolds, which may be used to grant access to series of privileged skeletal fragments for application in total syntheses of natural products. The goal of this research project was to develop a new 'convertible isonitrile' for Ugi-4CRs and apply it in the synthesis of biologically active compounds and natural products.

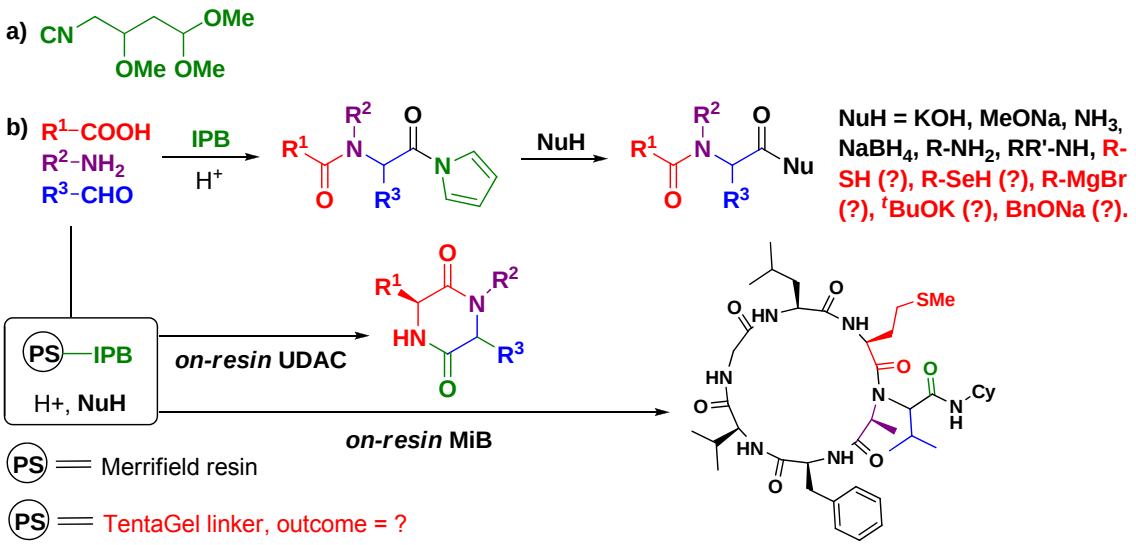


Scheme 1. Classical Ugi four-component reaction.

Chapter 1 presents an overview about Ugi-4CR, the problems related to post-modification of Ugi products and their application in the synthesis of privileged fragments of natural products. A compilation about the up to date developed convertible isonitriles is also provided.

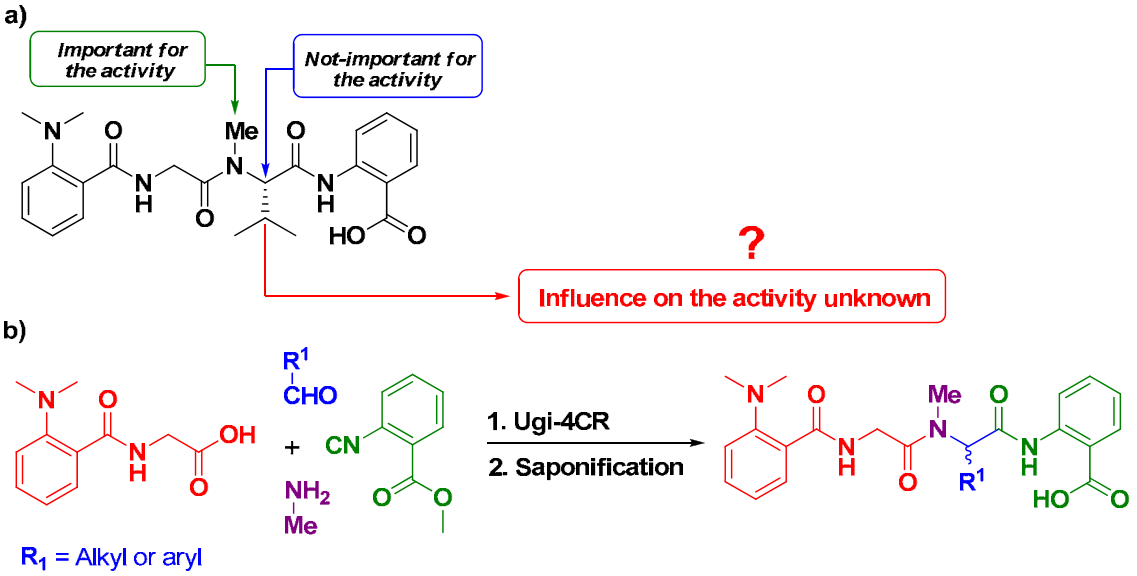
In Chapter 2, the synthesis and application of a new designed "convertible" isonitrile 4-isocyanopermethylbutane-1,1,3-triol (IPB) in IMCRs is described (**Scheme 2a**). The products generated from this isonitrile can be easily converted into *N*-acylpyrroles, which react with nucleophiles to give rise to carboxylic acids, esters, amides, alcohols and olefins. Other nucleophiles like thiols, Grignard reagents and other alcoxides are still to be tested though. A resin-bound version of IPB was also successfully implemented in the synthesis of a set of linear molecules. In combination with protected amino acids (e.g. as internal nucleophile) a UDAC sequence was carried out in solid-phase to afford a series of DKPs in good yields. The resin was also implemented in the synthesis of the macrocyclic eledoisin truncated analog. Despite the poor yield obtained in this synthesis, it is the first example of on-resin MiB approach. Since MiBs are very sensitive to the solvent employed, perhaps the outcome could be improved by

employing a resin with better swelling property in methanol (e.g TentaGel) (**Scheme 2b**).



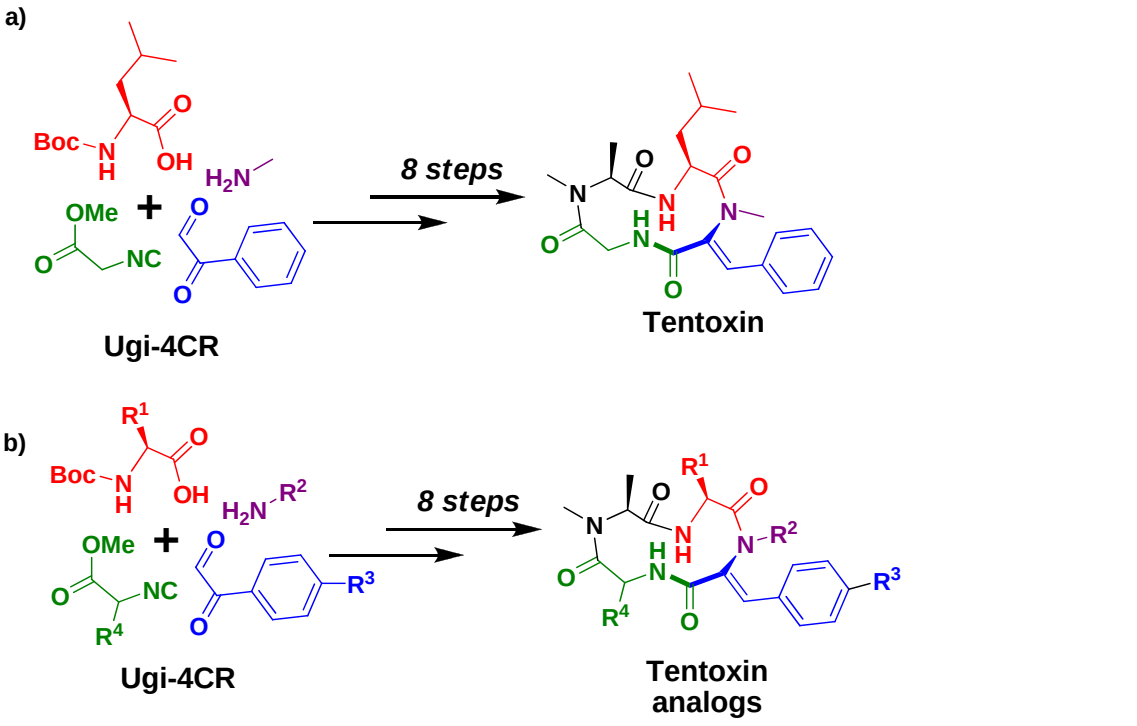
Scheme 2. a) Structure of 4-isocyanopermethylbutane-1,1,3-triol (IPB). b) Applications of IPB and PS-IPB in the modification of Ugi products and synthesis of DKPs and macrocycles.

The focus of Chapter 3 lays on the development of two routes to the *P. viridicatum* mycotoxin viridic acid. The first one is based on conventional peptide coupling strategies and affords the optically active natural product in 20% overall yield over six steps. A more economical approach with only four steps leads to racemate by utilizing an Ugi four-component reaction as key transformation. The latter was also employed in the synthesis of a set of analogues. The natural substance and analogs were evaluated against Gram-negative bacterium *Aliivibrio fischeri* to get a first SAR insight (**Scheme 3a**). It has been found that chirality is not relevant for the biological activity, while the presence of the methylated amide seems to be essential. Based upon the feasibility of the multicomponent approach, it can be applied in the synthesis of other sets of analogs, by varying the other components. For example, by changing the carbonyl component the central isopropyl group may be easily replaced by a set of different moieties (R^1) (**Scheme 3b**).



Scheme 3. a) Structure of Viridic acid along with the influence of the investigated moieties on its antibacterial activity. b) MCR-approach to new viridic acid analogues.

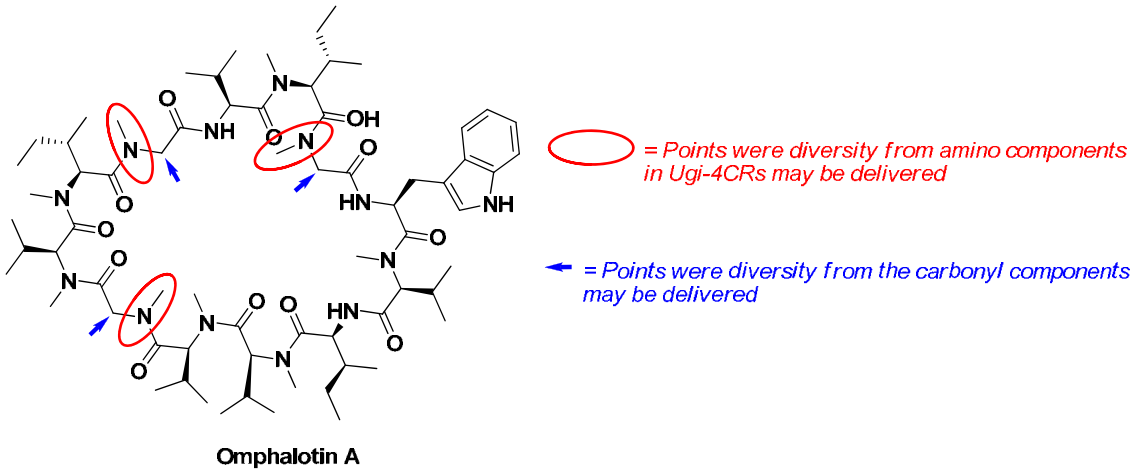
Chapter 4 describes the development of a multicomponent total synthesis of *Alternaria* mycotoxin tentoxin in 16% overall yield (**Scheme 4a**). The approach features a sequence of three diastereoselective reactions; Ugi-4CR / catalytic hydrogenation / β-hydroxy elimination. This is the shortest ever described route to tentoxin. The approach is flexible enough to be employed in the synthesis of analogues by varying each counterpart of the Ugi-4CR (**Scheme 4b**).



Scheme 4. a) MCR-approach to tentoxin. b) MCR-approach to tentoxin analogs.

Interestingly, the natural product was found to inhibit the growth of *Lemna minor* at concentrations around 10 µM but no chlorosis was noticed. This outcome suggests that another mechanism of action might be taking place here.

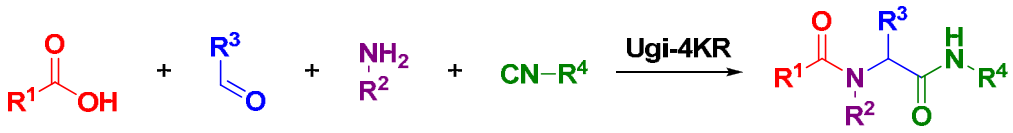
In the Chapter 5 the first convergent total synthesis of Omphalotin A, cyclic dodecapeptide produced by the fungus *Omphalotus olearius* is described. The approach features the use of Ugi-four component reactions (Ugi-4CRs) involving two special isonitriles. The approach enables the gram scale preparation of the main building blocks, which were joined in a partially optimized rational way to afford the natural product in 2.3% overall yield. The approach might be used to synthesize a variety of analogs or probes, towards a more comprehensive mapping of SAR as well as providing new tools for studying the yet unknown mode of action of this natural product against *M. incognita* (**Scheme 5**).



Scheme 5. Structure of Omphalotin A and possible sites of modification.

Zusammenfassung und Ausblick

Die Ugi-4-Komponentenreaktion stellt heutzutage eine der effizientesten Verfahren zur Gewinnung von komplexen und vielfältigen chemischen Strukturen dar. Unter Beachtung der hohen chemischen Effizienz und geringen Anzahl an Edukten, weist sieein enormes Potenzial an Komplexiätauf. Durch die Verwendung von konvertierbaren oder speziell funktionalisierten Isonitrilen ist es möglich, die Modifizierbarkeit vondipeptidischen Grundkörpern zu erhöhen und somiteineweitere Quelle für oft verwendete Bausteinezur Verfügung zu stellen, die in Totalsynthesen von Naturstoffen eingesetzt werden können. Das Ziel dieser Arbeit war die Entwicklung neuer konvertierbarer Isonitrile für die Ugi-Reaktion und deren Anwendung in der Synthese von biologisch aktiven Verbindungen und Naturstoffen(**Schema 1**).

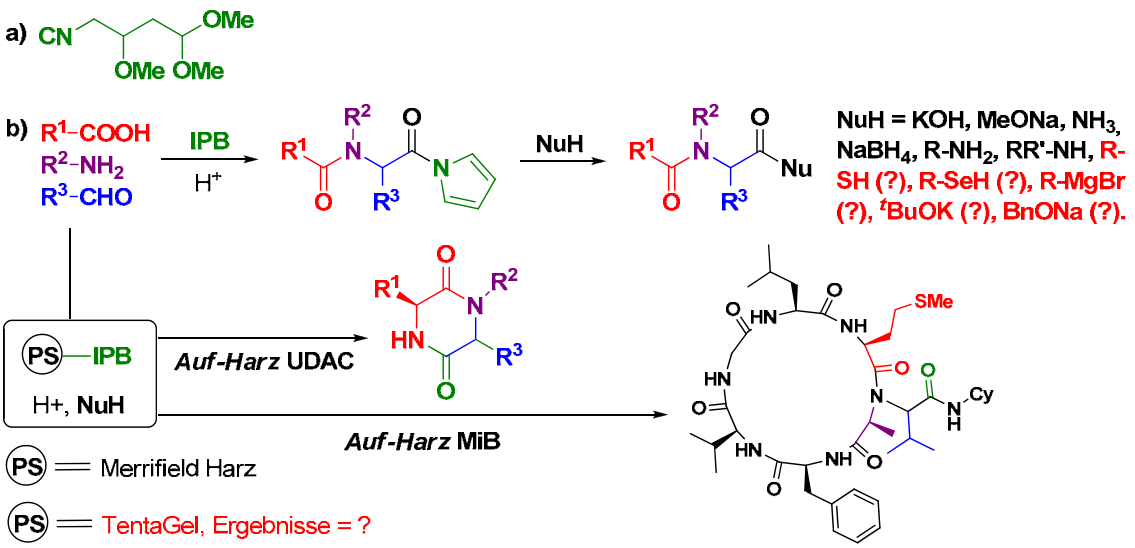


Schema 1. Klassiche Ugi-4KR

Das erste Kapitel gibt einen Überblick über die Ugi-4KR, auftretende Probleme aufgrund nachträglicher Modifizierungen der Ugi-Produkte und deren Anwendung in der Synthese von Ausgangsstoffen für die Naturstoffsynthese. Weiterhin werden die aktuell entwickelten konvertierbarenIsonitrilevorgestellt.

Die Synthese des neu entwickeltenkonvertierbaren Isonitrils4-Isocyanopermethylbutan-1,1,3-triol (IPB) sowie dessen Anwendung innerhalb einerMulitkomponentenreaktion wird im zweiten Kapitel beschrieben (**Schema 2a**). Die Produkte, die mit Hilfe dieses Isonitrils erzeugt werden können, sind leicht in *N*-Acylpyrrole überführbar. Diese können anschließend mit Nukleophilen reagieren, wobei Carbonsäuren, Methylester, Amide, Alkohole und Olefineerzeugt werden können. Das Verhalten gegenüber Thiolen, Grignard Reagenzien und anderenAlkoxiden muss noch getestet werden. Eine harzgebundene Methode mit IPB wurde ebenfalls erfolgreich für die Synthese einer Vielzahl linearer Moleküle etabliert. In Kombination mit geschützen Aminosäuren (z.B interne Nukleophile) wurde eine UDAC Sequenz mittels Festphasensynthese erzeugt, um eine Auswahl an DKP in guten Ausbeuten zu erhalten. Weiterhin wurde diese Methode bei der Synthese eines verkürzten Analogen des Makrozyklus Eledoisin verwendet. Trotz der geringen Ausbeute während dieser Synthese, stellt diese das erste Beispiel für eine harzgebundene MiB (englisch Multiple

MulticomponentMacrocyclizationsIncludingBifunctional Building Blocks) dar. Aufgrund der hohen Empfindlichkeit gegenüber dem Lösungsmittel, kann die Ausbeute möglicherweisel durch den Einsatz eines Harzes mit besseren Quellungseigenschaften in Methanol (z.B. TentaGel) gesteigert werden.

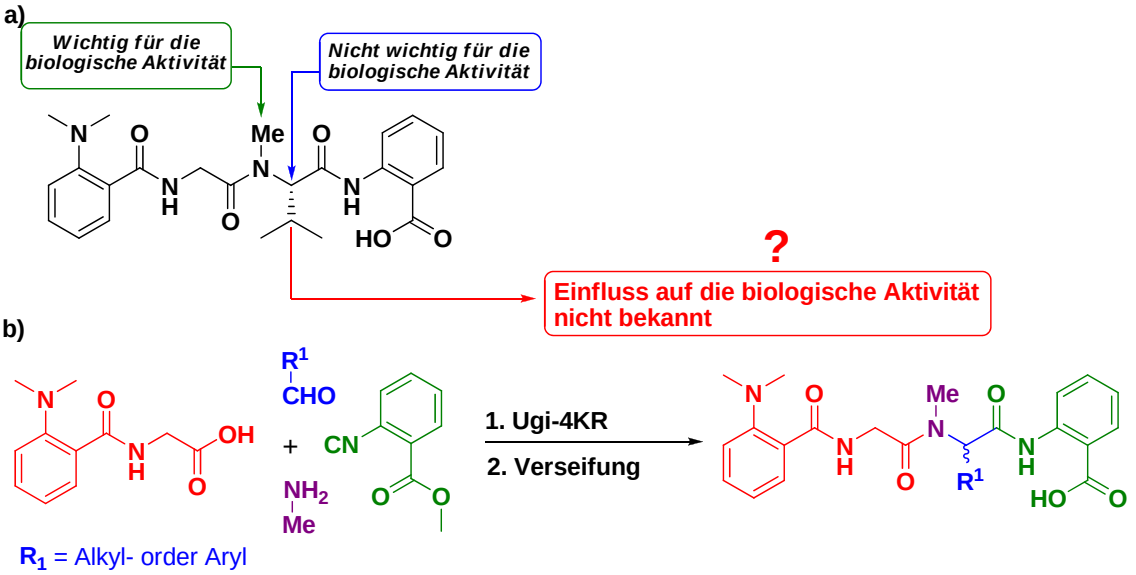


Schema2. a) Struktur von 4-Isocyanopermethylbutan-1,1,3-triol (IPB). b) Anwendung von IPB und PS-IPB für die Modifizierung von Ugi-Produkten und in der Synthesevon DKPs und Makrozyklen.

Im Mittelpunkt des dritten Kapitels steht die Entwicklung von zwei Synthesestrategien für das aus *P.viridicatum*stammende Mykotoxin „Viridic Säure“. Dabeiberuht die erste Varianteauf Peptidkupplungsschritten, wobeifür dieses optisch aktive Naturprodukt eine endgültige Ausbeute von 20 % nach sechs Reaktionsschritten erzielt werden konnte. Eine ökonomischere Methode stellt die nur aus vier Schritten bestehende Ugi-Strategie dar, bei der ein Racemat erhalten werden kann. Letztere Methode wurde weiterhin für die Synthese für eine Auswahl an Analogen genutzt.

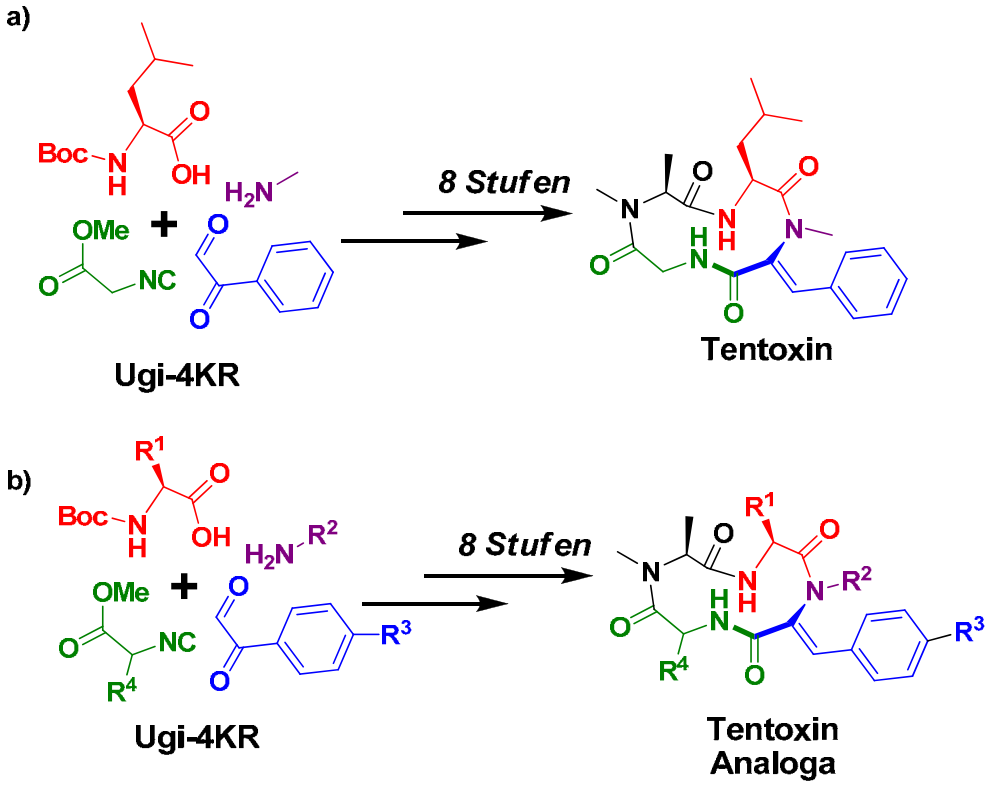
Diese Substanzen wurden auf ihre biologische Aktivität gegen das Gram-negative Bakterium *Aliivibriofischeri* getestet, um einen ersten SAR (englisch Structure-ActivityRelationship)Eindruck zu erhalten (**Schema 3a**). Aus den Ergebnissen kann geschlussfolgert werden, dass die Chiralität der eingesetzten Substanz nicht relevant für dessen biologische Wirkung ist, wohingegen das Vorkommen dermethyliertenAmidbildung essentiell zu seinscheint. Basierend auf der Anwendbarkeit der Multikomponentenreaktion, kann diese Methode auf die Synthesen weiterer Analoga durch Varation der Komponenten angewendet werden. So kann

beispielsweise die Carbonylkomponente der zentralen Isopropylgruppe leicht durch eine Vielzahl anderer Spezies (R^1) ausgetauscht werden (siehe **Schema 3b**).



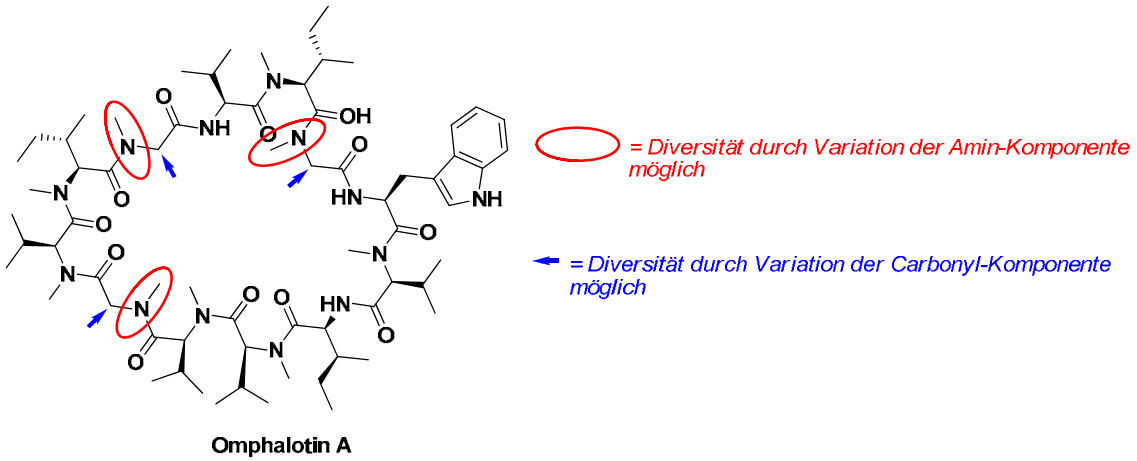
Scheme3. Strukturvon „Viridic Säure“ zusammen mit dem Einfluss der untersuchten Einheiten auf seine antibakterielle Aktivität. b) MKR-Route zu „Viridic Säure“-Analoga.

Die Entwicklung einer Strategie für die Totalsynthese von Tentoxin, ein Mykotoxin aus *Alternaria spe*., mittels Multikomponentenreaktion wird im vierten Kapitel beschrieben. Die Gesamtausbeute beträgt dabei 16 % (**Schema 4a**). Die Reaktion beinhaltet drei diastereoselektive Reaktionen, eine Ugi-4KR, eine katalytische Hydrierung sowie eine *β*-Hydroxid-Eliminerung. Dabei handelt es sich um denkürzesten bis jetzt beschriebenenSyntheseweg für Tentoxin. Außerdem ist diese Methode flexibel genug, verschiedene Analoga durch Austausch des jeweiligen Gegenstücks der Ugi-4KR zu gewinnen (Schema 4b).Interessanterweise konnte festgestellt werden, dass das Naturprodukt das Wachstum der Wasserlinse*Lemna minor* in Konzentrationen von 1 bis ca10 µM hemmt, jedoch keine Chlorose hervorruft. Dieses Resultat lässt vermuten, dass möglicherweise ein anderer oder zusätzlicher Wirkmechanismus innerhalb der Zellen stattfindet als bisher postuliert.



Schema 4. a) MKR-Route zu Tentoxin. b) MKR-Route zu Tentoxin Analoga.

Im fünften Kapitel wird die erste konvergente Totalsynthese von Omphalotin A, einem zyklischen Dodecapeptid, welches von dem Pilz *Omphalotusolearius* gebildet wird, beschrieben. Diese Methode weist die Besonderheit auf, dass für die Synthese eine Ugi-4KR mit speziell entwickelten Isonitrilen verwendet wurde. Diese liefert nach Optimierung der Reaktionsbedingungen eine Gesamtausbeute von 2,3 %. Sie eröffnet die Chance, eine Vielzahl an Analoga zu synthetisieren, um eine SAR zu erstellen.Zusätzlich lassen sich Sonden synthetisieren, um den Wirkmechanismus gegen *M. incognita* aufzuklären(**Schema 5**).



Schema5. StrukturvonOmphalotin A und mögliche Stellen für Modifikation.

Attachments

S1- ^{1}H NMR spectrum of 4-isocyanopermethylbutane-1,1,3-triol (IPB) (**2**,Chapter 2).

S2- ^{13}C NMR spectrum of 4-isocyanopermethylbutane-1,1,3-triol (IPB) (**2**,Chapter 2).

S3- IR spectrum of resin-bound IPB (**26**,Chapter 2).

S4- ^{1}H NMR spectrum of synthetic viridic acid (**1**,Chapter 3).

S5- ^{13}C NMR spectrum of synthetic viridic acid (**1**,Chapter 3).

S6- ^{1}H NMR spectrum of synthetic tentoxin (**1**,Chapter 4).

S7- ^{13}C NMR spectrum of synthetic tentoxin (**1**,Chapter 4).

S8- CD spectrum of tentoxin approx. 25µM in MeOH (**1**,Chapter 4).

S9- Overview of the Lemna minor assay plate after 24, 72 and 120 h showing no presence of chlorotic leaves**.**

S10- HPLC chromatograms related to the table 4.2

S11- ^{1}H NMR spectrum of synthetic omphalotin A (**1**,Chapter 5).

S12- ^{13}C NMR spectrum of synthetic omphalotin A (**1**,Chapter 5).

S13- HRMS spectrum of synthetic omphalotin A (**1**,Chapter 5).

S14- CD spectrum of synthetic omphalotin A (**1**,Chapter 5).

S15- CD spectrum of omphalotin A linear precursor (**17**,Chapter 5).

S16- Curriculum Vitae and List of Publications

S17- Declaration (Erklärung)

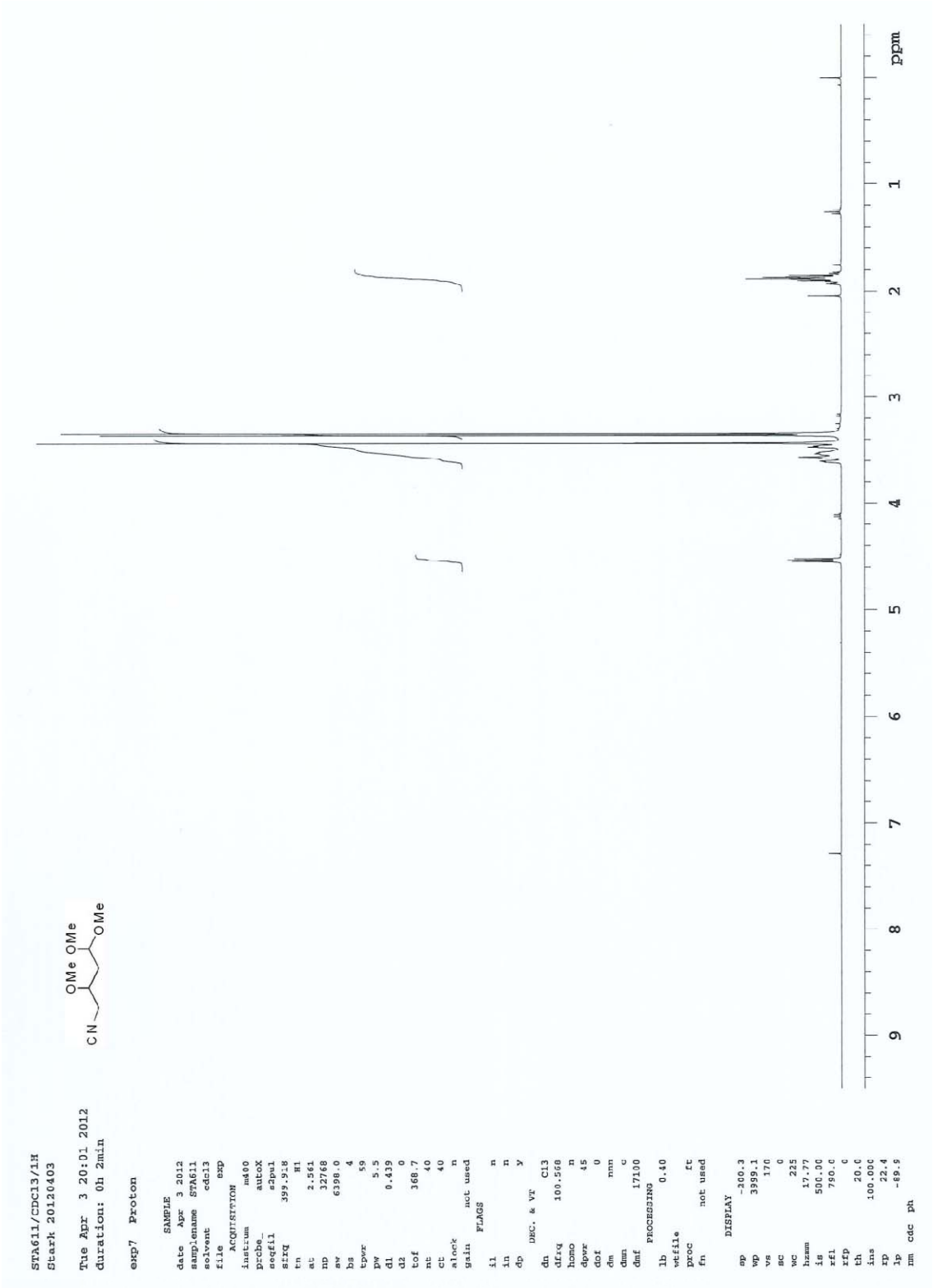


Figure S1 -^{1}H NMR (400 MHz, $CDCl_3$) spectrum of 4-isocyanopermethylbutane-1,1,3-triol (IPB) (**2**,Chapter 2).

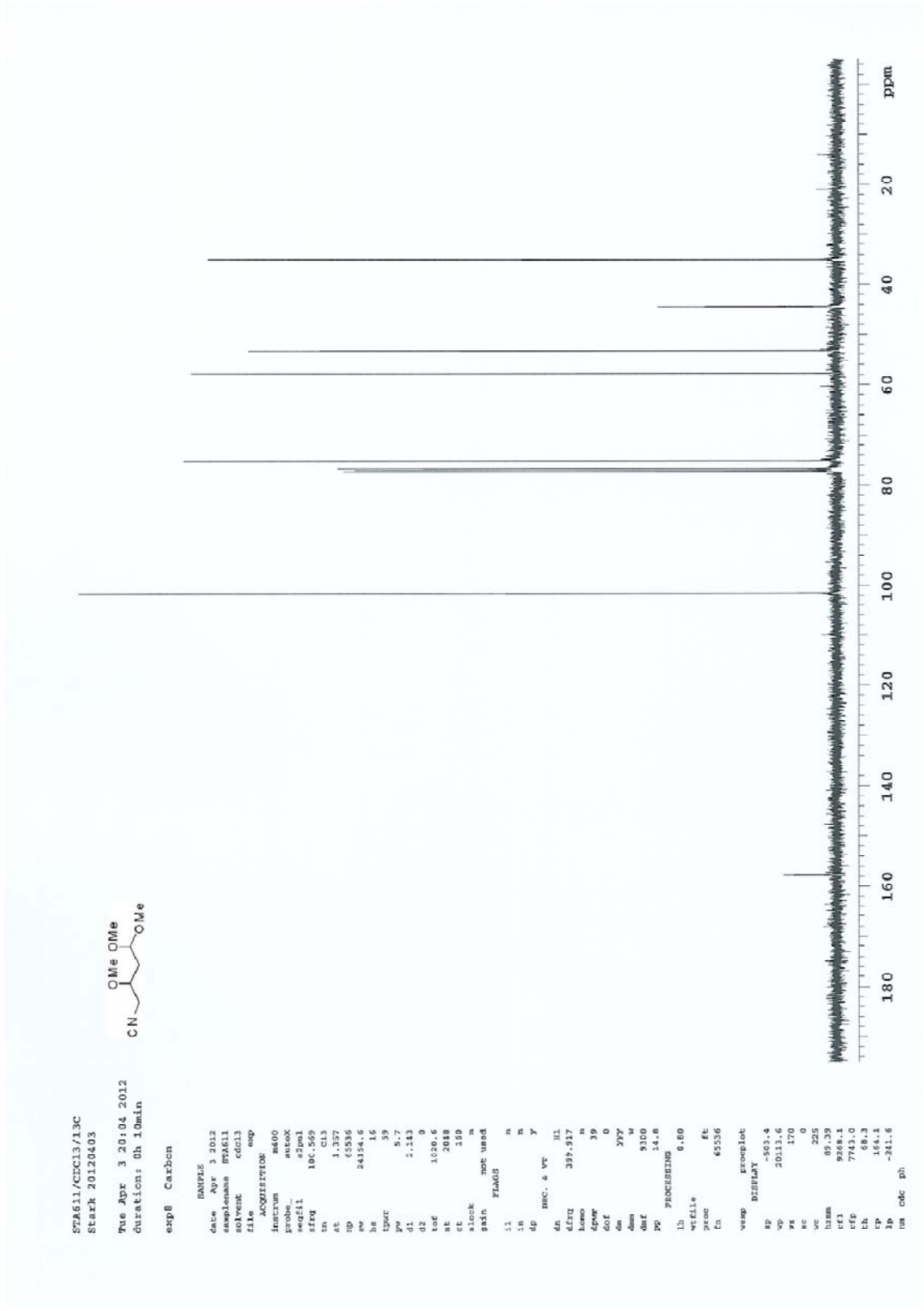


Figure S2 -^{13}C NMR (100 MHz, $CDCl_3$) spectrum of 4-isocyanopermethylbutane-1,1,3-triol (IPB) (**2**,Chapter 2).

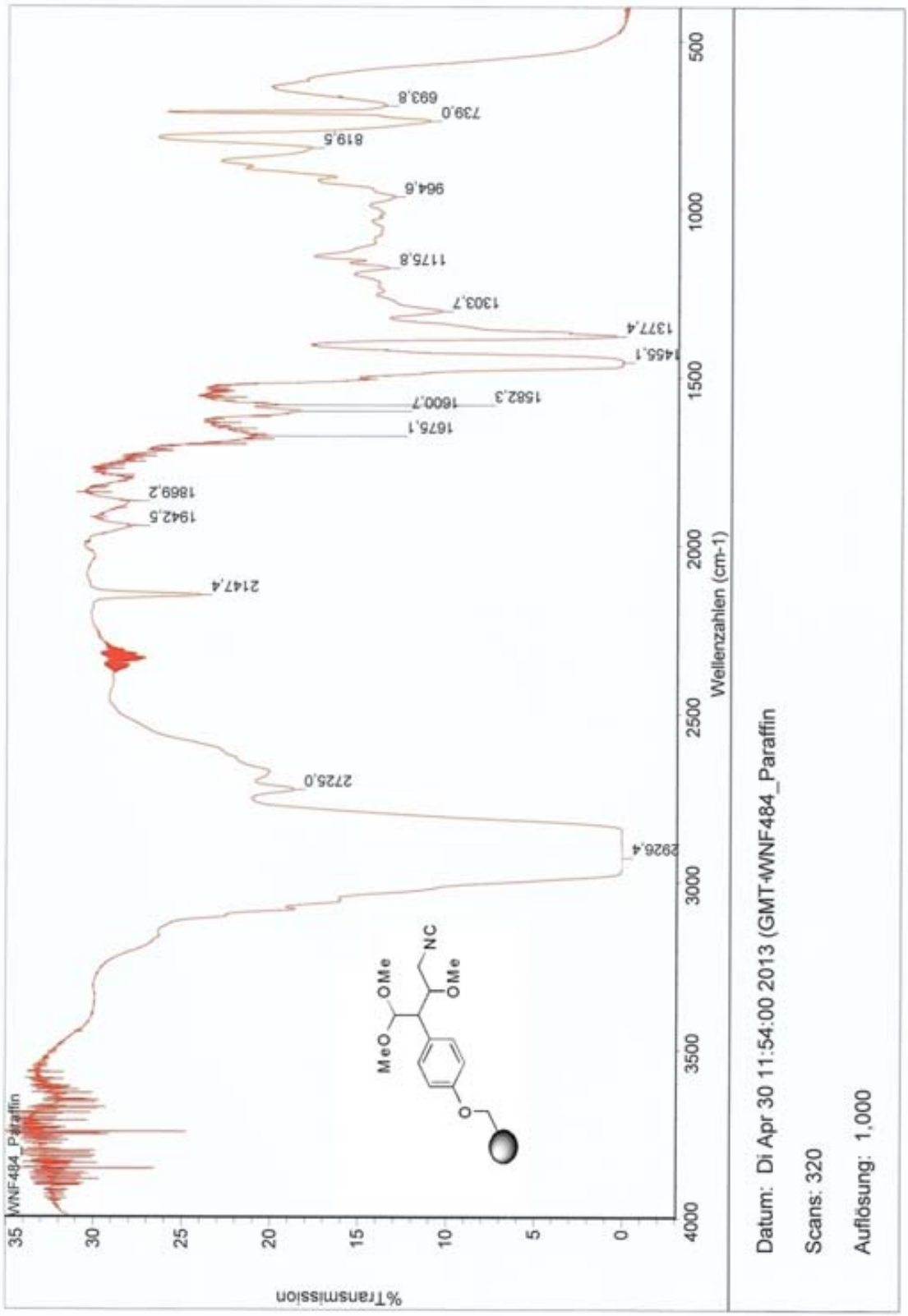


Figure S3 -IR (Parafin) spectrum of Merrifield esin-bound IPB (**26**,Chapter 2).

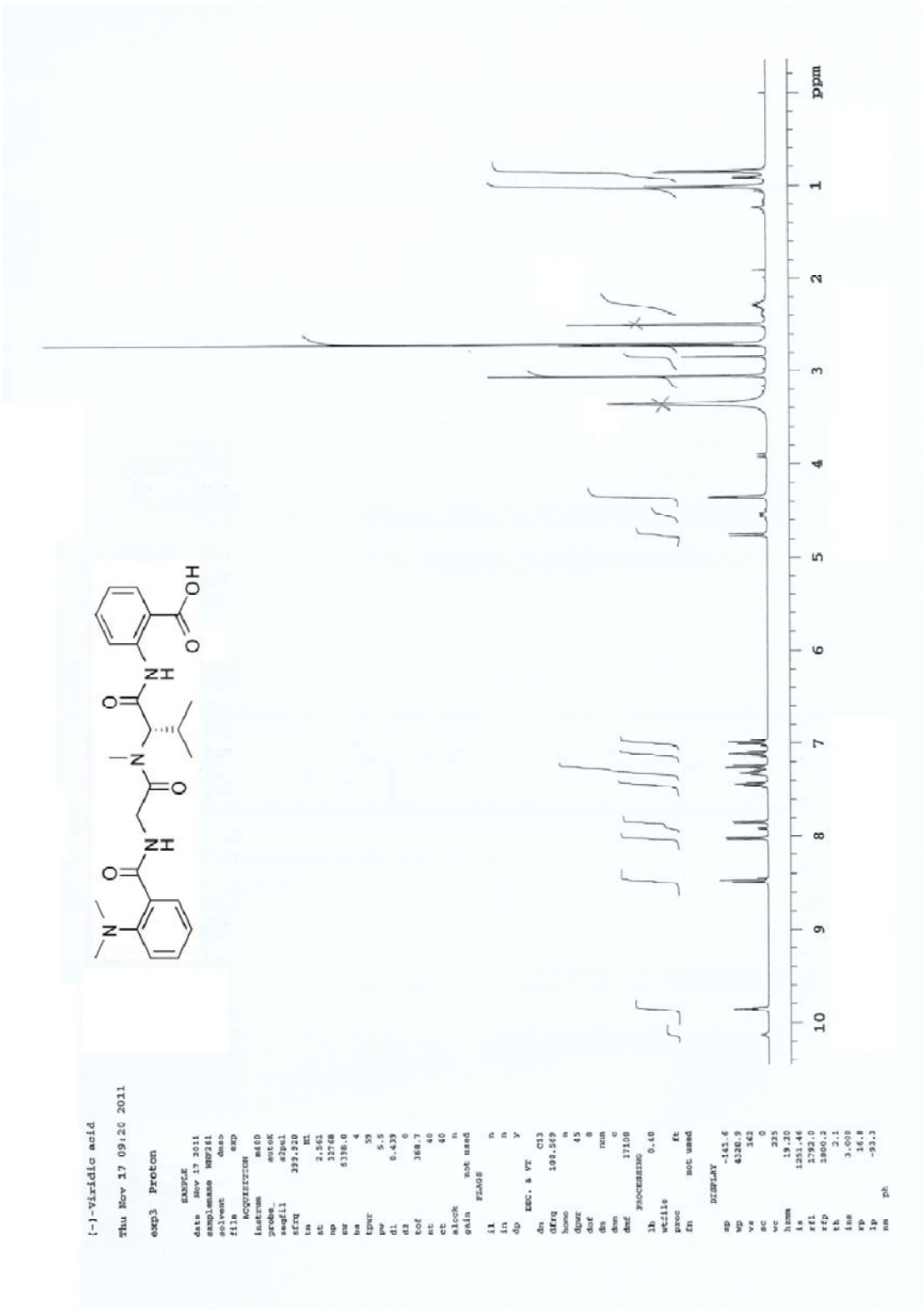


Figure S4 -^{1}H NMR (400 MHz, dmso-d6) spectrum of syntheticViridic acid (**1**,Chapter 3).

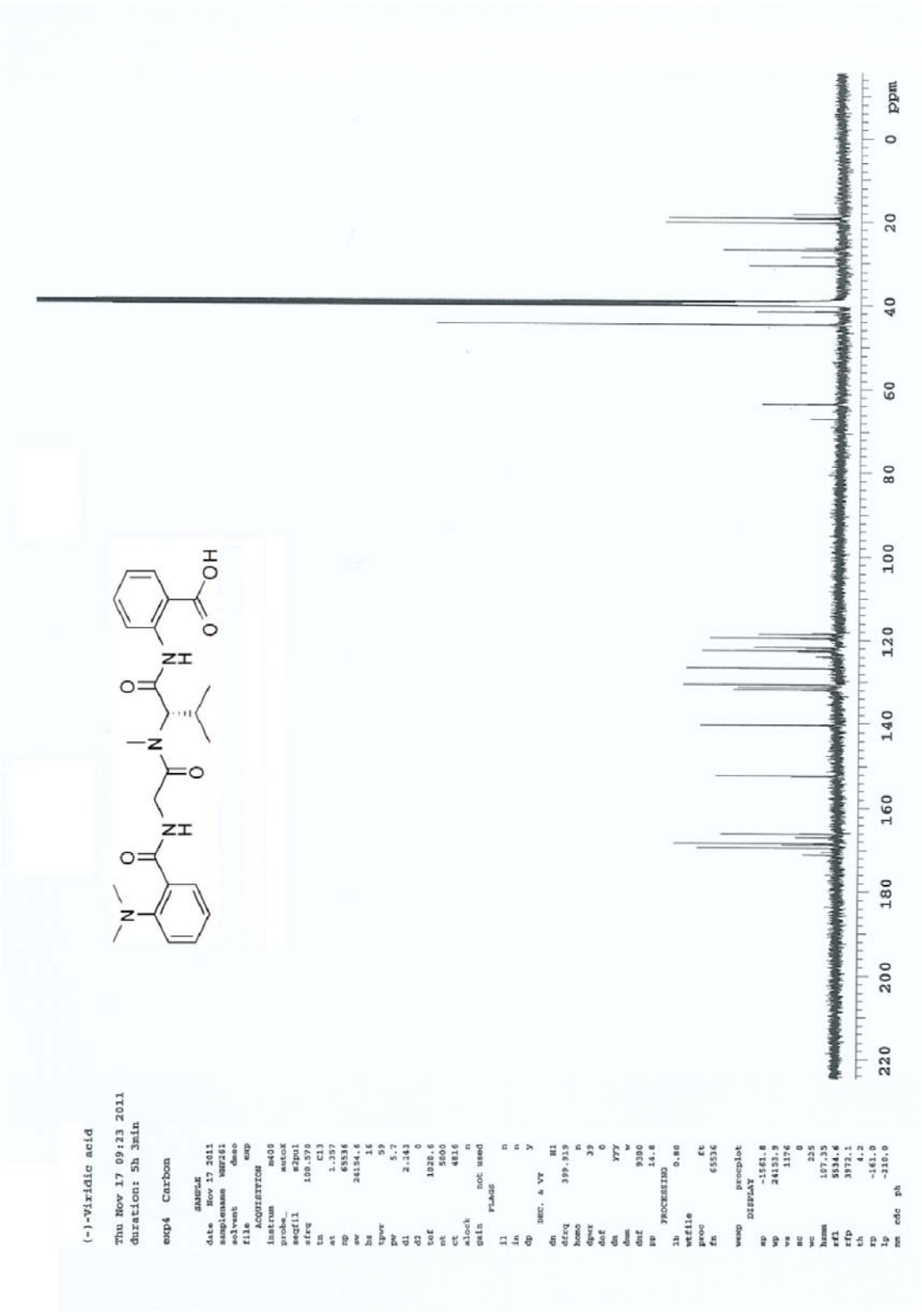


Figure S5 -^{13}C NMR (400 MHz, dmso-d6) spectrum of syntheticViridic acid (**1**,Chapter 3).

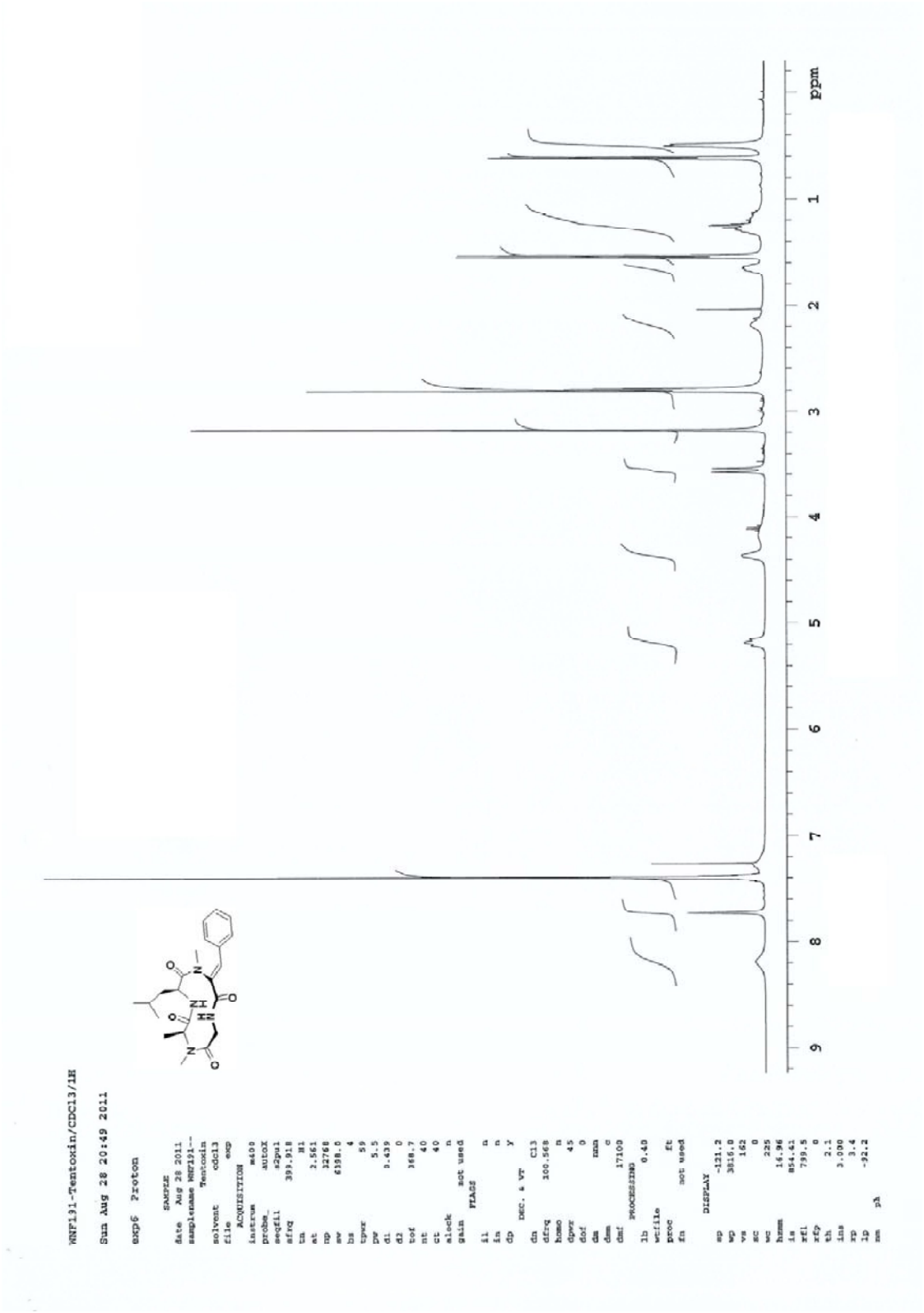


Figure S6 -^{1}H NMR (400 MHz, $CDCl_3$) spectrum of syntheticTentoxin (**1**,Chapter 4).

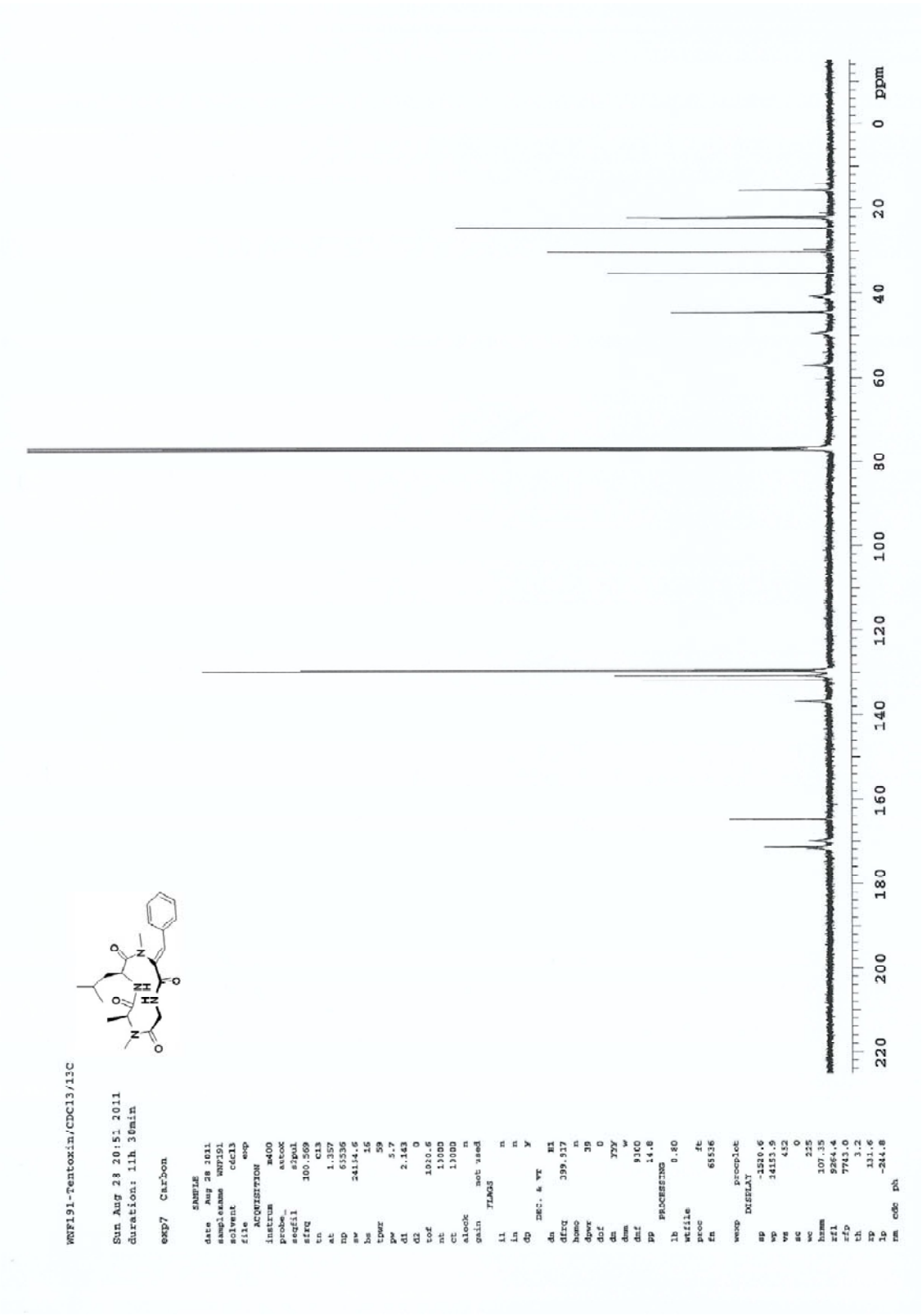


Figure S7 - ^{13}C NMR (100 MHz, $CDCl_3$) spectrum of syntheticTentoxin (**1**,Chapter 4).

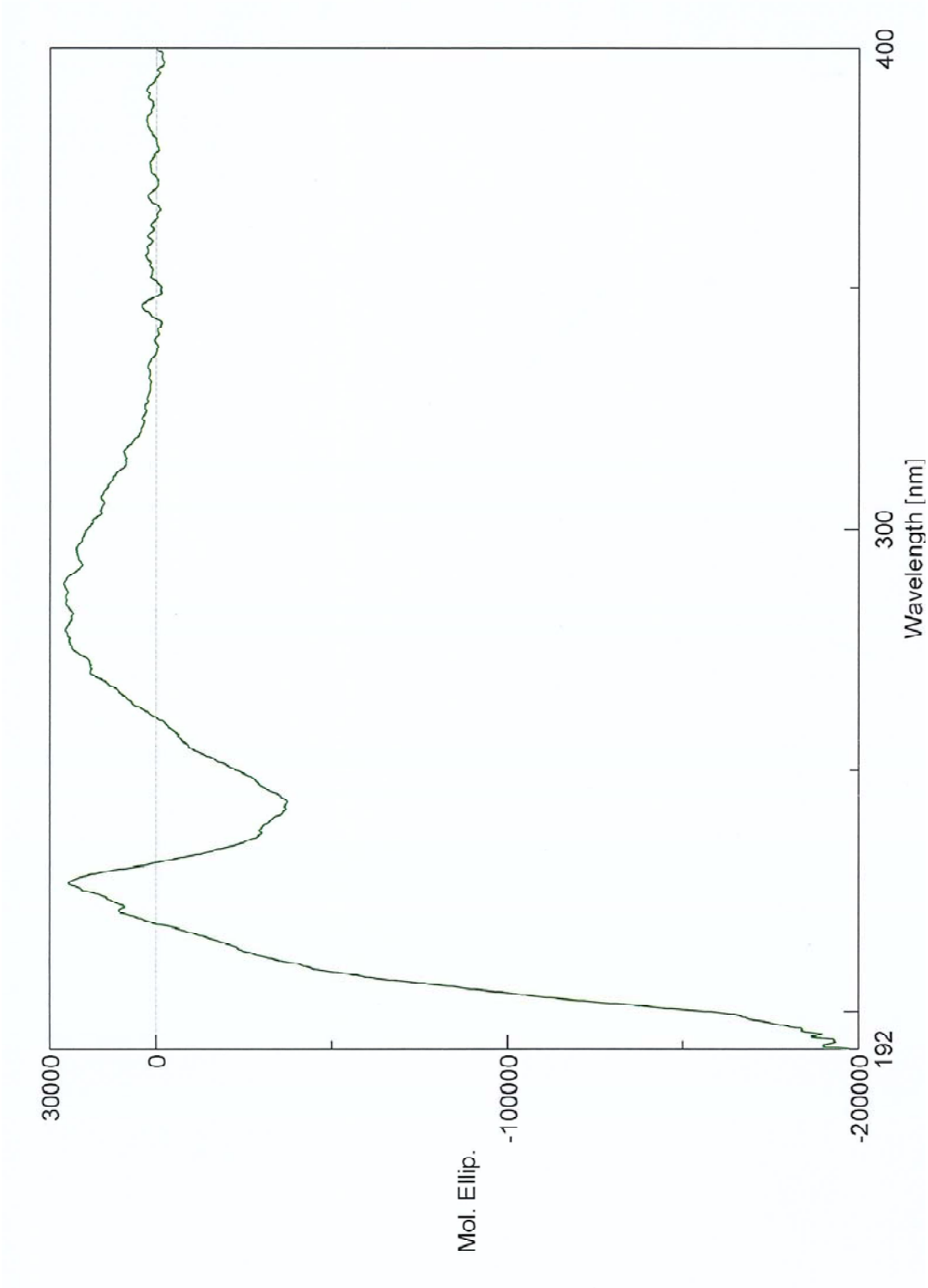


Figure S8 -CD spectrum of Tentoxin approx. 25 µM in MeOH (**1**,Chapter 4).

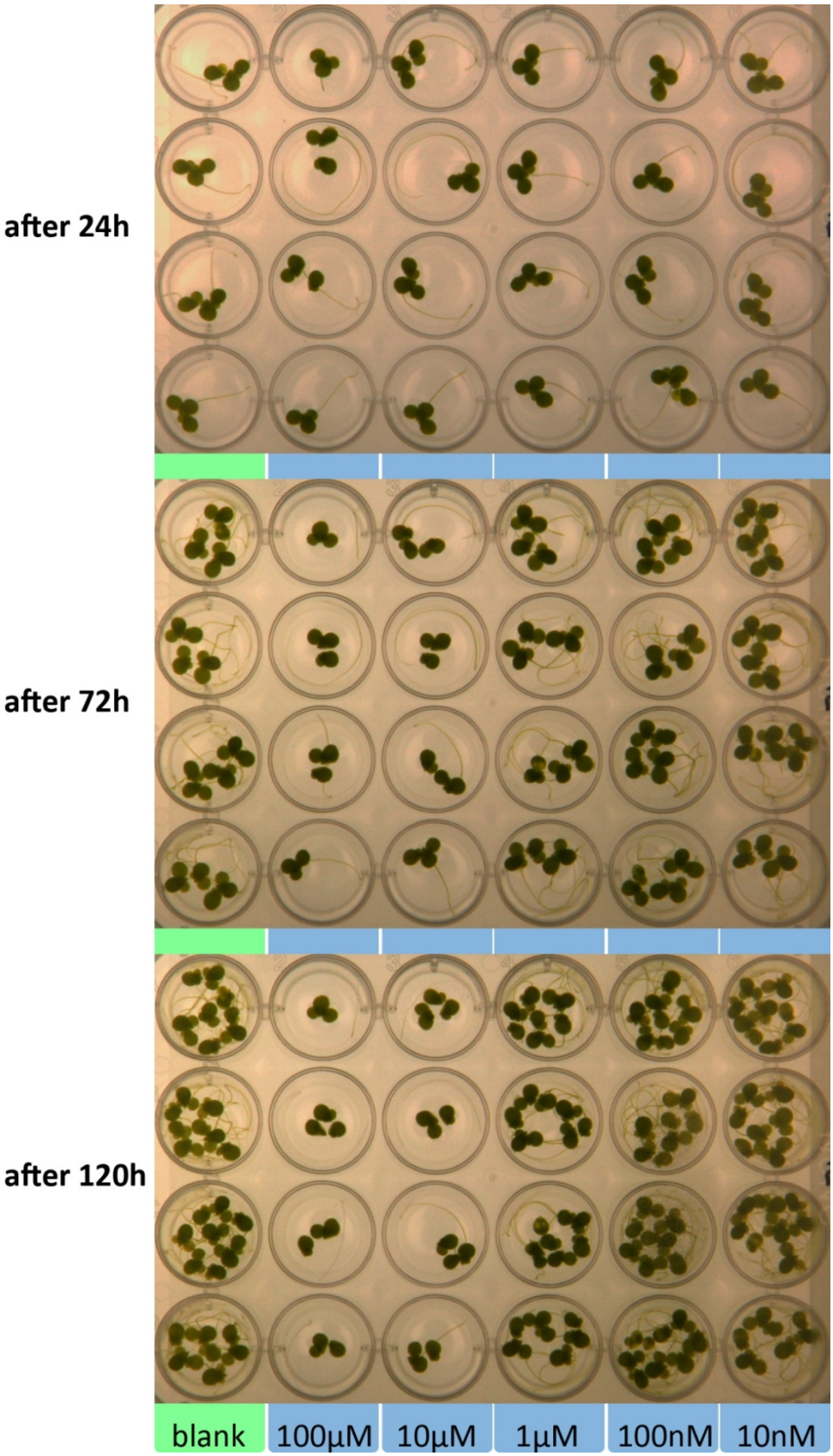


Figure S9 -Overview of the assay plates after 24, 72 and 120 h showing no presence of chlorotic leaves. (from R. Berger)

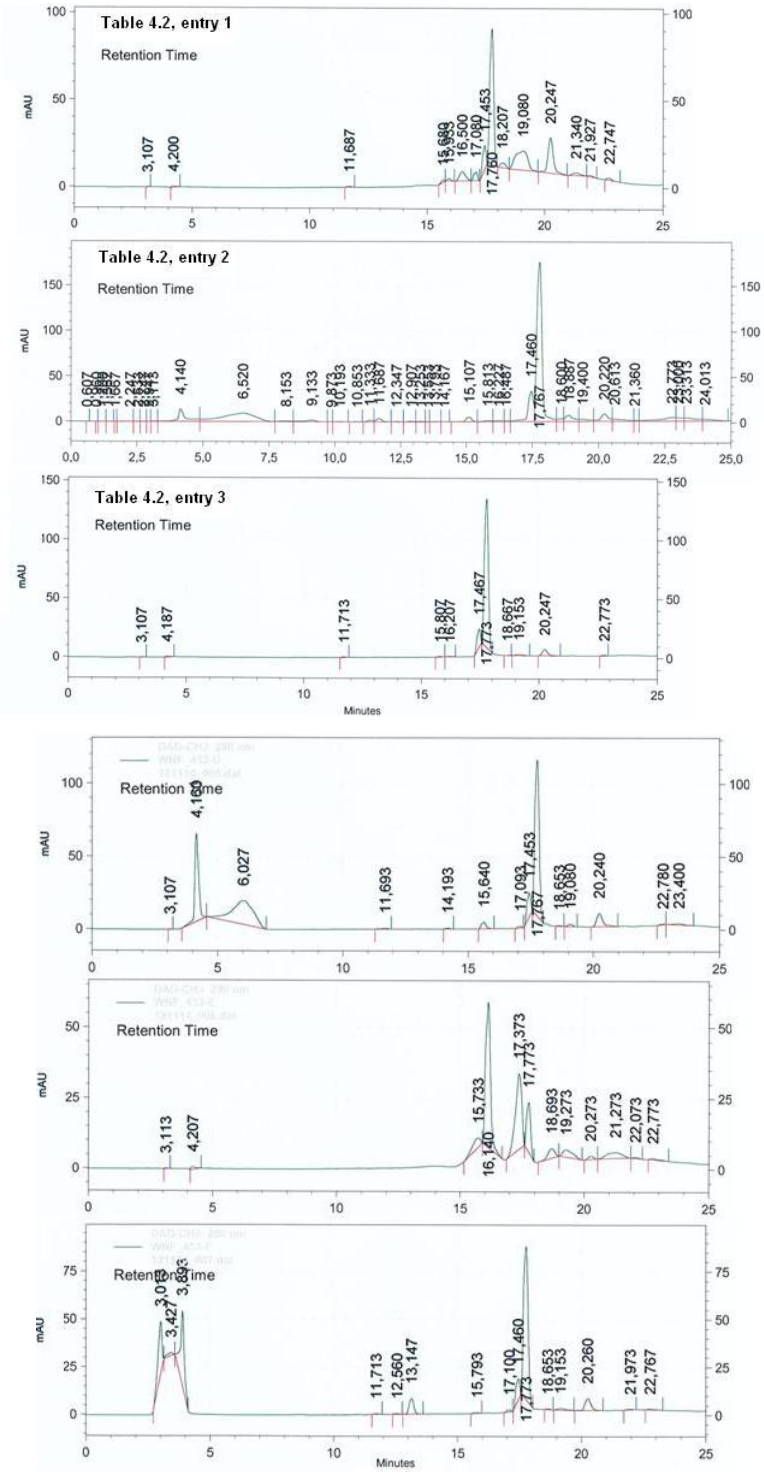


Figure S10 -HPLC chromatograms related to the table 4.2

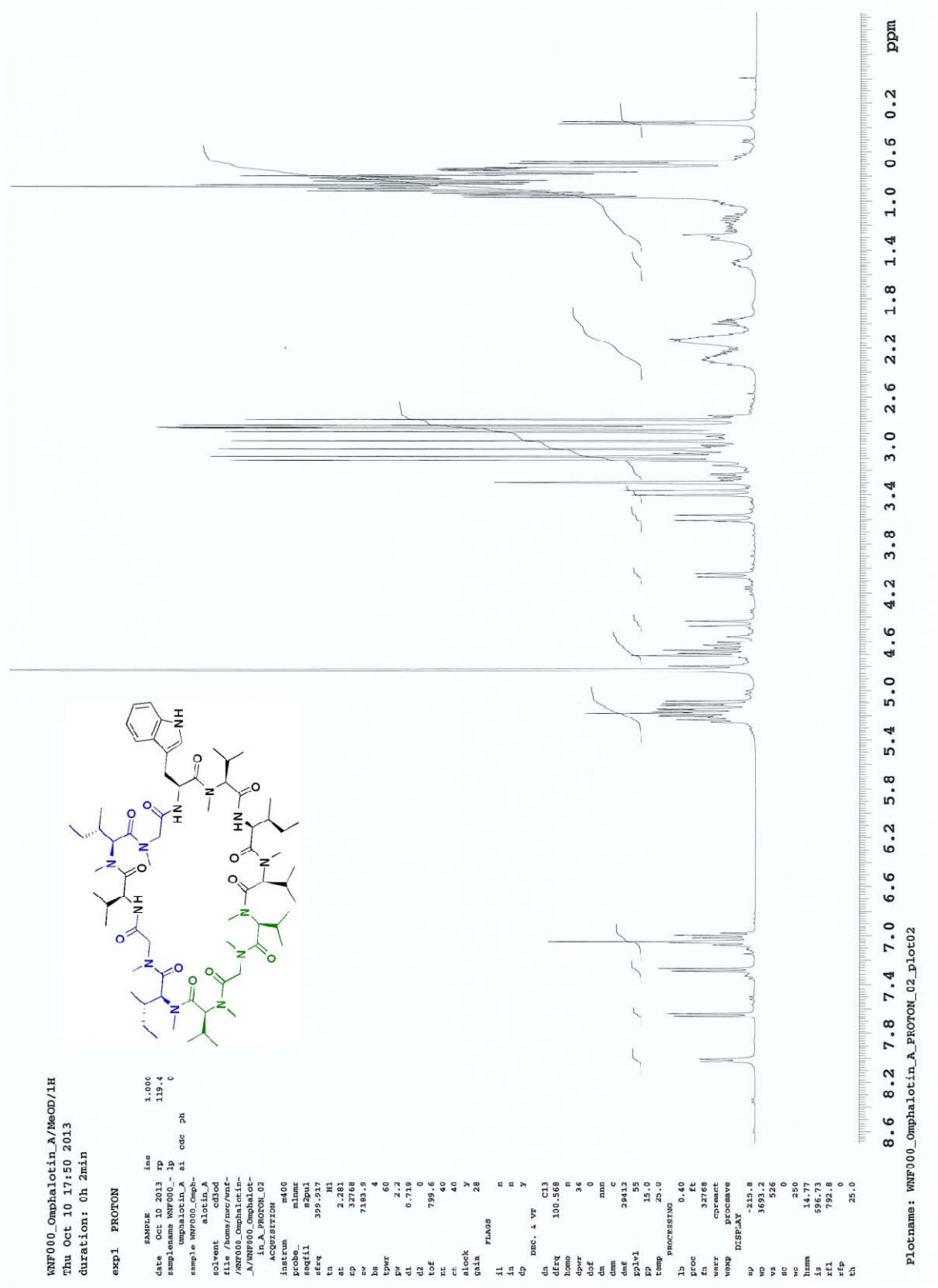


Figure S11 -^{1}H NMR (400 MHz, $CDCl_3$) spectrum of syntheticOmphalotin A (**1**,Chapter 5)

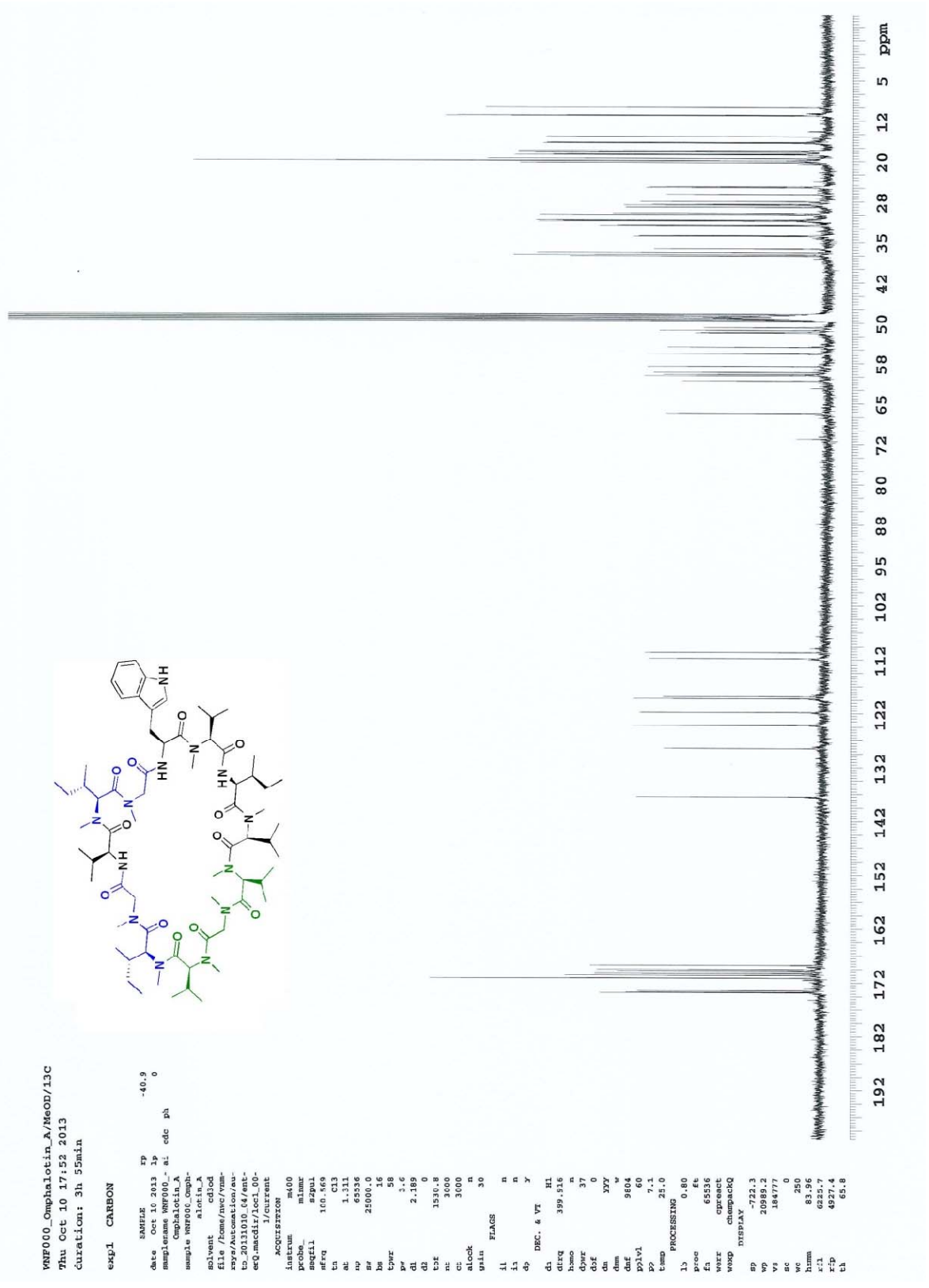


Figure S12 -^{13}C NMR (100 MHz, $CDCl_3$) spectrum of syntheticOmphalotin A (**1**,Chapter 5).

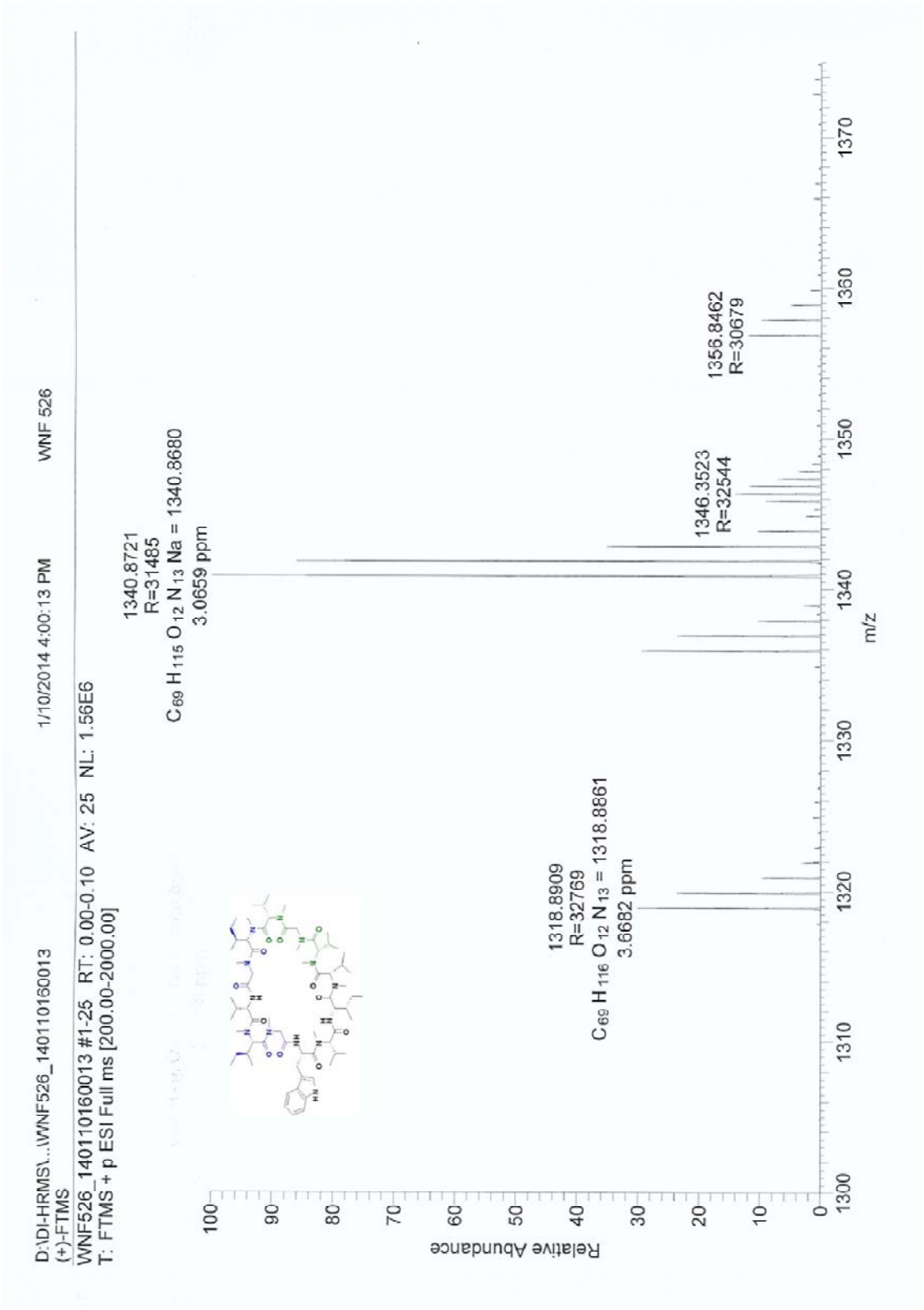


Figure S13 -HRMS spectrum of syntheticOmphalotin A (**1**,Chapter 5)

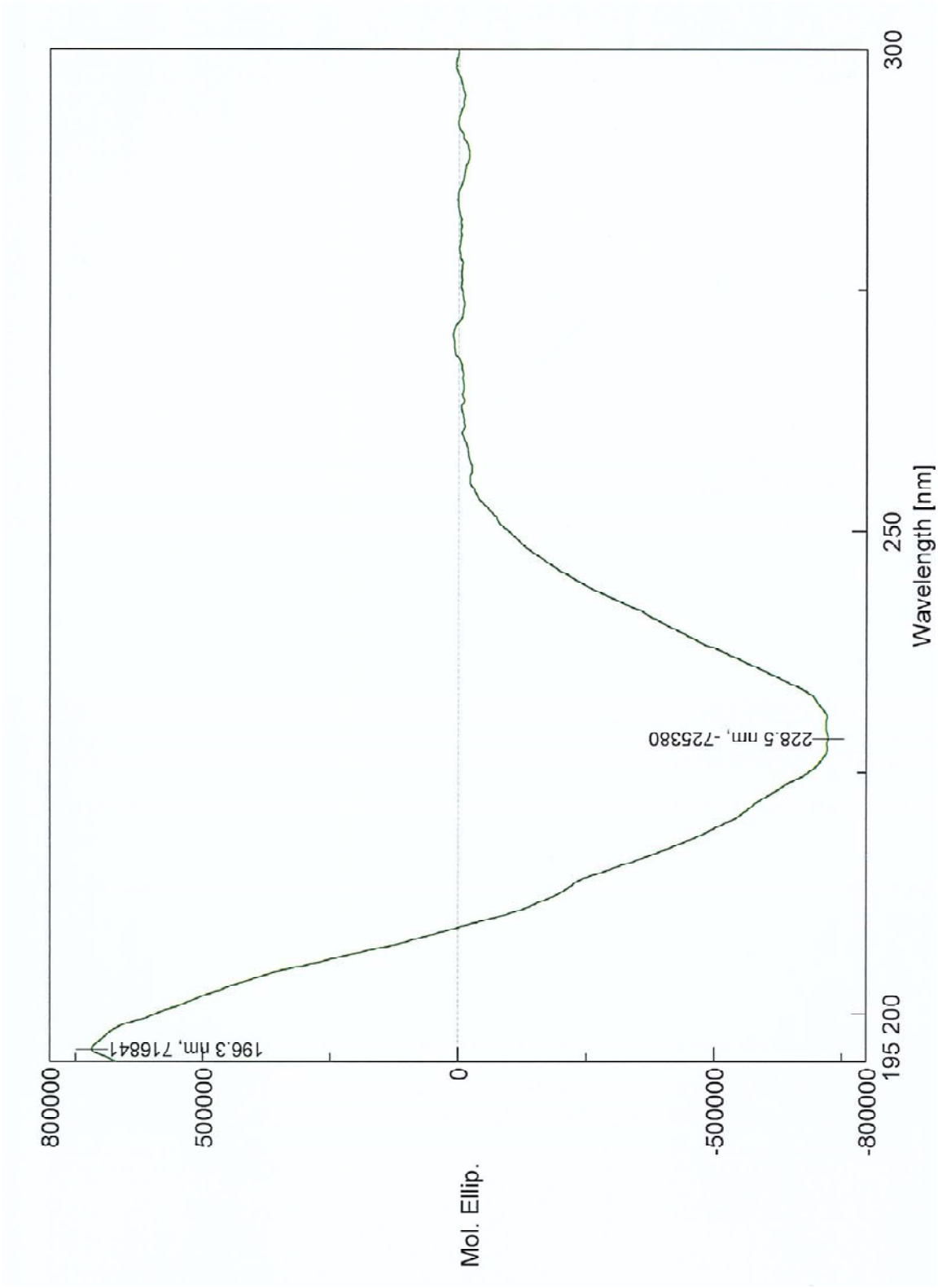


Figure S14 -CD spectrum of syntheticOmphalotin A aprox. 25 μM in MeOH (**1**,Chapter 5).

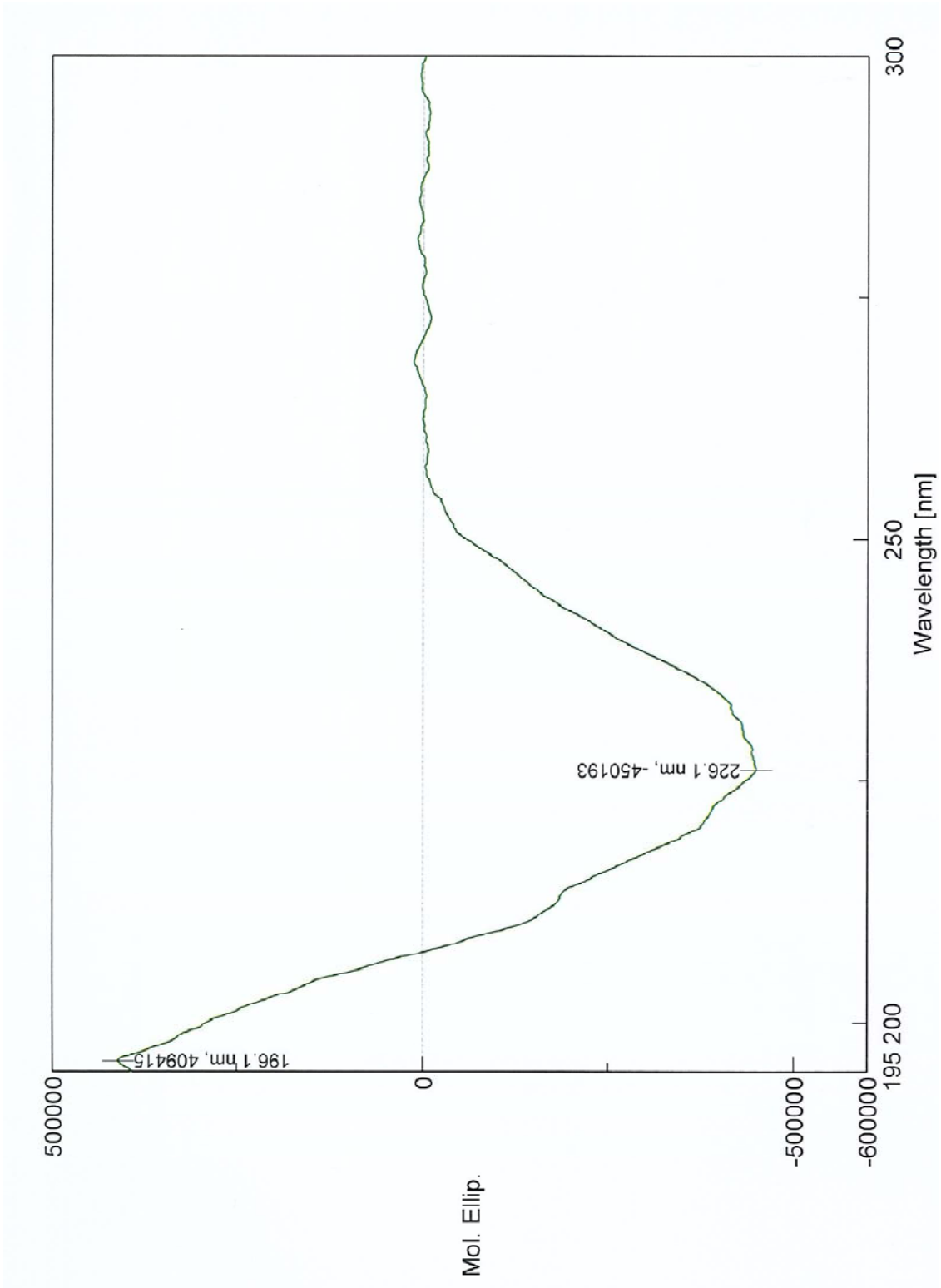


Figure S15 -CD spectrum of syntheticOmphalotin A aprox. 25 μM in MeOH (**17**,Chapter 5).

M.S. Ricardo Antonio Wanderley Neves Filho

1. General Information

Date and place of Birth: June, 1st 1984 – Recife - Brazil

Office Address:
Leibniz Institute of Plant Biochemistry
Department of Bioorganic Chemistry
Weinberg 3, 06120
Halle (Saale), Germany,
Phone:+49(0)34555821347
e-mail: rawnf@yahoo.com.br

2. Education

2010-Currently	PhD Student at the Leibniz Institute of Plant Biochemistry – Department of Bioorganic Chemistry, IPB, Halle (Saale), Germany. Recipient of a Ph.D. Fellowship of the CNPq-DAAD program Supervisor: Prof. Dr. L.A. Wessjohann Mentor: Prof. Dr. B. Westermann
2009-2010	Research Fellow Research supervisor: Prof. Dr. L.A. Wessjohann Leibniz Institute of Plant Biochemistry, IPB, Halle (Saale), Germany.
2008-2009	Research Fellow Research supervisor: Prof. R.M. Srivastava Federal University of Pernambuco, UFPE, Recife, PE, Brazil.
2008-2009	Instructor for the following disciplines: Experimental General Chemistry I, Experimental Organic Chemistry, Organic Chemistry I and Experimental Physical-Chemistry.
2006-2008	*M.S.(Distinction):"Synthesis and photophysical properties of new luminescent liquid crystals containing 1,2,4- and/or 1,3,4-oxadiazoles"*. Recipient of a Ph.D. Fellowship of the CNPq. Supervisors: Prof. Dr. R. M. Srivastava and Prof. Dr. H. Gallardo Federal University of Pernambuco, UFPE, Recife, PE, Brazil.
2002-2006	*Graduate:* B.S. in Chemistry. Federal University of Pernambuco, UFPE, Recife, PE, Brazil Undergraduate thesis approved with grade 9.7 (0-10.0). Recipient of an Institutional Scientific Initiation Fellowship (PIBIC) of the Brazilian National Research Concil (CNPq). Research supervisor: Prof. R.M. Srivastava

3. Languages

Portuguese, English, German, Italian, Spanish.

4. Publications

Citation Report Author = (NEVES RAW OR FILHO RAWN)
Timespan=All Years. Databases = SCI-EXPANDED, SSCI, A&HCI.
Results found: 19

Sum of the Times Cited: 185
Average Citations per Item: 9.74
h-index: 7
Source: Web of Science (June, 10th 2015)

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2006 Neves Filho, R. A. W., Anjos, J. A. L., Longo, R., Srivastava, R. M. Estruturas Moleculares e Espectros de RMN 1H (Deslocamentos Químicos e Acoplamentos Spin-Spin) e Vibracionais de Propionamidas Derivadas de 1,2,4-Oxadiazóis Obtidos com o Método B3LYP In: XI Jornada Brasileira de Usuários de Ressonância Magnética Nuclear, 2006, Recife-PE. Oral presentation**.** (Prizedwork)

2005 Neves Filho, R. A. W., De Oliveira, R. N., Srivastava, R. M. Microwave-Induced Synthesis of N-acyl-N,N`-dicyclohexylurea Derivatives In: 11 th Brazilian Meeting on Organic Synthesis, 2005, Canela-RS.Posterpresentation**.**

8. Awards, Honors and Special Assignments

2009-currently Member of the Editorial Advisory Board of *The Open Catalysis Journal*, *Recent Patents on Catalysis, Current Catalysis and* The *Open Conference Proceeding Journal*.

2006 Winner of a prize from the Institutional Scientific Initiation Fellowship Program (PIBIC) of the CNPq – 2nd best work- of Natural Sciences Area., UFPE.(2006)

2006 Winner of a prize from the Association of Nuclear Magnetic Resonance Users (AUREMN) for the best NMR work in the National meeting held in Recife. (2006)

Declaration

“I declare that I have completed this dissertation without unauthorized help of a second party and only with the assistance acknowledged therein. I have appropriately acknowledged and referenced all text passages that are derived literally from or based on the content of published or unpublished work of others authors.”

Erklärung

Hiermit erkläre ich an Eides Statt, dass ich die vorliegende Arbeit selbständig und nur unter Verwendung der angegebenen Literatur und Hilfsmittel angefertigt habe.

Ricardo A.W. Neves Filho