Journal of Medicinal Chemistry

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J. Med. Chem., **Just Accepted Manuscript** • DOI: 10.1021/acs.jmedchem.9b01457 • Publication Date (Web): 06 Jan 2020

Synthesis and Structure-Activity Relationship Studies of C2-Modified Analogs of the Antimycobacterial Natural Product Pyridomycin

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ABSTRACT

A series of derivatives of the antimycobacterial natural product pyridomycin have been prepared with the C2-side chain attached to the macrocyclic core structure by a C-C single bond, in place of the synthetically more demanding enol ester double bond found in the natural product. Hydrophobic C2-substituents of sufficient size generally provide for potent *anti-Mtb* activity of these dihydropyridomycins (MIC values around 2.5 μM), with several analogs thus approaching the activity of natural pyridomycin. Surprisingly, some of these compounds, in contrast to pyridomycin, are insensitive to overexpression of InhA in *Mtb*. This indicates that their *anti-Mtb* activity does not critically depend on inhibition of InhA and that their overall mode of action may differ from that of the original natural product lead.

INTRODUCTION

Tuberculosis (TB), while often regarded as a disease of the past, as of today is still one of the major global threats to public health. In 2017, the World Health Organization (WHO) reported an estimated 10 million new cases of infection and 1.6 million TB-related deaths.[1] In addition, 1.7 billion individuals globally present an asymptomatic latent tuberculosis infection and have a 5-10% lifetime risk of developing active disease.[1] A particular risk for the activation of dormant bacteria exists for individuals with an impaired (or suppressed) immune system, e. g., as the result of a human immunodeficiency virus (HIV) infection. However, bacterial activation may also be triggered by malnutrition, affliction with diabetes or unhealthy lifestyle habits (smoking, alcohol abuse).[1] Recently, the emergence of multidrug- and extensively drug-resistant (MDR and XDR, respectively) strains of the causative pathogen *Mycobacterium tuberculosis* (*Mtb*) has become a serious threat also in industrialized countries.[1,2] This development has been exacerbated by the lack of development of novel *anti*-TB drugs for several decades; only two new chemical entities have been approved for TB treatment since the approval of rifampicin in 1968 (Italy; FDA approval in 1971),[3] namely bedaquiline[4,5] and delamanid.[6,7] Of these, only bedaquiline acts *via* a truly novel mechanism of action.[4,5] Therefore, in order to eradicate TB and eliminate resistant strains, the development of improved antimycobacterial agents, ideally equipped with novel mechanisms of action, is urgently required.

Pyridomycin (**1**, Figure 1) is a bacterial secondary metabolite that was first isolated in 1953 from the *Streptomyces* strain 6706 (later termed *S. pyridomyceticus*) by Maeda and co-workers.[8] At the time of discovery, the compound was shown to exhibit significant antimycobacterial activity and low systemic toxicity in mice. However, little additional work on the pharmacology of pyridomycin (**1**) has been reported for several decades thereafter.

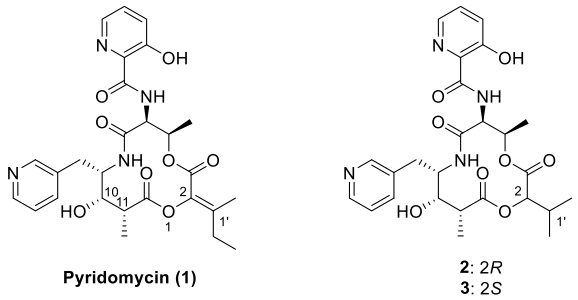


Figure 1. Structure of pyridomycin (**1**) and of 2,1'-dihydropyridomycins **2** and **3**.

As part of a comprehensive initiative directed towards the discovery of new *anti*-TB drugs (ERC FP7 program MM4TB), we recently revisited **1** as a potential lead structure for *anti*-TB drug discovery and we reconfirmed its antimycobacterial activity *in vitro* (MIC = 0.56 – 1.48 μM).[9] Of equal importance, we determined the molecular target of pyridomycin (**1**) as the mycobacterial NADH-dependent enoyl-[acyl-carrier-protein] reductase (InhA),[9] which is also the ultimate target of the clinical TB drug isoniazid (INH). INH acts as a prodrug and requires activation by katG, a mycobacterial catalase-peroxidase, to exert its antimycobacterial activity.[10] Resistance to INH most often results from mutations in the katG gene rather than in InhA itself.[11] In contrast, we have shown pyridomycin (**1**) to be a direct NADH-competitive inhibitor of the reductase; as a consequence, most INH-resistant strains remain susceptible to **1**,[9] which renders the compound a promising starting point for TB drug discovery.

The binding mode of **1** to InhA could be determined by X-ray crystallography, which showed that **1**, in contrast to the INH-NADH adduct formed after activation of INH by KatG[10] and other previously reported direct InhA inhibitors,[12–20] blocks *both* the NADH as well as the lipid binding site of the enzyme.[21] This binding mode makes pyridomycin unique among InhA inhibitors and effectively defines a new mode of action.

Only one total synthesis of pyridomycin (**1**) has been reported in the literature,[22,23] with the most challenging and difficult step being the installment of the exocyclic C2-C1'-double bond of the

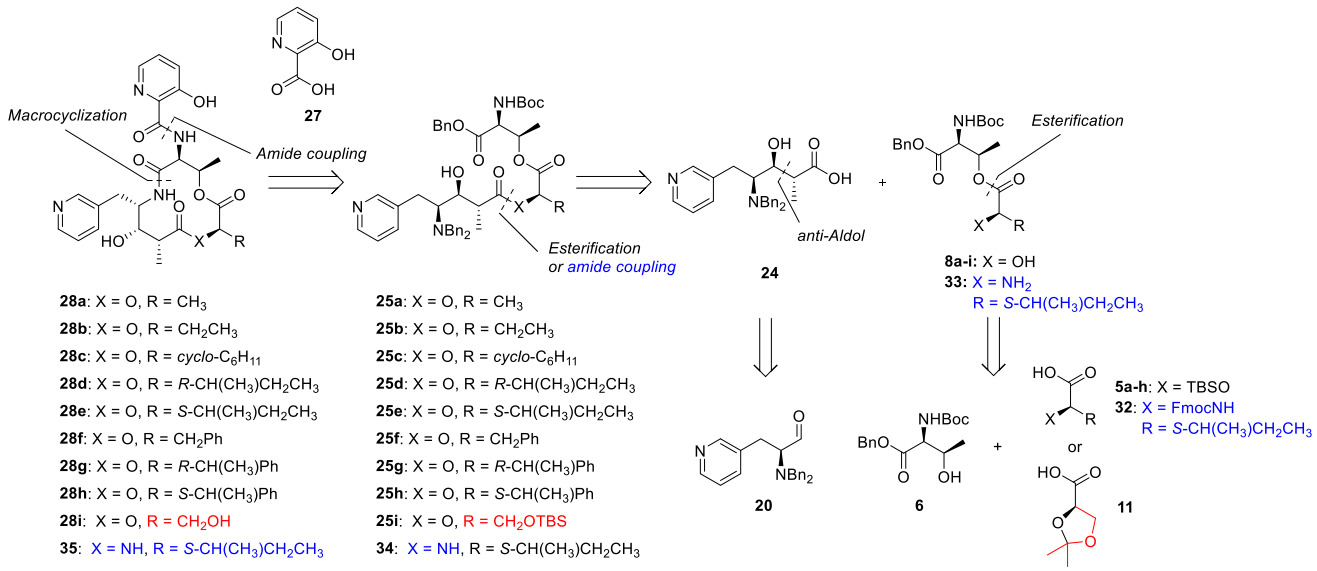
unique enol ester moiety. In order to circumvent this problem, we have investigated analogs of **1** with a single bond between C2 and C1' which we have termed "dihydropyridomycins".[24] Prototypical examples of this family of pyridomycin analogs are the 2-*iso*-propyl derivatives **2** and **3** (Figure 1), which retain a simple branched C2-side chain. Of these two analogs, the 2*R* isomer **2** was only *ca*. 4-fold less active than **1**, while it was 8-fold more potent than its 2*S* isomer **3** (Figure 1). These findings indicated that (a) the enol ester moiety in pyridomycin (**1**) is not crucial for antimycobacterial activity, while at the same time (b) highlighting the importance of the configuration of the newly created stereocenter (in comparison with **1**) at C2 for biological activity. Based on X-ray crystallography, the binding modes of dihydropyridomycin **2** and pyridomycin (**1**) to InhA are very similar, with the C2-substituent protruding into a hydrophobic pocket in both cases.[21] In light of these findings, we have now prepared a series of new 2*R* dihydropyridomycin derivatives with variations in the size and hydrophobicity of the C2-substituent and we have determined the effects of these variations on antimycobacterial activity. In addition, we have investigated the importance of the configuration at the C10 and C11 stereocenters of **1** for biological activity and we have assessed the effect of the replacement of the C12(O)-O1 ester group of the natural product by an amide moiety (which we thought could lead to improved metabolic stability).

RESULTS AND DISCUSSION

Chemistry. As outlined in Scheme 1, the synthesis of all pyridomycin analogs was to follow the same overall strategy that we had developed previously for the synthesis of **2** and **3**.[24] Thus, analogs **28** and **35** were to be obtained by late-stage amide coupling of 3-hydroxypicolinic acid (**27**) with the corresponding macrocyclic amines, which would in turn be formed by simultaneous cleavage

of the benzyl amino and benzyl ester groups by catalytic hydrogenation of amino acids **25** and **34**, respectively, followed by macrolactamization and cleavage of the BOC-protecting group.

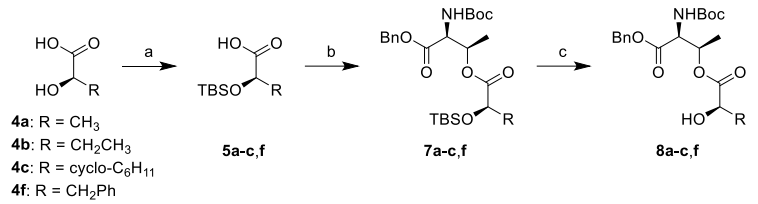
Scheme 1. Retrosynthesis of pyridomycin analogs **28** and **35**



Protected amino acids **25** and **34** would result from an esterification or an amide coupling reaction of acid **24** with alcohols **8** and amine **33**, respectively. Acid **24** can be accessed from aldehyde **20** by means of an *anti*-aldol reaction, as described previously,[24] while alcohols **8** and amine **33** were to be prepared by esterification of acids **5**, **11**, or **32** with Nα-BOC-Thr-OBn (**6**).

Synthesis of alcohols 8. The synthesis of alcohols **8a-c,f** departed from the commercially available hydroxy acids **4a-c,f**, which were converted into their TBS-ethers **5a-c,f** (*via* the bis-TBS derivative of the hydroxy acid and selective *in situ* cleavage of the TBS-ester group)[24] (Scheme 2). Yamaguchi esterification[25] of partially protected hydroxy acids **5a-c,f** with Nα-BOC-L-Thr-OBn (**6**) (which is commercially available but was synthesized here in two steps from L-Thr following literature procedures[24]) then delivered intermediates **7a-c,f**. Finally, TBS-deprotection with HF•pyridine furnished the desired free alcohols **8a-c,f**.

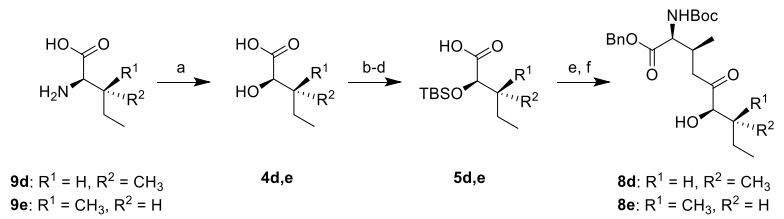
Scheme 2. Synthesis of alcohols **8a-c,f**[a]



[a]Reagents and conditions: (a) i. TBSCl, imidazole, DMF, rt, 24 h; ii. K_2CO_3, MeOH/H_2O (5:2), 0 °C to rt, 4 h, 79% to quant.; (b) 2,4,6-trichlorobenzoyl chloride, Et_3N, toluene, rt, 5 min, then alcohol **6**, DMAP, rt, 18 h, 12-92%; (c) HF·pyridine, THF, 0 °C to rt, 17 h, 82% to quant.

Alcohols **8d** and **8e**, which serve as precursors for dihydropyridomycins with an asymmetric C2-side chain that are the specific formal hydrogenation products of pyridomycin (**1**), were obtained in 6 steps from D-Ile (**9e**) and D-*allo*-Ile (**9d**), respectively (Scheme 3). Diazotation of the amino acids with aq. H_2SO_4 and $NaNO_2$ produced α-hydroxy acids **4d,e** in excellent yields with retention of configuration.[26,27]

Scheme 3. Synthesis of alcohols **8d,e**[a]



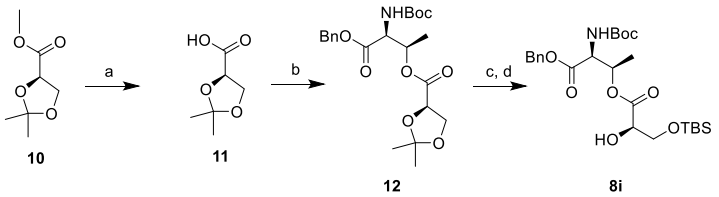
[a]Reagents and conditions: (a) H_2SO_4 1M, $NaNO_2$, H_2O, 0 °C, 19 h, then rt, 15 min, 97% and quant.; (b) benzyl bromide, Cs_2CO_3, DMF, 0 °C to rt, 20 h, 94% and 90%; (c) TBSOTf, lutidine, DCM, –78 °C, 30 min, then rt, 15 h, 93% and 77%; (d) H_2 (5 bar), Pd/C (10%), EtOAc, rt, 1 h, quant. and 81%; (e) 2,4,6-trichlorobenzoyl chloride, Et_3N, toluene, rt, 5 min, then alcohol **6**, DMAP, rt, 18 h, 92% and 68%; (f) HF·pyridine, THF, 0 °C to rt, 17 h, quant. The first yield given is for the respective intermediate leading to **8d**, the second for the one leading to **8e**.

Applying the conditions previously used for the conversion of **4a-c,f** into **5a-c,f** (excess TBSCl/imidazole in DMF, then K_2CO_3) failed to deliver the desired TBS-ethers **5d,e**. Hydroxy acids **4d,e** were thus transformed into the corresponding benzyl esters with BnBr/Cs_2CO_3 followed

by reaction with TBSCl; benzyl ester cleavage by catalytic hydrogenation then furnished acids **5d,e**. Finally, Yamaguchi esterification[25] of **5d,e** with **6** followed by TBS-deprotection gave free alcohols **8d,e**.

Compared to all other alcohols **8**, the synthesis of alcohol **8i** required a different protecting group regime, due to the need for a protecting group on the prospective C2-hydroxymethyl side chain. Starting from methyl ester **10**,[28] quantitative ester saponification gave carboxylic acid **11**, which was esterified with N_{α}-BOC-Thr-OBn (**6**) under Yamaguchi conditions[25] to give **12** in 71% yield (Scheme 4). Acetal cleavage with *p*-TsOH in MeOH followed by selective protection of the primary hydroxy group as a TBS-ether then furnished alcohol **8i** in 41% overall yield from **12**.

Scheme 4. Synthesis of alcohol **8i**[a]

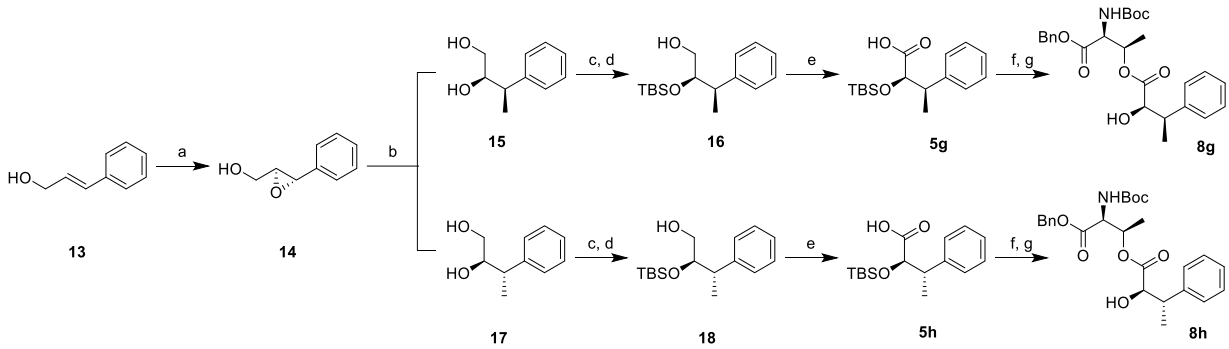


[a]Reagents and conditions: (a) KOH (aq. 2M), MeOH, rt, 1 h, 99%; (b) 2,4,6-trichlorobenzoyl chloride, Et_3N, THF/DMF (2:3), rt, 5 min, then alcohol **6**, DMAP, rt, 18 h, 71%; (c) *p*-TsOH, MeOH, rt, 20 h, 69%; (d) TBSCl, imidazole, DCM, rt, 18 h, 60%.

The syntheses of the two diastereoisomeric alcohols **8g** and **8h** were more elaborate than those of all other building blocks **8**, as they required the *de novo* installation of two chiral centers in precursor acids **5g** and **5h**. Both syntheses proceeded through epoxide **14** as a common intermediate that was obtained by enantioselective Sharpless epoxidation of *trans*-cinnamyl alcohol (**13**) in 74% yield.[29,30] Epoxide **14** was opened either in an *anti*-fashion with MeLi, to furnish diol **15** in 74% yield as a single diastereoisomer,[31] or in a *syn*-fashion with Me_3Al,[32] to provide **17** in 77% yield as an inseparable mixture of diastereoisomers (*d.r.* = 3.4:1). The diols **15**

and **17** were doubly TBS-protected and the primary silyl-ether was selectively cleaved using camphorsulfonic acid (CSA). The resulting alcohols **16** and **18** were oxidized to the corresponding acids **5g** and **5h** in good yields (94% and 86%, respectively) following a two-stage Swern-Pinnick protocol.[33–35]

Scheme 5. Synthesis of alcohols **8g,h**[a]



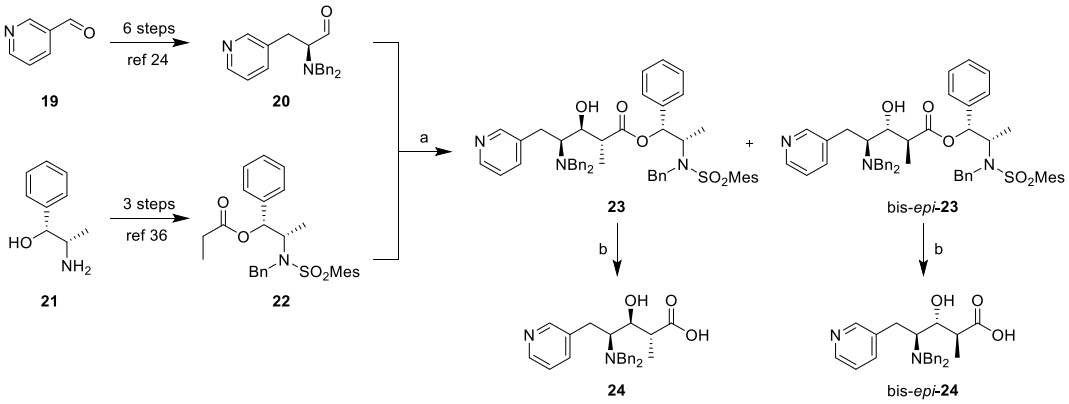
[a]Reagents and conditions: (a) L-(+)-DET, TTIP, *t*-BuOOH, DCM, –20 °C, 16 h, 74%; (b) for **15**, CuCN, MeLi, Et_2O, –78 °C to 0 °C, 3 h, 74%; for **17**, Me_3Al, toluene, 0 °C, 2 h, 77% (3.4:1 mixture of diastereoisomers); (c) TBSCl, imidazole, DMF, 0 °C to rt, 12-15 h, 93% and 88% (7.73:1 mixture of diastereoisomers); (d) CSA, DCM/MeOH 1:1, 0 °C, 3-5 h, 60% and 48%; (e) i. $(COCl)_2$, DMSO, Et_3N, DCM, –78 °C to rt; ii. $NaClO_2$, $NaH_2PO_4 \cdot H_2O$, *t*-BuOH/THF/H_2O (4.5:3:1), rt, 2 h, 94% and 86%; (f) **6**, 2,4,6-trichlorobenzoyl chloride, Et_3N, DMAP, toluene, 0 °C to rt, 2 h, 75% and 60%; (g) HF·pyridine, THF, pyridine, 0 °C to rt, 16 h, 73% and 77%. The first yield is for the respective intermediate leading to **8g**, the second for the one leading to **8h**.

Yamaguchi esterification[25] of **5g,h** with **6** followed by TBS-removal with HF•pyridine finally furnished the desired alcohols **8g** and **8h** in 73% and 77% yield, respectively. Importantly, while the undesired *anti* isomer could not be completely removed from the various intermediates *en route* from **17** to **8h** (ca. 5-10% of the *anti* product present), **8h** was obtained as a single diastereoisomer.

Synthesis of acids 24 and bis-*epi*-24. Acid **24** was synthesized from aldehyde **20** and chiral ester **22** *via* a Masamune *anti*-aldol reaction[36] as previously described (Scheme 6).[24] The reaction gave a separable mixture of diastereomeric aldol products in a 2:1 ratio, from which the desired

product **23** and bis-*epi*-**23** were isolated as pure isomers in 45% and 18% yield, respectively. The configuration of **23** was previously established by X-crystallography of a derived lactam;[24] in this work, we have also obtained an X-ray crystal structure of the minor diastereoisomer, which confirmed its structure as bis-*epi*-**23** (see Supporting Information). Finally, base-mediated ester hydrolysis with LiOH in MeOH/THF/H_2O gave the free carboxylic acids **24** and bis-*epi*-**24**, respectively, in high yields.

Scheme 6. Synthesis of acids **24** and bis-*epi*-**24**[a]

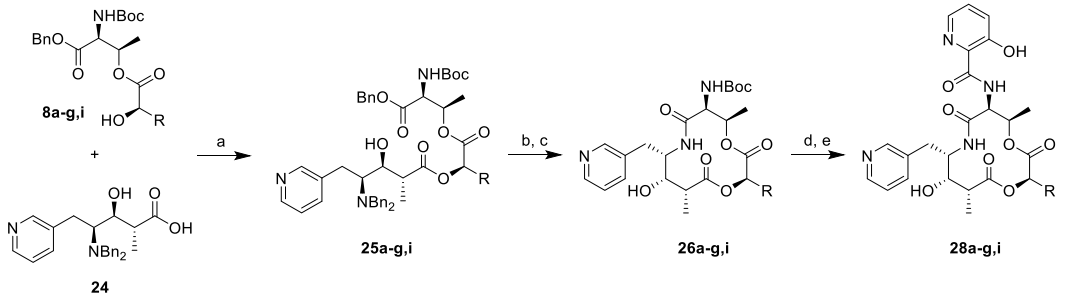


[a]Reagents and conditions: (a) *c*-Hex_2BOTf, Et_3N, DCM, –78 °C, 45% (**23**)/18% (bis-*epi*-**23**); (b) LiOH, MeOH/H_2O/THF (3:2:2), rt, 24 h, 83-96%.

Synthesis of analogs 28a-g,i: Analogs **28a-g,i** were all synthesized according to the general strategy outlined in Scheme 1. Thus, Yamaguchi esterification[25] of acid **24** with alcohols **8a-g,i** at low temperature (–78 °C to –35 °C)[24] provided **25a-g,i** in low to moderate yields (Scheme 7). Simultaneous removal of all benzyl-protecting groups by catalytic hydrogenation provided the corresponding free amino acids, which were submitted to macrolactamization with HATU[37] and DIPEA in DCM/DMF (100:1) at high dilution to deliver macrocycles **26a-g,i** in yields between 23% and 62%. The exocyclic amino group was then deprotected with TFA (in the case of **26i**, this resulted in concomitant cleavage of the TBS-ether in the C2-substituent) and the free amines were

coupled to 3-HPA (**27**) with HATU,[37] furnishing the targeted pyridomycin analogs **28a-g,i** in yields between 30% and 65% over two steps. Importantly, an X-ray crystal structure of **26c** could be obtained and unequivocally confirmed the absolute configuration of the compound (see Supporting Information).

Scheme 7. Synthesis of C2-modified pyridomycin analogs **28a-g,i**[a]



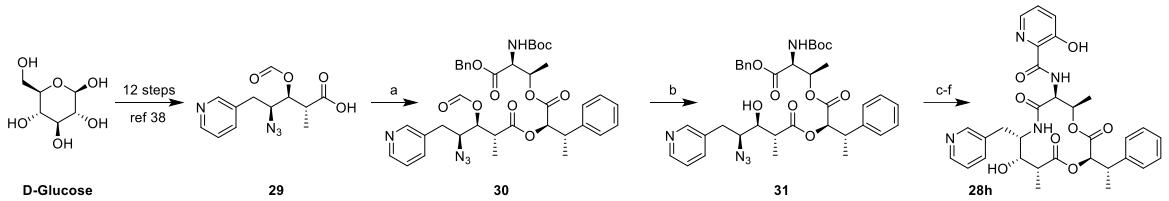
[a]Reagents and conditions: (a) 2,4,6-trichlorobenzoyl chloride, Et_3N, DMAP, toluene, –78 °C to –35 °C, 43 h, then 0 °C, 2 h, 18-72%; (b) H_2 (1 bar), Pd/C, MeOH, rt, 5 h; (c) HATU, DIPEA, DCM/DMF, rt, 18 h, 23-62% over 2 steps; (d) TFA, DCM, 0 °C, 10 min, then rt, 2 h; (e) 3-HPA (**27**), HATU, DIPEA, MeCN, rt, 18 h, 30-65% over 2 steps.

Synthesis of analog 28h. Unfortunately, the attempted Yamaguchi esterification[25] of acid **24** with alcohol **8h** failed to deliver the desired ester; and the same was true for other esterification methods investigated. As an alternative to acid **24**, we chose to investigate the esterification of alcohol **8h** with acid **29** (Scheme 8), which was obtained in 12 steps from D-glucose following a route that had been described previously by Kinoshita and co-workers[38] as part of their total synthesis of pyridomycin (**1**) (with some of the steps requiring optimization; see Supporting Information).

Gratifyingly, acid **29** could be successfully esterified with alcohol **8h** under Steglich conditions (DCC, DMAP)[39] to deliver ester **30** in high yield (78%). The formyl protecting group was then removed with $KHCO_3$ in MeOH, delivering ester **31** in 89% yield. Subsequent catalytic

hydrogenation led to concomitant benzyl-removal and azide reduction, furnishing the corresponding amino acid. HATU-mediated macrolactamization followed by BOC-removal with TFA and HATU-mediated coupling of the ensuing macrocyclic amine with 3-HPA (**27**) finally furnished analog **28h**.

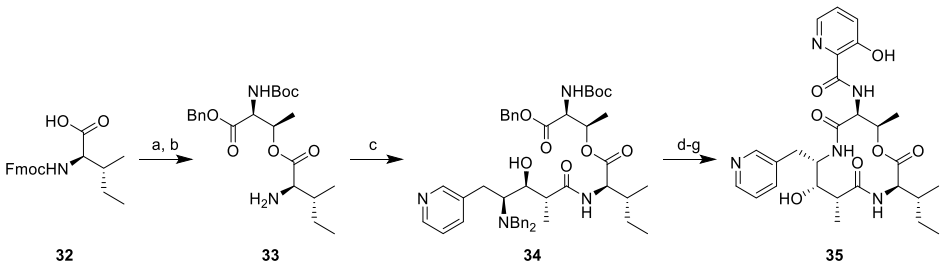
Scheme 8. Synthesis of C2-modified pyridomycin analog **28h**[a]



[a]Reagents and conditions: (a) **8h**, DCC, DMAP, DCM, 0 °C to rt, 5 h, 78%; (b) $KHCO_3$, MeOH, rt, 1 h, 89%; (c) Pd/C, MeOH, rt, 1.5 h; (d) HATU, DIPEA, DCM, DMF, rt, 16 h, 31% over 2 steps; (e) TFA, DCM, 3 h; (f) 3-HPA (**27**), HATU, DIPEA, DMF, 16 h, 43% over 2 steps.

Synthesis of analog 35. Instead of protected hydroxy acids **5** or **11**, the synthesis of analog **35** involved the esterification of Nα-BOC-Thr-OBn (**6**) with Nα-Fmoc-D-Ile (**32**) (Scheme 9).

Scheme 9. Synthesis of pyridomycin analog **35**[a]

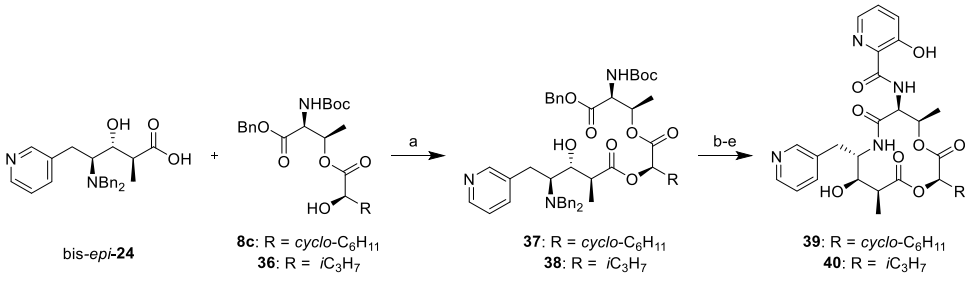


[a]Reagents and conditions: (a) 2,4,6-trichlorobenzoyl chloride, Et_3N, THF, rt, 5 min, then alcohol **6**, DMAP, toluene, rt, 1 h, 98%; (b) piperidine, MeCN, rt, 30 min, 64%; (c) **24**, HATU, DIPEA, MeCN/THF (10:1), 0 °C to rt, 48 h, 75%; (d) H_2 (1 atm), Pd/C (10%), MeOH, rt, 2.75 h, 96% (crude); (e) HATU, DIPEA, DCM, DMF (1%), rt, 18 h, 83%; (f) TFA, DCM, rt, 3 h, quant. (crude); 3-HPA (**27**), HATU, DIPEA, DMF, rt, 18 h, 25%.

The reaction was again conducted under Yamaguchi conditions[25] and furnished the desired ester in excellent yield (98%). Fmoc-cleavage with piperidine then gave the free amine **33.** HATU-mediated coupling of the latter with acid **24** delivered amide **34** in 75% yield. Global debenzylation with hydrogen over Pd/C and subsequent macrolactamization with HATU and DIPEA under the previously established conditions furnished the protected macrocycle in high yield. Cleavage of the BOC-group and coupling of the free amine with 3-HPA (**27**) (HATU/DIPEA) finally gave analog **35** in 20% yield for the four-step sequence from **34** (after RP-HPLC purification).

Synthesis of analogs 39 and 40. Given the availability of significant amounts of acid bis-*epi*-**24** from a number of runs of the aldol reaction between **20** and **22**, we also pursued the synthesis of dihydropyridomicin analogs **39** and **40** with inverted configurations at the C10 and C11 stereocenters (pyridomycin numbering; Scheme 10), in order to gain information on the effect of these stereochemical changes on antimycobacterial activity. The synthesis of these analogs from bis-*epi*-**24** and alcohols **8c** and **36**, respectively, followed the same strategy as for analogs **28a-i** (Scheme 10).

Scheme 10. Synthesis of analogs **39** and **40** with inverted configurations at C10 and C11[a]



[a]Reagents and conditions: (a) 2,4,6-trichlorobenzoyl chloride, Et_3N, DMAP, toluene, –78 °C to –35 °C, 43 h, then 0 °C, 2 h, 51-52%; (b) H_2 (1 bar), Pd/C, MeOH, rt, 5 h; (c) HATU, DIPEA, DCM/DMF, rt, 18 h, 33-42% over 2 steps; (d) TFA, DCM, 0 °C, 10 min, then rt, 2 h; (e) 3-HPA (**27**), HATU, DIPEA, MeCN, rt, 18 h, 37-51% over 2 steps.

Antimycobacterial Activity and InhA Inhibition. Pyridomycin analogs **28a-i**, **35**, **39**, and **40** were assessed for their antibacterial activity against *Mtb* strain H37Rv. For those compounds that displayed significant antimycobacterial activity, their IC_{50} for inhibition of InhA (S94A) was also determined with either 2-*trans*-octenoyl-CoA or 2-*trans*-dodecenoyl-CoA as the substrate.

As illustrated by the MIC values summarized in Table 1, all dihydropyridomycins with 2-substituents bulkier than methyl display good antimycobacterial activity, which in several cases (**28b-h**) surpasses the activity of our initial dihydropyridomycin **2** and thus begins to approach the activity of pyridomycin (**1**) itself. In contrast, C2-methyl analog **28a** is significantly less active than ethyl derivative **28b**, while the presence of a polar side chain as in **28i** results in a >150-fold loss in potency. Although we cannot rule out the possibility that the observed loss of activity for **28i** results from a decreased ability of the compound to penetrate the mycobacterial cell wall, the data are in general agreement with the structural data for the complexes between InhA and **1** or **2**, respectively;[21] in these structures, the C2-substituent protrudes into a hydrophobic pocket. At the same time, it is still surprising that a relatively small substituent such as ethyl essentially produces the same activity as the bulkier and more hydrophobic substituents present in **28c-h** which would be expected to engage in more extensive hydrophobic interactions with the hydrophobic protein cavity. As for asymmetric C2-side chains as in **28d/e** and **28g/h**, the *R* diastereoisomer (**28d**) is found to be more potent for the **28d/e** pair, while no difference is observed between **28g** and **28h**. However, even the activity difference between **28d** and **28e** is small and may not be biologically significant. Likewise, no meaningful activity difference is observed between C2-benzyl derivative **28f** and the branched variants **28g/h**.

Table 1. Antimycobacterial and InhA-inhibitory activity of pyridomycin (**1**) and of dihydropyridomycins **2**, **3**, **28a-i**, **35**, **39** and **40**

Compound	MIC (μM)[a]	Relative MIC (MIC/MIC(**1**))[b]	IC_{50} InhA (S94A) (μM)[c]
Pyridomycin (**1**)	0.55 – 3.05	-	3.43 ± 0.60
2	3.2	4	32.8 ± 9.20
3	26.0	32	NM[d]
28a	49.9	15	ND[e]
28b	4.9	2	127 ± 2.31
28c	1.3	1.7	11.1 ± 0.703
28d	1.5	2	31.5 ± 8.61
28e	2.9	3.9	87.2 ± 23.4
28f	4.3	2	29.12 ± 1.93
28g	2.6	2.3	ND[e]
28h	2.6	2.3	ND[e]
28i	96.8	167	ND[e]
35	184.6	250	ND[e]
39	94.6	83	ND[e]
40	135.8	148	291 ± 2.42

[a]Minimal inhibitory concentrations (MIC) were measured against *Mtb* strain H37Rv using the resazurin method.[40] Data are average values from at least duplicate experiments. MIC's were determined in Middlebrook 7H9 broth (Difco) supplemented with 0.2% glycerol, 0.05% Tween 80 and 10% albumin-dextrose-catalase (ADC), which is routinely used in the screening for and testing of new antimycobacterial agents. The stability of pyridomycin (**1**) in this medium after 7 days was found to be >50% (for an initial concentration of 10 μg/mL). Some of the analogs **28** were also tested in the absence of Tween 80, which led to an increase in MIC of 2-4 fold (see Supporting Information). The exact reasons for these changes are unclear, but it appears conceivable that Tween 80 enhances the penetration of pyridomycin and pyridomycin analogs into *Mtb* by disrupting the outer capsule of the bacterial cell envelope.[41] The differences in MIC's between experiments in the presence or absence of Tween 80 do not affect the general SAR conclusions derived from the data shown in the Table 1. [b]The relative MIC is defined as the ratio of the MIC's of the respective analog and the pyridomycin (**1**) control in the same experiment. [c]Half maximal inhibitory concentrations (IC_{50}) were measured against InhA (S94A) with 2-*trans*-octenoyl-CoA or 2-*trans*-dodecenoyl-CoA as the substrate. For experimental details *cf*. ref.[9] Data are average values from experiments carried out in triplicates ± SD . [d]NM, no activity detectable up to a concentration of 75 μM. [e]ND, no values determined.

Unfortunately, the substitution of an ester for an amide moiety in analog **35** resulted in the complete loss of antimycobacterial activity. This may be attributed to an intolerable conformational change of the macrocycle or to unfavorable interactions of the protein with the more polar amide group. Finally, analogs **39** and **40** did not exhibit any antimycobacterial activity, which highlights the importance of the natural configurations of the stereocenters at C10 and C11 (although we cannot exclude that changing only one of these stereocenters, rather than both simultaneously, would have a less pronounced effect).

The data in Table 1 also show that there is no clear correlation between the antimycobacterial activity of pyridomycin analogs **28** and the inhibitory activity against their purported target enzyme InhA *in vitro* (where determined). There is no straightforward explanation for this discrepancy, but the data might indicate that at least some of our dihydropyridomycins could exhibit additional activity(ies) that are unrelated to InhA, perhaps by targeting other short chain enoyl reductase(s) in *Mtb* that is (are) different from but still similar to InhA.[42,43] To address this question, MIC values were determined for a group of compounds against the wild type H37Rv strain as well as the corresponding InhA-overexpressing H37Rv::pMVinhA strain[44] (Table 2). Pyridomycin (**1**) and isoniazid (INH) were used as positive controls, and both compound showed significantly decreased activity against the InhA-overexpressing strain (*ca*. 16- and 31-fold reduction in MIC, respectively). A similar decrease in activity was also observed for analogs **28b,f**, thus indicating that they, too, target InhA. In contrast, and quite intriguingly, **28c,e** retained essentially full antimycobacterial activity against H37Rv::pMVinhA. Thus, while **28c,e** show inhibition of InhA in a biochemical assay, their antibacterial activity appears not to depend critically on inhibition of bacterial InhA and they act through (an)other mechanism(s) of action that is (are) unaffected by the overexpression of InhA.

Table 2. Antimycobacterial activity of selected analogs and INH against wild type H37Rv and against InhA-overexpressing H37Rv::pMVinhA[44]

Compound	MIC H37Rv::pMV261 (μM)[a]	MIC H37Rv::pMVinhA (μM)[a]	Resistance impact (fold MIC increase)
Pyridomycin (**1**)	0.74	11.56	16
2	3.1	24.2	8
28b	4.9	>19.1[b]	>4
28c	0.7	0.7	1
28e	2.9	2.9	1
28f	4.3	>17.3[b]	>4
Isoniazid (INH)	1.2	37.3	31

[a]Minimal inhibitory concentrations (MIC) were measured against *Mtb* strain H37Rv using the resazurin method[9,40] under the conditions detailed in footnote[a] of Table 1. H37Rv::pMVinhA refers to the *Mtb* strain H37Rv containing the pMV261::inhA plasmid; H37Rv::pMV261 refers to H37Rv transfected with the empty plasmid vector pMV261.[44] [b]Highest concentration tested.

Experiments aimed at identifying possible additional targets of these compounds are currently ongoing, but so far have not yielded any conclusive results. Independent of this, the data in Table 2 clearly demonstrate that rather subtle changes in the structure of the C2-substituent in dihydropyridomycins can completely overcome drug resistance due to overexpression of InhA.

CONCLUSION

In summary, we have achieved the synthesis of 12 new 2,1'-dihydropyridomycins and we have determined their antimycobacterial activities and, for selected examples, their capacity for inhibition of InhA. By doing so, we have established the importance of the natural configuration of the C10 and C11 stereocenters and the significance of the C12(O)-O1 ester linkage for antibacterial activity. Most importantly, we have identified several new dihydropyridomycins with

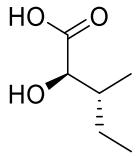
antimycobacterial activities starting to approach the activity of the lead natural product. At the same time, the (cellular) SAR for lipophilic C2-substituents larger than methyl is surprisingly flat. It remains to be seen if further modifications of the most potent analogs, for example the attachment of substituents to the phenyl part of analogs **28f** or **28g,h** could lead to enhanced activity. In addition, modifications in the hydroxypicolinic acid part of the pyridomycin/dihydropyridomycin structure will be required to enhance metabolic stability, in order to produce analogs suitable for eventual *in vivo* applications. These questions will be addressed in a forthcoming publication.

EXPERIMENTAL SECTION

General Information. All manipulations were conducted under an argon atmosphere using flame-dried glassware and standard syringe/septa and Schlenk techniques. Absolute solvents were purchased from Fluka (absolute over molecular sieves). Commercial chemicals were used without further purification. Solvents for extractions, flash column chromatography (FC) and thin layer chromatography (TLC) were purchased as commercial grade and distilled prior to use. TLC was performed on Merck TLC aluminum sheets (silica gel 60 F254). Spots were visualized with UV light (λ = 254 nm) or through staining with $Ce_2(SO_4)_3$/phosphomolybdic acid/H_2SO_4 (CPS) or $KMnO_4/K_2CO_3$. FC was performed using Fluka silica gel 60 for preparative column chromatography (particle size 40-63 μm). NMR spectra were recorded on a Bruker Avance 400 MHz or 500 MHz NMR spectrometer at 300 K. Chemical shifts (δ) are reported in ppm and are referenced to the solvent signal as an internal standard ($CDCl_3$ δ 7.26 ppm for ^1H and δ 77.00 ppm for ^{13}C spectra; CD_3OD δ 3.31 ppm for ^1H and δ 49.00 ppm for ^{13}C spectra; $(CD_3)_2SO$ δ 2.50 ppm for ^1H and δ 39.43 ppm for ^{13}C spectra). Data are reported as follows: s = singlet, d = doublet, t =

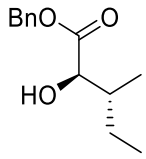
triplet, q = quartet, quint = quintet, sext = sextet, m = multiplet, br. = broad signal, *J* = coupling constant in Hz. The multiplicity of signals is reported based on appearance (i.e. doublet of doublets that are apparent triplets are described as triplets). All ^{13}C-NMR spectra were measured with complete proton decoupling. ^{1}H- and ^{13}C-signals were assigned using two-dimensional correlation experiments (COSY, HSQC, HMBC). IR spectra were recorded on a Jasco FT/IR-6200 instrument on thin films. Optical rotations were measured on a Jasco P-1020 polarimeter operating at the sodium D line (λ = 589 nm) and are reported as follows: $[\alpha]_D^T$, concentration (*c* in g/100 mL) and solvent. Melting points were obtained in open capillary tubes using a Büchi melting point apparatus B-540 and are uncorrected. Mass spectra were recorded by the MS service of the Laboratory of Organic Chemistry (LOC) of the ETH Zürich; HR-MS (ESI) spectra were measured on a Bruker Daltonics maxis (UHR-TOF) and HR-MS (EI) on a Waters Micromass AutoSpec Ultima instrument. The purity of final products submitted for biological testing was determined by analytical RP-HPLC to be ≥95%. (For HPLC traces, see the Supporting Information). As the sole exception, HPLC analysis was not performed on compound **28d**; the purity of this compound was estimated to be >90% based on the ^{1}H-NMR spectrum included in the Supporting Information.

(2R,3R)-2-Hydroxy-3-methylpentanoic acid (4d)



To a solution of D-Ile (**9d**) (500 mg, 3.81 mmol, 1.00 eq.) in 1M sulfuric acid (10 mL) was added a solution of $NaNO_2$ (2.10 g, 30.5 mmol, 8.00 eq) in water (10 mL) dropwise over 4 h a 0 °C. The reaction mixture was stirred at 0 °C for 15 h, allowed to warm to rt, and stirred for further 15 min. The reaction was quenched by adding NaCl until saturation of the solution. The aqueous layer was extracted with EtOAc (3 x 30 mL), and the combined organic layers were washed with water (2 x 10 mL) and with brine (2 x 10 mL), dried over $MgSO_4$, filtered, and concentrated *in vacuo* to deliver **4d** (489 mg, 97%) as a yellow oil. The crude product was used in the following step without further purification. **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.17. **$[\alpha]_D^{20}$** = +12.6° (*c* 0.70, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 4.18 (d, *J* = 3.6 Hz, 1H), 1.94 – 1.85 (m, 1H), 1.49 – 1.39 (m, 1H), 1.36 – 1.24 (m, 1H), 1.03 (d, *J* = 6.9 Hz, 3H), 0.93 (t, *J* = 7.4 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 179.0, 74.8, 39.1, 23.8, 15.5, 11.9. **IR** (thin film): υ (cm^{-1}) = 3449, 2965, 2937, 2879, 1718, 1646, 1461, 1382, 1273, 1242, 1216, 1169.13, 1135, 1071, 1045, 1017. **HR-MS** (ESI): Calcd. for $C_6H_{13}O_3$ $[M+H]^+$ 133.0859; found 133.0859 *m/z*.

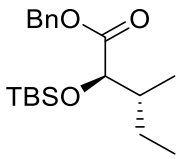
Benzyl (2R,3R)-2-hydroxy-3-methylpentanoate (4dA)



To a solution of **4d** (453 mg, 3.43 mmol, 1.00 eq.) in DMF (20 mL) was added benzyl bromide (448 µL, 3.77 mmol, 1.10 eq.) and Cs_2CO_3 (581 mg, 1.78 mmol, 0.52 eq.) at 0 °C. The white

suspension was vigorously stirred at rt for 20 h. H_2O (200 mL) was added and the mixture was extracted with Et_2O (3 x 200 mL). The combined organic layers were washed with water (2 x 20 mL), dried over $MgSO_4$, filtered, and concentrated *in vacuo* to deliver **4dA** (716 mg, 94%) as a yellow oil. The crude product was used in the following step without further purification. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.48. **[α]$_D^{20}$** = +3.3° (*c* 0.96, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.41 – 7.28 (m, 5H), 5.22 (dd, *J* = 19.5, 12.1 Hz, 2H), 4.12 (dd, *J* = 5.9, 3.7 Hz, 1H), 2.68 (d, *J* = 6.1 Hz, 1H), 1.88 – 1.74 (m, 1H), 1.38 – 1.27 (m, 1H), 1.27 – 1.16 (m, 1H), 0.97 (d, *J* = 6.9 Hz, 3H), 0.86 (t, *J* = 7.4 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.0, 135.3, 128.8, 128.7, 128.6, 75.0, 67.4, 39.3, 23.8, 15.6, 11.9. **IR** (thin film): υ (cm^{-1}) = 3501, 2964, 2935, 2877, 1729, 1498, 1455, 1379, 1266, 1214, 1135, 1072, 1045, 963, 749, 697. **HR-MS** (ESI): Calcd. for $C_{13}H_{18}NaO_3$ $[M+Na]^+$ 245.1148 *m/z*; found 245.1154 *m/z*.

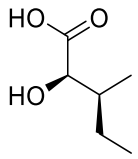
Benzyl (2R,3R)-2-((tert-butyldimethylsilyl)oxy)-3-methylpentanoate (4dB)



A solution of **4dA** (700 mg, 3.15 mmol, 1.00 eq.) and 2,6-lutidine (880 µL, 7.56 mmol, 2.40 eq.) in DCM (32 mL) was cooled to −78 °C. TBSOTf (868 µL, 3.78 mmol, 1.20 eq.) was added dropwise over 15 min. The mixture was stirred for 30 min at −78 °C, was slowly allowed to warm to 0 °C and stirred at rt for 15 h. The solution was quenched by the addition of sat. aq. $NaHCO_3$, diluted with Et_2O (15 mL each), and extracted with Et_2O (3 x 10 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 9:1 to 6:1 to 4:1) provided **4dB** (987 mg, 93%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.79. **[α]$_D^{20}$** = +33.6° (*c* 0.71, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.28 (m, 5H),

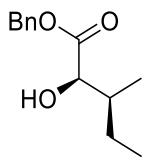
5.15 (dd, *J* = 19.8, 12.2 Hz, 2H), 4.05 (d, *J* = 5.0 Hz, 1H), 1.82 (m, 1H), 1.50 – 1.40 (m, 1H), 1.24 – 1.12 (m, 1H), 0.89 (d, *J* = 8.3 Hz, 3H overlapping s, 9H), 0.85 (t, *J* = 7.9 Hz, 3H), 0.01 (s, 3H), 0.00 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 173.5, 135.9, 128.6, 128.4, 76.7, 66.4, 39.6, 25.9, 24.1, 18.4, 15.60, 11.6, -4.8, -5.2. **IR** (thin film): υ (cm^{-1}) = 3026, 2959, 2930, 2880, 2857, 1752, 1731, 1461, 1380, 1252, 1172, 1139, 1069, 1005, 968, 860, 836. **HR-MS** (ESI): Calcd. for $C_{19}H_{32}NaO_3Si$ $[M+Na]^+$ 359.2013 *m/z*; found 359.2020 *m/z*.

(2R,3S)-2-Hydroxy-3-methylpentanoic acid (4e)



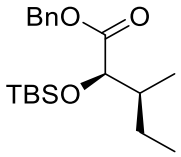
4e was prepared from D-*allo*-Ile (**9e**) according to the procedure described for **4d**. **4e** (568 mg, quant.) was obtained as a yellow oil. The crude product was used in the following step without further purification. **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.08. **$[\alpha]_D^{20}$** = +7.4 ° (*c* 0.35, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 4.29 (d, *J* = 2.0 Hz, 1H), 1.93 – 1.86 (m, 1H), 1.62 – 1.50 (m, 1H), 1.41 – 1.31 (m, 1H), 0.97 (t, *J* = 7.4 Hz, 3H), 0.88 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 180.0, 73.0, 38.5, 26.2, 13.3, 11.9. **IR** (thin film): υ (cm^{-1}) = 3521, 2965, 2924, 2863, 1726, 1464, 1373, 1251, 1201, 1145. **HR-MS** (ESI): Calcd. for $C_6H_{11}Na_2O_3$ $[M-H+2Na]^+$ 177.0498; found 177.0496 *m/z*.

Benzyl (2R,3S)-2-hydroxy-3-methylpentanoate (4eA)



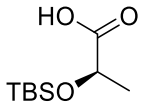
4eA was prepared from **4e** according to the procedure described for **4dA**. **4eA** (829 mg, 90%) was obtained as a yellow oil. The crude product was used in the following step without further purification. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.50. **[α]$_D^{20}$** = +4.3° (*c* 0.81, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.34 (m, 5H), 5.22 (dd, *J* = 17.4, 12.3 Hz, 2H), 4.23 (dd, *J* = 5.8, 2.9 Hz, 1H), 2.65 (d, *J* = 5.8 Hz, 1H), 1.83 (ddd, *J* = 14.2, 7.0, 2.8 Hz, 1H), 1.61 – 1.46 (m, 1H), 1.58 – 1.47 (m, 1H), 0.94 (t, *J* = 7.4 Hz, 3H), 0.78 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.5, 135.4, 128.8, 128.7, 128.6, 73.2, 67.5, 38.6, 26.1, 13.3, 12.0. **IR** (thin film): υ (cm^{-1}) = 3537, 3019, 2964, 2935, 2877, 1730, 1456, 1381, 1251, 1214, 1139, 1112, 1048, 753, 668. **HR-MS** (ESI): Calcd. for $C_{13}H_{18}NaO_3$ $[M+Na]^+$ 245.1148; found 245.1143 *m/z*.

Benzyl (2R,3S)-2-((tert-butyldimethylsilyl)oxy)-3-methylpentanoate (4eB)



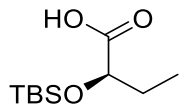
4eB was prepared from **4eA** according to the procedure described for **4dB**. FC (hexane/EtOAc 9:1 to 6:1 to 4:1) provided **4eB** (956 mg, 77%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.80. **[α]$_D^{20}$** = +42.9° (*c* 0.93, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.38 – 7.30 (m, 5H), 5.15 (dd, *J* = 14.3, 12.3 Hz, 2H), 4.15 (d, *J* = 3.7 Hz, 1H), 1.83 – 1.75 (m, 1H), 1.48 – 1.38 (m, 1H), 1.27 – 1.16 (m, 1H), 0.92 – 0.88 (t, *J* = 7.4 Hz, 3H overlapping with s, 9H), 0.84 (d, *J* = 6.8 Hz, 3H), 0.01 (s, 3H), 0.01 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 173.9, 136.1, 128.7, 128.7, 128.5, 75.3, 66.5, 39.8, 26.3, 26.0, 18.5, 13.9, 12.1, -4.6, -5.2. **IR** (thin film): υ (cm^{-1}) = 2959, 2931, 2881, 2857, 1752, 1461, 1381, 1361, 1252, 1214, 1142, 1072, 836, 752. **HR-MS** (ESI): Calcd. for $C_{19}H_{32}NaO_3Si$ $[M+Na]^+$ 359.2013 *m/z*; found 359.2015 *m/z*.

(R)-2-((tert-Butyldimethylsilyl)oxy)propanoic acid (5a)



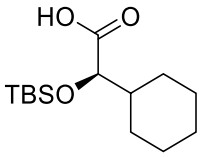
To a stirred solution of *D*-lactic acid (**4a**) (513 mg, 5.69 mmol, 1.00 eq.) in DMF (6 mL) at 0 °C were added imidazole (762 mg, 11.2 mmol, 1.97 eq.) and TBSCl (3.30 g, 21.9 mmol, 3.85 eq.). The white suspension was stirred at rt for 23 h and diluted with hexane/EtOAc (1:1, 90 mL). The organic layer was washed with aq. 10% citric acid (50 mL) and H_2O (90 mL), dried over $MgSO_4$, filtered, and concentrated *in vacuo*. The crude intermediate (2.29 g) was dissolved in MeOH (40 mL), cooled to 0 °C and a solution of K_2CO_3 (1.52 g, 11.1 mmol, 1.95 eq.) in H_2O (12.5 mL) was added. The reaction mixture was stirred at rt for 4 h, was diluted with DCM (150 mL) and was acidified to pH = 4 using citric acid (aq. 10%). The aqueous layer was extracted with DCM (3 x 150 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo* to deliver **5a** (588 mg, crude) as a slightly brown syrup. **5a** (crude) was used in the following step without further purification. **TLC** (SiO_2, hexane/EtOAc 1:3 +0.5% CH_3COOH, $KMnO_4$): R_f = 0.18. **^{1}H-NMR** (400 MHz, $CDCl_3$) δ (ppm) = 7.61 (br. s, 1H), 4.35 (q, *J* = 6.7 Hz, 1H), 1.43 (d, *J* = 6.8 Hz, 3H), 0.91 (s, 9H), 0.12 (s, 6H).

(R)-2-((tert-Butyldimethylsilyl)oxy)butanoic acid (5b)



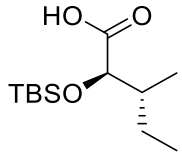
5b was prepared from (*R*)-hydroxybutyric acid (**4b**) according to the procedure described for **5a**. **5b** (crude) was used in the following step without further purification. **TLC** (SiO_2, hexane/EtOAc, 4:1 +1% CH_3COOH, CPS): R_f = 0.32. **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 4.27 (t, *J* = 5.0 Hz, 1H), 1.93 – 1.66 (m, 2H), 0.99 – 0.94 (m, 3H), 0.93 (s, 9H), 0.13 (s, 6H).

(R)-2-((tert-Butyldimethylsilyl)oxy)-2-cyclohexylacetic acid (5c)



5c was prepared from (*R*)-hexahydromandelic acid (**4c**) according to the procedure described for **5a**. FC (hexane/EtOAc 6:1 to 4:1) provided **5c** (547 mg, 79% over 2 steps) as a colorless oil. **TLC** (SiO_2, DCM/MeOH 95:5, UV): R_f = 0.50. **[α]$_D^{20}$** = +15.3° (*c* 1.41, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 4.05 (d, *J* = 4.0 Hz, 1H), 1.85 – 1.54 (m, 6H), 1.19 (d, *J* = 86.5 Hz, 5H), 0.93 (s, 9H), 0.11 (s, 3H), 0.09 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.5, 76.6, 42.5, 29.2, 27.0, 26.1, 26.0, 26.0, 25.7 (3 C), 18.2, -5.0, -5.2. **IR** (thin film): υ (cm^{-1}) = 2929, 2856, 1721, 1451, 1254, 1179, 1142, 900, 837, 777, 406. **HR-MS** (ESI): Calcd. for $C_{14}H_{28}NaO_3Si$ $[M+Na]^+$ 295.1700 *m/z*; found 295.1702 *m/z*.

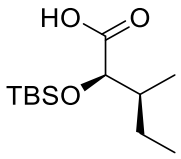
(2R,3R)-2-((tert-Butyldimethylsilyl)oxy)-3-methylpentanoic acid (5d)



A solution of **4dB** (280 mg, 832 µmol, 1.00 eq.) and Pd/C (10%, 89.0 mg, 83.2 µmol, 0.10 eq.) in EtOAc (16 mL) was stirred under a H_2-atmosphere (5 bar). After 1 h, the suspension was filtered through a plug of celite. The filtrate was evaporated to deliver **5d** (207 mg, quant.) as a colorless oil. The crude product was used in the following step without further purification. **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.50. **[α]$_D^{20}$** = +5.4° (*c* 0.94, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 4.17 (d, *J* = 3.8 Hz, 1H), 1.83 – 1.77 (m, 1H), 1.55 – 1.46 (m, 1H), 1.35 – 1.24 (m, 1H), 0.98 – 0.88 (d, *J* = 8.2 Hz, 3H, overlapping with t, *J* = 7.5 Hz, 3H), 0.94 (s, 9H), 0.12 (d, *J* = 5.1

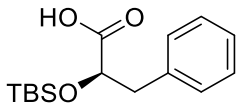
Hz, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 171.4, 76.3, 40.0, 25.8, 24.3, 15.2, 11.9, −4.9, −5.1. **IR** (neat, cm^{-1}): 2961, 2932, 2859, 1722, 1461, 1254, 1146, 837, 776. **HR-MS** (ESI): Calcd. for $C_{12}H_{25}O_3Si$ $[M-H]^-$ 245.1578 *m/z*; found 245.1575 *m/z*.

(2R,3S)-2-((tert-Butyldimethylsilyl)oxy)-3-methylpentanoic acid (5e)



5e was prepared from (**4eB**) according to the procedure described for **5d**. **5e** (26.7 mg, 81%) was obtained as a colorless oil. The crude product was used in the following step without further purification. **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.61. **[α]$_D^{20}$** = +18.3° (*c* 1.10, EtOH). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 4.20 (d, *J* = 3.0 Hz, 1H), 1.88 – 1.74 (m, 1H), 1.57 – 1.42 (m, 1H), 1.29 – 1.15 (m, 1H), 0.96 – 0.91 (t, *J* = 7.0 Hz, 3H, overlapping with d, *J* = 6.8 Hz, 3H),0.95 (s, 9H), 0.12 (d, *J* = 4.3 Hz, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 173.3 (extracted from HMBC), 75.8, 39.9, 25.8, 25.8, 13.7, 12.1, -4.7, -5.1. **HR-MS** (ESI): Calcd. for $C_{12}H_{27}O_3Si$ $[M+H]^+$ 247.1724 *m/z*; found 247.1730 *m/z*.

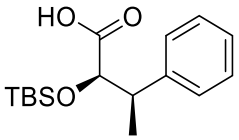
(R)-2-((tert-Butyldimethylsilyl)oxy)-3-phenylpropanoic acid (5f)



5f was prepared from D-(+)-3-phenyllactic acid (**4f**) according to the procedure described for **5a**. **5f** (2.24 g, crude) was used in the following step without further purification. **TLC** (SiO_2, hexane/EtOAc, 4:1, UV) : R_f = 0.31. **^{1}H-NMR** (400 MHz, $CDCl_3$) δ (ppm) = 7.33 – 7.19 (m, 5H),

4.42 (dd, *J* = 7.8, 3.8 Hz, 1H), 3.12 (dd, *J* = 13.7, 3.7 Hz, 1H), 2.95 (dd, *J* = 13.7, 7.8 Hz, 1H), 0.85 (s, 9H), -0.06 (s, 3H), -0.17 (s, 3H).

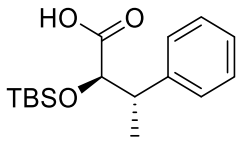
(2R,3R)-2-((tert-Butyldimethylsilyl)oxy)-3-phenylbutanoic acid (5g)



To a stirred solution of oxalyl chloride (115 mg, 0.91 mmol, 3.00 eq.) in DCM (1.5 mL) was slowly added DMSO (0.13 mL, 1.82 mmol, 6.00 eq.) at –78 °C. The mixture was stirred for 10 min and a solution of **16** (85.0 mg, 0.30 mmol, 1.00 eq.) in DCM (1.5 mL) was added at –78 °C. The reaction was stirred at –78 °C for 20 min and Et_3N (0.34 mL, 2.42 mmol, 8.00 eq.) was added at –78 °C. The mixture was allowed to warm to rt (over 20 min). The reaction was quenched by the addition of brine (2 mL) and was extracted with DCM (3 x 10 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 5:1) provided the aldehyde (yield considered as quantitative) as a slight yellow oil. To a solution of the aldehyde (303 µmol, 1.00 eq.) in *t*-BuOH/THF 4.5:3 (3 mL) and 2-methyl-2-butene (1.61 mL, 15.2 mmol, 50.0 eq.) was sequentially added at rt a solution of $NaH_2PO_4 \cdot 2H_2O$ (189 mg, 1.21 mmol, 4.00 eq.) in H_2O (0.18 mL) and a solution of $NaClO_2$ (87.7 mg, 0.97 mmol, 3.20 eq.) in H_2O (0.18 mL). The mixture was stirred for 2 h at rt. The reaction was quenched with sat. aq. $NaHCO_3$ solution (3 mL) and extracted with EtOAc (3 x 10 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 5:1 to 1:1) provided **5g** (84.1 mg, 94%) as a white solid. **TLC** (SiO_2; hexane/EtOAc 1:1, UV, CPS): R_f = 0.39. **$[\alpha]_D^{20}$** = +29.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.32 – 7.20 (m, 5H), 4.31 (dd, *J* = 4.9, 2.2 Hz, 1H), 3.26 – 3.14 (m, 1H), 1.38 (d, *J* = 7.3, 1.5 Hz, 3H), 0.90 (s, 9H), 0.03 (s, 6H). **^{13}C-NMR** (101

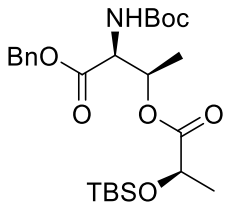
MHz, $CDCl_3$): δ (ppm) = 177.2, 128.7, 128.4, 127.3, 77.2, 44.00, 25.8, 17.7, -5.3. **IR** (thin film): υ (cm^{-1}) = 2957, 2930, 2886, 2858, 2359, 1721, 1472, 1463, 1455, 1256, 1146, 1115, 1061, 859, 838, 778, 763, 699, 666, 539, 525, 446, 441, 426, 418, 407. **HR-MS** (ESI): Calcd. for $C_{16}H_{26}NaO_3Si$ $[M+Na]^+$ 317.1543 *m/z*; found 317.1543 *m/z*.

(2R,3S)-2-((tert-Butyldimethylsilyl)oxy)-3-phenylbutanoic acid (5h)



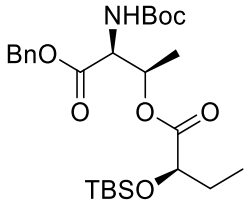
5h was prepared from **18** according to the procedure described for **5g**. FC (hexane/EtOAc 3:1) provided **5h** (338 mg, 86%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 1:1, UV, CPS): R_f = 0.45. **[α]$_D^{20}$** = +8.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.37 – 7.18 (m, 5H), 4.33 (d, *J* = 3.3 Hz, 1H), 3.34 (qd, *J* = 7.1, 3.3 Hz, 1H), 1.34 (d, *J* = 7.1 Hz, 3H), 0.86 (s, 9H), -0.15 (s, 3H), -0.36 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 176.1, 142.0, 128.6, 128.5, 128.3, 127.2, 77.3, 43.3, 25.8, 18.2, 13.2, -5.7. **IR** (thin film): υ (cm^{-1}) = 2955, 2929, 2885, 2858, 1721, 1472, 1463, 1454, 1362, 1256, 1147, 1114, 1099, 1055, 1011, 939, 887, 857, 837, 778, 761, 699, 671. **HR-MS** (ESI): Calcd. for $C_{16}H_{26}NaO_3Si$ $[M+Na]^+$ 317.1543 *m/z*; found 317.1543 *m/z*.

Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-((tert-butyldimethylsilyl)oxy)propanoyl)-L-threoninate (7a)



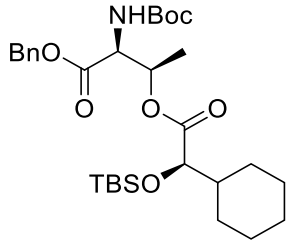
To a stirred solution of crude acid **5a** (433 mg, 0.873 mmol, 1.00 eq.) in toluene (20 mL) at rt were added Et_3N (0.92 mL, 6.62 mmol, 3.13 eq.) and 2,4,6-trichlorobenzoyl chloride (0.42 mL, 2.69 mmol, 1.27 eq.). After 5 min, *N*-Boc-L-Threonine benzyl ester **6** (691 mg, 2.23 mmol, 1.06 eq.) and DMAP (559 mg, 4.58 mmol, 2.16 eq.) were added and the suspension was stirred at rt for 18 h. The reaction was quenched with sat. aq. $NaHCO_3$ (20 mL). The aqueous layer was extracted with EtOAc (4 x 50 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 9:1 to 4:1) provided **7a** (322 mg, 12% over 2 steps) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 4:1, UV): R_f = 0.52. **$[\alpha]_D^{20}$** = +38.1° (*c* 0.67, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.29 (m, 5H), 5.46 (qd, *J* = 6.3, 2.5 Hz, 1H), 5.17 (d, *J* = 10.0 Hz, 1H), 5.13 (d, *J* = 12.2 Hz, 1H), 5.09 (d, *J* = 12.1 Hz, 1H), 4.47 (dd, *J* = 9.7, 2.4 Hz, 1H), 4.19 (q, *J* = 6.7 Hz, 1H), 1.46 (s, 9H), 1.30 (d, *J* = 6.4 Hz, 3H), 1.26 (d, *J* = 6.7 Hz, 3H), 0.90 (s, 9H), 0.08 (s, 3H), 0.04 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.9, 170.1, 156.0, 135.1, 128.8, 128.7, 128.6, 80.4, 71.1, 68.2, 67.8, 57.3, 28.4, 25.8, 21.4, 18.4, 17.1, -4.8, -5.1. **IR** (thin film): υ (cm^{-1}) = 3449, 2980, 2955, 2931, 2890, 2858, 1749, 1720, 1499, 1473, 1456, 1381, 1367, 1346, 1314, 1251, 1200, 1159, 1143, 1085, 1063, 1035, 975, 939, 902, 894, 867, 830, 813, 778, 748, 697, 667, 601, 582, 540, 534. **HR-MS** (ESI): Calcd. for $C_{25}H_{41}NNaO_7Si$ $[M+Na]^+$ 518.2544 *m/z*; found 518.2545 *m/z*.

Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-((tert-butyldimethylsilyl)oxy)butanoyl)-L-threoninate (7b)



7b was prepared from **6** and **5b** according to the procedure described for **7a**. FC (hexane/EtOAc 7:1) provided **7b** (933 mg, 61% over 2 steps) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc, 7:1, UV): R_f = 0.50. **[α]$_D^{20}$** = +44.4° (*c* 1.74, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.39 – 7.29 (m, 5H), 5.47 (qd, *J* = 6.3, 2.4 Hz, 1H), 5.18 (d, *J* = 9.9 Hz, 1H), 5.16 – 5.03 (m, 2H), 4.47 (dd, *J* = 9.7, 2.4 Hz, 1H), 4.05 (dd, *J* = 7.2, 4.6 Hz, 1H), 1.67 – 1.52 (m, 2H), 1.46 (s, 9H), 1.30 (d, *J* = 6.4 Hz, 3H), 0.91 (s, 9H), 0.90 – 0.86 (m, 3H), 0.06 (s, 3H), 0.03 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.6, 170.1, 156.0, 135.1, 128.8, 128.7, 128.6, 80.4, 73.1, 71.0, 67.8, 57.4, 28.4, 28.4, 25.8, 18.4, 17.2, 9.6, -4.8, -5.2. **IR** (thin film): υ (cm^{-1}) = 3448, 2956, 2931, 2887, 2857, 1752, 1721, 1499, 1456, 1366, 1316, 1289, 1250, 1163, 1142, 1084, 1065, 1005, 837, 779, 698. **HR-MS** (ESI): Calcd. for $C_{26}H_{44}NO_7Si$ $[M+H]^+$ 510.2882 *m/z*; found 510.2880 *m/z*.

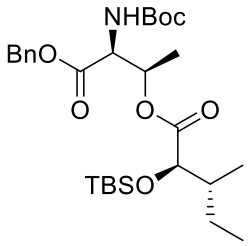
Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-((tert-butyldimethylsilyl)oxy)-2-cyclohexylacetyl)-L-threoninate (7c)



7c was prepared from **6** and **5c** according to the procedure described for **7a**. FC (hexane/EtOAc 95:5) provided **7c** (164 mg, 92%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 95:5, UV): R_f = 0.28. **[α]$_D^{20}$** = +42.1° (*c* 1.66, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.38 – 7.29 (m, 5H), 5.48 (qd, *J* = 6.2, 2.3 Hz, 1H), 5.25 – 5.11 (m, 2H), 5.02 (d, *J* = 12.2 Hz, 1H), 4.46 (dd, *J* = 9.7, 2.3 Hz, 1H), 3.91 (d, *J* = 4.5 Hz, 1H), 1.78 – 1.67 (m, 2H), 1.67 – 1.57 (m, 2H), 1.53 – 1.37 (m, 11H), 1.30 (d, *J* = 6.4 Hz, 3H), 1.24 – 1.03 (m, 5H), 0.91 (s, 9H), 0.03 (s, 3H), 0.01 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.2, 170.0, 156.0, 135.0, 128.7, 128.6, 128.4, 80.4,

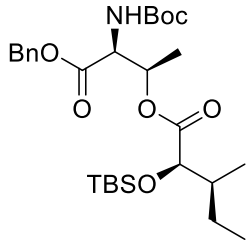
76.4, 71.1, 67.7, 57.4, 42.4, 29.5, 28.4, 27.0, 26.3, 26.2, 26.1, 25.9, 18.4, 17.2, -4.8, -5.2. **IR** (thin film): υ (cm^{-1}) = 2930, 2856, 1753, 1722, 1499, 1367, 1253, 1163, 1140, 1085, 1065, 837, 7783. **HR-MS** (ESI): Calcd. for $C_{30}H_{49}NNaO_7Si$ $[M+Na]^+$ 586.3171; found 586.3167 *m/z*.

(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl (2R,3R)-2-((tert-butyl-dimethylsilyl)oxy)-3-methylpentanoate (7d)



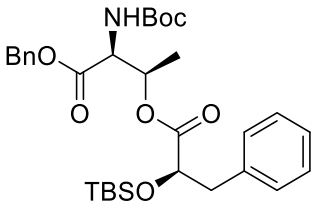
7d was prepared from **6** and **5d** according to the procedure described for **7a**. FC (hexane/EtOAc 40:1 to 4:1) provided **7d** (223 mg, 92%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.55. **$[\alpha]_D^{20}$** = +40.8° (*c* 0.70, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.30 (m, 5H), 5.49 (qd, *J* = 6.2, 2.0 Hz, 2H), 5.17 (d, *J* = 7.7 Hz, 1H), 5.10 (dd, *J* = 51.5, 12.2 Hz, 1H), 4.46 (dd, *J* = 9.7, 2.3 Hz, 1H), 3.97 (d, *J* = 4.4 Hz, 1H), 1.78 – 1.66 (m, 1H), 1.46 (s, 9H), 1.38 – 1.32 (m, 1H), 1.30 (d, *J* = 6.4 Hz, 3H), 1.22 – 1.11 (m, 1H), 0.91 (s, 9H), 0.90 – 0.86 (m, 6H), 0.04 (s, 3H), 0.02 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.3, 170.1, 156.0, 135.1, 128.8, 128.7, 128.5, 76.5, 71.1, 67.7, 57.4, 39.5, 28.4, 25.9, 23.8, 17.3, 15.6, 11.7, -4.8, -5.2. **IR** (thin film): υ (cm^{-1}) = 2962, 2932, 2858, 1752, 1723, 1253, 1165, 1143, 1066, 838, 779. **HR-MS** (ESI): Calcd. for $C_{28}H_{47}NNaO_7Si$ $[M+Na]^+$ 560.3014 *m/z*; found 560.3020 *m/z*.

(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl (2R,3S)-2-((tert-butyl-dimethylsilyl)oxy)-3-methylpentanoate (7e)



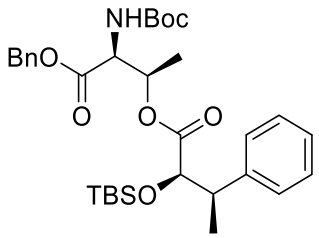
7e was prepared from **6** and **5e** according to the procedure described for **7a**. FC (hexane/EtOAc 40:1 to 4:1) provided **7d** (554 mg, 68%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.55. **[α]D^{20}** = +35.0° (*c* 0.75, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.39 – 7.32 (m, 5H), 5.48 (qd, *J* = 6.1, 2.5 Hz, 1H), 5.18 (d, *J* = 9.6 Hz, 1H), 5.10 (dd, *J* = 52.2, 12.2 Hz, 1H), 4.47 (dd, *J* = 9.7, 2.4 Hz, 1H), 4.07 (d, *J* = 3.2 Hz, 1H), 1.69 – 1.61 (m, 1H), 1.46 (s, 9H), 1.4 – 1.37 (m, 1H), 1.30 (d, *J* = 6.4 Hz, 3H), 1.26 – 1.16 (m, 1H), 0.92 – 0.90 (s, 9H overlapping with t, 3H), 0.79 (d, *J* = 6.8 Hz, 3H), 0.04 (s, 3H), 0.01 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.6, 170.1, 156.0, 135.0, 128.8, 128.6, 128.5, 80.4, 74.8, 71.1, 67.8, 57.4, 39.6, 28.4, 26.2, 25.9, 17.2, 13.6, 12.1, -4.7, -5.2. **IR** (thin film): υ (cm^{-1}) = 3460, 2961, 2933, 2859, 1752, 1723, 1499, 1461, 1367, 1252, 1164, 1146, 1065, 837, 776. **HR-MS** (ESI): Calcd. for $C_{28}H_{47}NNaO_7Si$ $[M+Na]^+$ 560.3014 *m/z*; found 560.3014 *m/z*.

Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-((tert-butyldimethylsilyl)oxy)-3-phenylpropanoyl)-L-threoninate (7f)



7f was prepared from **6** and **5f** according to the procedure described for **7a**. FC (hexane/EtOAc 4:1) provided **7f** (466 mg, 39%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc, 4:1, UV): R_f = 0.66. **$[\alpha]_D^{20}$** = +29.2° (*c* 1.58, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.38 – 7.30 (m, 5H), 7.30 – 7.25 (m, 2H), 7.23 – 7.16 (m, 3H), 5.49 (qd, *J* = 6.2, 2.3 Hz, 1H), 5.17 (d, *J* = 9.9 Hz, 1H), 5.14 (d, *J* = 12.3 Hz, 1H), 5.08 (d, *J* = 12.1 Hz, 1H), 4.48 (dd, *J* = 9.7, 2.3 Hz, 1H), 4.22 (dd, *J* = 9.2, 3.6 Hz, 1H), 2.89 (dd, *J* = 13.5, 3.6 Hz, 1H), 2.69 (dd, *J* = 13.5, 9.2 Hz, 1H), 1.47 (s, 9H), 1.28 (d, *J* = 6.4 Hz, 3H), 0.78 (s, 9H), -0.14 (s, 3H), -0.29 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.1, 170.0, 156.0, 137.4, 135.1, 130.0, 128.8, 128.7, 128.6, 128.3, 126.8, 80.5, 73.6, 71.3, 67.8, 57.3, 41.5, 28.4, 25.7, 18.3, 17.1, -5.3, -5.7. **IR** (thin film): υ (cm^{-1}) = 3448, 2953, 2931, 2857, 1751, 1719, 1498, 1457, 1365, 1314, 1252, 1162, 1128, 1082, 1064, 1000, 942, 832, 776, 754, 740, 699, 670. **HR-MS** (ESI): Calcd. for $C_{31}H_{45}NNaO_7Si$ $[M+Na]^+$ 594.2858 *m/z*; found 594.2860 *m/z*.

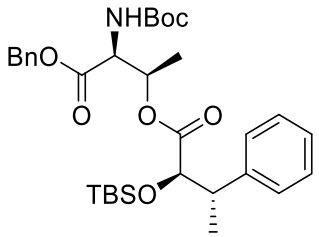
Benzyl N-(tert-butoxycarbonyl)-O-((2R,3R)-2-((tert-butyldimethylsilyl)oxy)-3-phenylbutanoyl)-L-threoninate (7g)



7g was prepared from **6** and **5g** according to the procedure described for **7a**. FC (hexane/EtOAc 5:1) provided **7g** (100 mg, 75%) as a colorless oil. **TLC** (SiO_2; hexane/EtOAc 5:1, UV, CPS): R_f = 0.32. **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.29 (m, 5H), 7.25 – 7.16 (m, 5H), 5.39 (qd, *J* = 6.4, 2.4 Hz, 1H), 5.21 – 4.98 (m, 3H), 4.40 (dd, *J* = 9.7, 2.4 Hz, 1H), 4.15 (d, *J* = 6.2 Hz, 1H), 3.06 (p, *J* = 7.0 Hz, 1H), 1.47 (s, 9H), 1.22 (d, *J* = 7.2 Hz, 3H), 1.11 (d, *J* = 6.4 Hz, 3H), 0.84

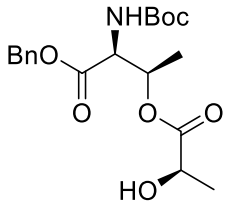
(s, 9H), -0.09 (s, 3H), -0.19 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 171.9, 170.1, 156.0, 142.4, 135.1, 128.8, 128.7, 128.5, 128.2, 126.9, 80.4, 77.4, 71.3, 67.8, 57.4, 44.3, 29.8, 28.4, 25.8, 25.7, 18.2, 17.9, 17.1, -5.1, -5.5.

Benzyl N-(tert-butoxycarbonyl)-O-((2R,3S)-2-((tert-butyldimethylsilyl)oxy)-3-phenylbutanoyl)-L-threoninate (7h)



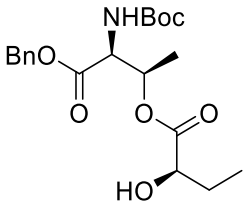
7h was prepared from **6** and **5h** according to the procedure described for **7a**. FC (hexane/EtOAc 5:1) provided **7h** (637 mg, 60%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 5:1, UV, $KMnO_4$): R_f = 0.46. **[α]$_D^{20}$** = +23.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.37 – 7.26 (m, 7H), 7.25 – 7.18 (m, 3H), 5.45 (m, 1H), 5.20 – 4.99 (m, 3H), 4.45 (dd, *J* = 9.6, 2.6 Hz, 1H), 4.21 (d, *J* = 3.9 Hz, 1H), 3.21 – 3.09 (m, 1H), 1.47 (s, 9H), 1.27 (d, *J* = 6.4 Hz, 3H), 1.22 (d, *J* = 7.1 Hz, 3H), 0.83 (s, 9H), -0.16 (s, 3H), -0.36 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 171.7, 170.0, 142.9, 138.0, 135.1, 129.2, 128.8, 128.7, 128.6, 128.4, 128.3, 126.9, 125.5, 80.5, 76.7, 71.4, 67.8, 57.4, 43.6, 28.5, 25.8, 17.3, 13.8, -5.3, -5.8. **IR** (thin film): υ (cm^{-1}) = 2956, 2931, 2894, 2857, 1753, 1721, 1498, 1472, 1455, 1383, 1367, 1313, 1284, 1254, 1163, 1146, 1114, 1098, 1085, 1064, 1010, 837, 778, 740, 699. **HR-MS** (ESI): Calcd. for $C_{32}H_{47}NNaO_7Si$ $[M+Na]^+$ 608.3014 *m/z*; found 608.3007 *m/z*.

Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-hydroxypropanoyl)-L-threoninate (8a)



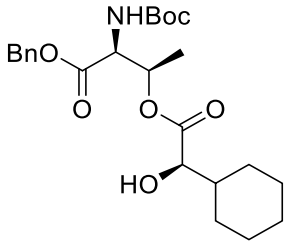
To a stirred solution of **7a** (241 mg, 0.486 mmol, 1.00 eq.) in THF (8.0 mL) was added HF·pyridine (70% HF, 30% pyridine, 2.00 mL, 76.0 mmol, 158 eq.) at 0 °C. The reaction was stirred at rt for 21 h and quenched by addition of sat. aq. $KHCO_3$ until pH 8 and of EtOAc (20 mL). The aqueous layer was extracted with EtOAc (3 x 50 mL) and the combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 3:1) provided **8a** (164 mg, 87%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 4:1, UV) R_f = 0.15. **$[\alpha]_D^{20}$** = +21.8° (*c* 0.57, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.45 – 7.29 (m, 5H), 5.48 (qd, *J* = 6.3, 2.6 Hz, 1H), 5.21 – 5.06 (m, 3H), 4.52 (dd, *J* = 9.6, 2.5 Hz, 1H), 4.15 (q, *J* = 6.8 Hz, 1H), 2.60 (s, 1H), 1.46 (s, 9H), 1.32 (d, *J* = 6.4 Hz, 3H), 1.22 (d, *J* = 6.9 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.7, 169.9, 155.9, 135.0, 128.8, 128.7, 80.7, 72.4, 67.9, 66.9, 57.2, 28.4, 20.5, 16.9. **IR** (thin film): υ (cm^{-1}) = 3441, 3357, 3065, 3034, 2980, 2936, 2875, 1744, 1717, 1521, 1500, 1456, 1392, 1382, 1368, 1348, 1315, 1280, 1253, 1214, 1164, 1128, 1085, 1063, 996, 939, 868, 837, 792, 771, 752, 732, 698. **HR-MS** (ESI): Calcd. for $C_{19}H_{27}NNaO_7$ $[M+Na]^+$ 404.1680 *m/z*; found 404.1678 *m/z*.

Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-hydroxybutanoyl)-L-threoninate (8b)



8b was prepared from **7b** according to the procedure described for **8a**. FC (hexane/EtOAc 3:2) provided **8b** (189 mg, quant.) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 3:2, UV): R_f = 0.51. **[α]$_D^{20}$** = +20.7° (*c* 1.02, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.42 – 7.29 (m, 5H), 5.48 (qd, *J* = 6.5, 2.6 Hz, 1H), 5.20 (d, *J* = 9.7 Hz, 1H), 5.17 – 5.06 (m, 2H), 4.52 (dd, *J* = 9.6, 2.7 Hz, 1H), 4.03 (ddd, *J* = 7.3, 5.9, 4.2 Hz, 1H), 2.62 (d, *J* = 5.9 Hz, 1H), 1.78 – 1.61 (m, 1H), 1.46 (s, 9H), 1.45 – 1.38 (m, 1H), 1.32 (d, *J* = 6.4 Hz, 3H), 0.89 (t, *J* = 7.4 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.2, 170.0, 155.9, 135.0, 128.8, 128.7, 80.6, 72.4, 71.8, 67.9, 57.2, 28.4, 27.6, 17.0, 9.1. **IR** (thin film): υ (cm^{-1}) = 3445, 3357, 2979, 2937, 1742, 1717,1500, 1456, 1391, 1367, 1316, 1246, 1212, 1164, 1128, 1084, 1063, 995, 865, 757, 697. **HR-MS** (ESI): Calcd. for $C_{20}H_{29}NNaO_7$ $[M+Na]^+$ 418.1836 *m/z*; found 418.1843 *m/z*.

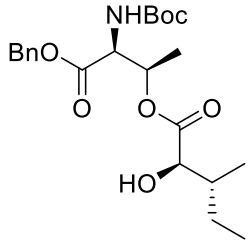
Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-cyclohexyl-2-hydroxyacetyl)-L-threoninate (8c)



8c was prepared from **7c** according to the procedure described for **8a**. FC (hexane/EtOAc 4:1) provided **8c** (126 mg, 96%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 4:1, UV): R_f = 0.20. **[α]$_D^{20}$** = +28.5° (*c* 2.02, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.39 – 7.29 (m, 5H), 5.49 (qd, *J* = 6.2, 2.4 Hz, 1H), 5.26 (d, *J* = 9.6 Hz, 1H), 5.16 (d, *J* = 12.2 Hz, 1H), 5.05 (d, *J* = 12.2 Hz, 1H), 4.52 (dd, *J* = 9.6, 2.3 Hz, 1H), 3.94 (d, *J* = 3.2 Hz, 1H), 2.64 (br. s, 1H), 1.82 – 1.53 (m, 5H), 1.46 (s, 9H), 1.32 (d, *J* = 6.4 Hz, 3H), 1.30 – 1.04 (m, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ = 173.7, 169.9, 155.9, 134.9, 128.8, 128.7, 128.4, 80.5, 75.0, 72.5, 67.7, 57.2, 41.8, 29.2, 28.4, 26.3, 26.1, 26.0, 26.0, 17.0. **IR** (thin film): υ (cm^{-1}) = 2929, 2855, 1716, 1499, 1453, 1367, 1346,

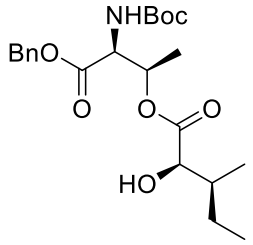
1314, 1284, 1250, 1212, 1163, 1116, 1086, 1063, 999, 938, 866, 753, 697. **HR-MS** (ESI): Calcd. for $C_{24}H_{35}NNaO_7$ $[M+Na]^+$ 472.2306 *m/z*; found 472.2308 *m/z*.

(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl (2R,3R)-2-hydroxy-3-methylpentanoate (8d)



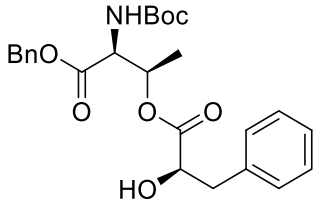
8d was prepared from **7d** according to the procedure described for **8a**. FC (hexane/EtOAc 6:1 to 4:1) provided **8d** (41.0 mg, quant.) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.39. $[\alpha]_D^{20}$ = +20.2° (*c* 1.04, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.32 (m, 5H), 5.50 (qd, *J* = 6.3, 2.3 Hz, 1H),), 5.17 (d, *J* = 6.5 Hz, 1H), 5.12 (dd, *J* = 37.7, 12.3 Hz, 2H), 4.51 (dd, *J* = 9.6, 2.4 Hz, 1H), 3.99 (dd, *J* = 6.1, 3.6 Hz, 1H), 2.55 (d, *J* = 6.1 Hz, 1H), 1.77 – 1.67 (m, 1H), 1.46 (s, 9H), 1.33 (d, *J* = 6.4 Hz, 3H), 1.19 (m, 2H), 0.95 (d, *J* = 6.9 Hz, 3H), 0.88 (t, *J* = 7.4 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 173.9, 169.9, 155.94, 135.0, 128.8, 128.8, 128.8, 128.5, 80.6, 75.3, 72.5, 67.8, 57.3, 38.9, 28.4, 23.5, 17.1, 15.4, 11.8. **IR** (neat, cm^{-1}): 3456, 2965, 2939, 2879, 1723, 1498, 1453, 1365, 1251, 1215, 1164, 1139, 1061. **HR-MS** (ESI): Calcd. for $C_{22}H_{33}NNaO_7$ $[M+Na]^+$ 446.2149 *m/z*; found 446.2147 *m/z*.

(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl (2R,3S)-2-hydroxy-3-methylpentanoate (8e)



8e was prepared from **7e** according to the procedure described for **8a**. FC (hexane/EtOAc 40:1 to 10:1) provided **8e** (41.0 mg, quant.) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 7:3, UV): R_f = 0.37. **$[\alpha]_D^{20}$** = +37.8° (*c* 0.45, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.39 – 7.32 (m, 5H), 5.49 (dd, *J* = 6.4, 2.6 Hz, 1H), 5.21 (d, *J* = 9.6 Hz, 1H), 5.12 (dd, *J* = 39.8, 12.2 Hz, 2H), 4.52 (dd, *J* = 9.6, 2.4 Hz, 1H), 4.10 (dd, *J* = 5.8, 2.7 Hz, 1H), 2.60 (d, *J* = 5.7 Hz, 1H), 1.72 – 1.62 (m, 1H), 1.53 – 1.47 (m, 1H), 1.46 (s, 9H), 1.33 (d, *J* = 6.4 Hz, 3H), 1.31 – 1.23 (m, 1H), 0.93 (t, *J* = 7.4 Hz, 3H), 0.72 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.4, 169.9, 155.9, 134.9, 128.8, 128.8, 128.5, 80.6, 73.1, 72.6, 67.9, 57.3, 38.4, 28.4, 26.1, 17.0, 13.1, 11.9. **IR** (thin film): υ (cm^{-1}) = 3456, 2968, 2936, 1741, 1720, 1501, 1457, 1383, 1316, 1249, 1214, 1165, 1064. **HR-MS** (ESI): Calcd. for $C_{22}H_{33}NNaO_7$ $[M+Na]^+$ 446.2149 *m/z*; found 446.2141 *m/z*.

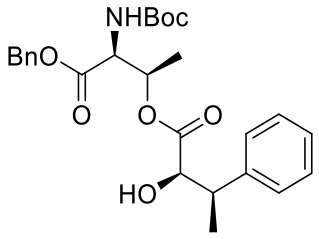
Benzyl N-(tert-butoxycarbonyl)-O-((R)-2-hydroxy-3-phenylpropanoyl)-L-threoninate (8f)



8f was prepared from **7f** according to the procedure described for **8a**. FC (hexane/EtOAc 4:1) provided **8f** (161 mg, 82%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 4:1, UV): R_f = 0.23.

[α]D20 = +17.4° (*c* 1.90, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.39 – 7.33 (m, 5H), 7.32 – 7.27 (m, 2H), 7.26 – 7.21 (m, 1H), 7.20 – 7.16 (m, 2H), 5.48 (qd, *J* = 6.3, 2.6 Hz, 1H), 5.20 – 5.09 (m, 3H), 4.52 (dd, *J* = 9.7, 2.4 Hz, 1H), 4.34 – 4.27 (m, 1H), 2.93 (dd, *J* = 14.1, 4.5 Hz, 1H), 2.67 (dd, *J* = 14.1, 7.8 Hz, 1H), 2.59 (d, *J* = 5.0 Hz, 1H), 1.47 (s, 9H), 1.29 (d, *J* = 6.4 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) =173.2, 170.0, 155.9, 136.5, 135.0, 129.5, 128.8, 128.8, 128.7, 128.6, 127.0, 80.6, 72.6, 71.5, 67.9, 57.2, 40.5, 28.4, 16.9. **IR** (thin film): υ (cm^{-1}) = 3403, 2978, 2935, 1741, 1715, 1497, 1455, 1367, 1314, 1248, 1162, 1087, 1062, 1031, 994, 864, 748, 698. **HR-MS** (ESI): Calcd. for $C_{25}H_{31}NNaO_7$ $[M+Na]^+$ 480.1993 *m/z*; found 480.1990 *m/z*.

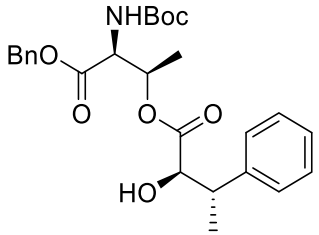
Benzyl N-(tert-butoxycarbonyl)-O-((2R,3R)-2-hydroxy-3-phenylbutanoyl)-L-threoninate (8g)



8g was prepared from **7g** according to the procedure described for **8a**. FC (hexane/EtOAc 3:1) provided **8g** (281 mg, 73%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 3:1, UV, CPS): R_f = 0.34. **[α]D20** = +26.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.27 (m, 7H), 7.25 – 7.18 (m, 3H), 5.37 (qd, *J* = 6.2, 1.9 Hz, 1H), 5.22 – 5.07 (m, 2H), 5.03 (d, *J* = 9.7 Hz, 1H), 4.46 (dd, *J* = 9.7, 2.6 Hz, 1H), 4.23 (dd, *J* = 6.9, 4.0 Hz, 1H), 3.18 – 3.06 (m, 1H), 2.44 (d, *J* = 7.0 Hz, 1H), 1.47 (s, 9H), 1.35 (d, *J* = 7.2 Hz, 3H), 1.18 (d, *J* = 6.4 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.7, 170.0, 158.3, 155.9, 140.7, 135.0, 128.8, 128.6, 128.5, 128.4, 128.3, 127.3, 80.6, 75.7, 72.6, 67.9, 57.2, 43.3, 28.4, 17.3, 17.0. **IR** (thin film): υ (cm^{-1}) = 2978, 2933, 1742, 1716, 1497, 1455, 1383, 1367, 1315, 1249, 1215, 1164, 1123, 1086, 1062, 1028, 993,

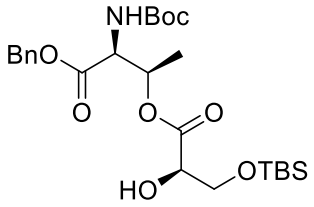
866, 754, 699, 541. **HR-MS** (ESI): Calcd. for $C_{26}H_{33}NNaO_7$ $[M+Na]^+$ 494.2149 *m/z*; found 494.2158 *m/z*.

Benzyl N-(tert-butoxycarbonyl)-O-((2R,3S)-2-hydroxy-3-phenylbutanoyl)-L-threoninate (8h)



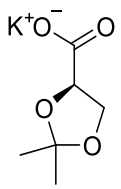
8h was prepared from **7h** according to the procedure described for **8a**. FC (hexane/EtOAc 3:1) provided **8h** (396 mg, 77%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 3:1, UV, CPS): R_f = 0.40. **$[\alpha]_D^{20}$** = +12.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.37 – 7.28 (m, 9H), 7.27 – 7.19 (m, 1H), 5.49 (qd, *J* = 6.5, 3.1 Hz, 1H), 5.26 – 4.99 (m, 3H), 4.51 (dd, *J* = 9.6, 2.7 Hz, 1H), 4.27 (s, 1H), 3.12 – 3.01 (m, 1H), 2.65 (s, 1H), 1.47 (s, 9H), 1.30 (d, *J* = 6.5 Hz, 3H), 1.19 (d, *J* = 7.1 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 173.2, 170.0, 155.9, 142.8, 136.0, 128.8, 128.6, 128.3, 128.0, 127.0, 80.7, 74.9, 72.8, 67.9, 57.2, 43.0, 28.4, 17.0, 14.6. **IR** (thin film): υ (cm^{-1}) = 3408, 2978, 2935, 1742, 1716, 1498, 1455, 1384, 1367, 1315, 1249, 1213, 1164, 1126, 1086, 1063, 988, 768, 699. **HR-MS** (ESI): Calcd. for $C_{26}H_{33}NNaO_7$ $[M+Na]^+$ 494.2149 *m/z*; found 494.2150 *m/z*.

Benzyl N-(tert-butoxycarbonyl)-O-((R)-3-((tert-butyldimethylsilyl)oxy)-2-hydroxypropanoyl)-L-threoninate (8i)



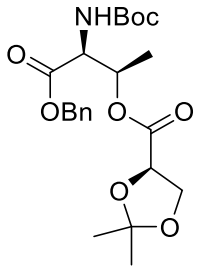
12A (80.0 mg, 201 µmol, 1.00 eq.), TBSCl (32.0 mg, 211 µmol, 1.05 eq.) and imidazole (18.0 mg, 211 µmol, 1.05 eq) were dissolved in DCM (1.5 mL) at rt. The mixture was stirred at rt for 18 h and was partitioned between sat. aq. NH_4Cl (3 mL) and Et_2O (5 mL). The aqueous layer was extracted with Et_2O (3 x 5 mL), the combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 9:1 to 2:3) provided **8i** (61.4 mg, 60%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 2:3, UV): R_f = 0.29. **$[\alpha]_D^{20}$** = +5.4° (*c* 0.63, MeOH). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.38 – 7.31 (m, 5H), 5.47 (dt, *J* = 6.0, 4.6 Hz, 1H), 5.31 – 5.22 (m, 1H), 5.18 – 5.07 (m, 2H), 4.49 (dd, *J* = 9.5, 2.1 Hz, 1H), 4.13 – 4.06 (m, 1H), 3.78 (dd, *J* = 3.5, 2.8 Hz, 2H), 1.45 (s, 9H), 1.32 (d, *J* = 6.4 Hz, 3H), 0.88 (s, 9H), 0.07 (s, 3H), 0.07 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 171.4, 169.8, 155.8, 135.0, 135.0, 128.6, 128.5, 128.5, 80.4, 72.2, 71.9, 67.6, 65.0, 57.1, 28.3, 25.9, 17.0, -5.3, -5.4. **IR** (thin film): υ (cm^{-1}) = 2933, 2856, 1745, 1718, 1504, 1461, 1370, 1313, 1250, 1165, 1122, 1063, 1001, 838, 779, 743, 696. **HR-MS** (ESI): Calcd. for $C_{25}H_{41}NNaO_8Si$ $[M+Na]^+$ 534.2494 *m/z*; found 534.2491 *m/z*.

Potassium (R)-2,2-dimethyl-1,3-dioxolane-4-carboxylate (11)[28]



To a stirred solution of (*R*)-2,2-dimethyl-1,3-dioxolane-4-carboxylic acid (**10**) (226 µL, 1.56 mmol, 1.00 eq.) in MeOH (4 mL) was added aq. 2M KOH (780 µL, 1.56 mmol, 1.00 eq.). The solution was stirred for 1 h at rt. The solvents were removed *in vacuo* to deliver **11** (285 mg, 99%) as a white wax. **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 4.49 – 4.37 (m, 1H), 4.26 – 4.16 (m, 1H), 3.87 (dd, *J* = 7.9, 7.4 Hz, 1H), 1.44 (s, 3H), 1.36 (s, 3H). **^{13}C-NMR** (101 MHz, CD_3OD): δ (ppm) = 178.2, 111.1, 77.5, 69.0, 26.4, 27.0. **HR-MS** (ESI): Calcd. for $C_6H_9O_4$ $[M-H]^-$ 145.0506 *m/z*; found 145.0503 *m/z*. The analytical data are in agreement with those reported in *J. Agric. Food Chem.* **2010**, *58*(10), 6458-6464.

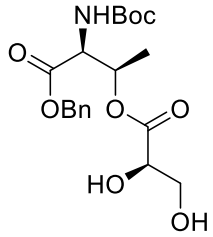
(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl (R)-2,2-dimethyl-1,3-dioxolane-4-carboxylate (12)



To a stirred solution of **11** (102 mg, 554 µmol, 1.00 eq.) and Et_3N (162 µL, 1.16 mmol, 2.10 eq.) in THF (1 mL) and DMF (1.5 mL) was added 2,4,6-trichlorobenzoyl chloride (90.9 µL, 581 µmol, 1.05 eq.). After the solution became turbid (5 min), **6** (180 mg, 581 µmol, 1.05 eq.) and DMAP (67.6 mg, 554 µmol, 1.00 eq.) were added at rt. The mixture was stirred for 18 h at rt. Sat. aq. $NaHCO_3$ (3 mL) was added and the aqueous layer was extracted with EtOAc (3 x 5 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 4:1) provided **12** (159 mg, 71%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 4:1, UV): R_f = 0.28. **$[\alpha]_D^{20}$** = +23.3° (*c* 2.19, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.36 –

7.27 (m, 5H), 5.47 (dt, *J* = 8.6, 4.9 Hz, 1H), 5.20 (d, *J* = 9.6 Hz, 1H), 5.09 (d, *J* = 2.8 Hz, 2H), 4.48 (dd, *J* = 9.6, 2.2 Hz, 1H), 4.39 (dd, *J* = 7.2, 5.0 Hz, 1H), 4.14 – 4.03 (m, 1H), 3.93 – 3.81 (m, 1H), 1.43 (s, 9H), 1.43 (s, 3H), 1.35 (s, 3H), 1.28 (d, *J* = 6.5 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 170.0, 169.9, 155.7, 134.9, 128.6, 128.6, 111.3, 80.3, 73.8, 71.6, 67.7, 67.1, 57.0, 28.2, 25.8, 25.5, 16.8. **IR** (thin film): υ (cm^{-1}) = 2981, 1744, 1715, 1500, 1456, 1382, 1369, 1315, 1248, 1198, 1162, 1103, 1063, 995, 865, 842, 753, 699, 606, 580, 517, 491, 467. **HR-MS** (ESI): Calcd. for $C_{22}H_{31}NNaO_8$ $[M+H]^+$ 460.1942 *m/z*; found 460.1942 *m/z*.

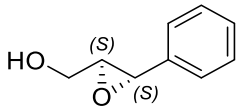
Benzyl N-(tert-butoxycarbonyl)-O-((R)-2,3-dihydroxypropanoyl)-L-threoninate (12A)



To a stirred solution of **12** (20.0 mg, 4.57 µmol, 1.00 eq.) in MeOH (1 mL) was added *p*-TsOH (1.00 mg, 0.46 µmol, 0.10 eq.). The solution was stirred at rt for 2 h but was not complete. *p*-TsOH (4.5 mg, 1.84 µmol, 0.40 eq.) was added and the solution was stirred at rt for 18 h. Et_3N (8 µL) was added and the solvents were removed *in vacuo*. FC (hexane/EtOAc 2:3) provided **12A** (12.5 mg, 69%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 2:3, UV): R_f = 0.20. **[α]$_D^{20}$** = +39.1° (*c* 0.63, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.40 – 7.32 (m, 5H), 5.58 – 5.48 (m, 1H), 5.31 (d, *J* = 9.4 Hz, 1H), 5.19 – 5.09 (m, 2H), 4.52 (d, *J* = 9.3 Hz, 1H), 4.17 (t, *J* = 3.4 Hz, 1H), 3.75 (d, *J* = 2.9 Hz, 2H), 1.46 (s, 9H), 1.34 (d, *J* = 6.5 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.0, 170.5, 155.8, 134.8, 128.7, 80.6, 72.6, 71.7, 68.1, 64.1, 57.0, 28.3, 16.9. **IR** (thin film): υ (cm^{-1}) = 3436, 2978, 2937, 1742, 1714, 1503, 1456, 1385, 1367, 1315, 1250, 1213, 1164,

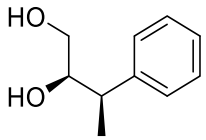
1115, 1062, 998, 865, 753, 699, 581, 493, 462. **HR-MS** (ESI): Calcd. for $C_{19}H_{27}NNaO_8$ $[M+Na]^+$ 420.1629 *m/z*; found 420.1632 *m/z*.

((2S,3S)-3-Phenyloxiran-2-yl)methanol (14)



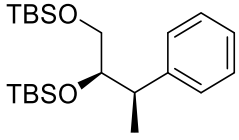
To a stirred solution of L-(+)-DET (1.46 mL, 8.57 mmol, 0.23 eq.) in DCM (340 mL) with activated molecular sieves (3 Å, 5 g) was added titanium(IV)isopropoxide (1.65 mL, 5.59 mmol, 0.15 eq.) and *tert*-butyl hydroperoxide (5.5M in DCM, 13.6 mL, 74.53 mmol, 2.00 eq.) at –20 °C. The mixture was stirred for 1 h at –20 °C. Then, a solution of *trans*-cinnamyl alcohol (**13**) (5.00 g, 37.3 mmol, 1.00 eq.) in DCM (7.45 mL) was added dropwise and the reaction mixture was stirred for 16 h at –20 °C. The reaction was quenched by the addition of 10% aq. NaOH solution (ca. 50 mL) and NaCl (ca. 50 g). After stirring the reaction mixture for 10 min, it was filtered through a pad of celite. The aqueous layer was extracted with EtOAc (3x 150 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (toluene/EtOAc 3:1) provided **14** (4.14 g, 74%) as a white solid. **TLC** (SiO_2, hexane/EtOAc 1:1, UV, $KMnO_4$): R_f = 0.36. $[\alpha]_D^{20}$ = –50.8° (*c* 1.00, $CHCl_3$) {lit.[45] $[\alpha]_D^{20}$ = –49.6° (*c* 2.4, $CHCl_3$)}. **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.39 – 7.27 (m, 5H), 4.06 (dddd, *J* = 12.8, 5.2, 2.4, 0.5 Hz, 1H), 3.93 (d, *J* = 2.2 Hz, 1H), 3.81 (ddd, *J* = 12.7, 7.8, 3.8 Hz, 1H), 3.23 (dt, *J* = 3.8, 2.3 Hz, 1H), 1.72 (dd, *J* = 7.9, 5.2 Hz, 1H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 136.8, 128.7, 128.5, 125.9, 62.5, 61.4, 55.7.

(2R,3R)-3-Phenylbutane-1,2-diol (15)



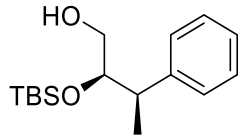
To a stirred suspension of CuCN (1.79 g, 20.0 mmol, 2.00 eq.) in Et_2O (200 mL) was slowly added MeLi (3.1M in THF, 11.6 mL, 36.0 mmol, 3.60 eq.) at –78 °C. The mixture was stirred at –78 °C for 1 h and a solution of **14** (1.50 g, 10.0 mmol, 1.00 eq.) in Et_2O (5 mL) was added slowly to the reaction mixture. The reaction was allowed to warm to 0 °C (over 1 h). The reaction mixture was stirred for 3 h at 0 °C while it was turning into a deep yellow suspension. The reaction was quenched by the addition of sat. aq. NH_4Cl solution (ca. 60 mL) and the mixture was extracted with EtOAc (3x 100 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 1:1) provided **15** (1.23 g, 74%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 1:1, UV, CPS): R_f = 0.16. **$[\alpha]_D^{20}$** = +14.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.38 – 7.30 (m, 2H), 7.25 (tdd, *J* = 5.4, 3.3, 2.0 Hz, 3H), 3.84 – 3.71 (m, 2H), 3.61 – 3.50 (m, 1H), 2.87 (p, *J* = 7.2 Hz, 1H), 1.30 – 1.24 (m, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 143.2, 129.0, 128.9, 128.1, 127.1, 76.4, 64.7, 43.0, 18.0. **IR** (thin film): υ (cm^{-1}) = 3372, 3028, 2965, 2930, 2878, 1495, 1453, 1379, 1335, 1098, 1083, 1047, 1030, 1014, 1002, 943, 910, 864, 777, 763, 678, 607, 573, 558, 515, 504, 486, 476, 456, 445, 415. **HR-MS** (ESI): Calcd. for $C_{10}H_{14}NaO_2$ $[M+Na]^+$ 189.0886 *m/z*; found 189.0888 *m/z*.

(R)-2,2,3,3,8,8,9,9-Octamethyl-5-((R)-1-phenylethyl)-4,7-dioxa-3,8-disiladecane (15A)



To a stirred solution of the **15** (157 mg, 0.95 mmol, 1.00 eq.) and imidazole (386 mg, 5.67 mmol, 6.00 eq.) in DMF (1.9 mL) was added TBSCl (428 mg, 2.84 mmol, 3.00 eq.) at 0 °C. The reaction mixture was allowed to warm to rt (over 1 h) and then stirred for 15 h. The reaction was quenched by the addition of sat. aq. $NHCl_4$ solution (ca. 5 mL) and extracted with EtOAc (3x 15 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 10:1) provided **15A** (348 mg, 93%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 9:1, UV, CPS): R_f = 0.79. **[α]$_D^{20}$** = +15.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.33– 7.14 (m, 5H), 3.75 (ddd, *J* = 6.8, 5.1, 4.1 Hz, 1H), 3.41 (dd, *J* = 10.0, 5.1 Hz, 1H), 3.20 (dd, *J* = 10.0, 6.9 Hz, 1H), 3.03 (dd, *J* = 7.3, 4.1 Hz, 1H), 1.30 (d, *J* = 7.3 Hz, 3H), 0.89 (s, 9H), 0.86 (s, 9H), 0.02 (s, 3H), -0.01 (s, 3H),. -0.02 (s, 3H), -0.02 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 143.5, 129.3, 127.8, 126.1, 77.2, 65.3, 42.1, 26.1, 26.1, 18.5, 18.4, 18.3, -4.1, -4.9, -5.3. **IR** (thin film): υ (cm^{-1}) = 2955, 2928, 2885, 2857, 1495, 1471, 1463, 1388, 1362, 1254, 1122, 1096, 1058, 1039, 1020, 1005, 988, 952, 939, 878, 831, 813, 773, 698, 672, 551, 516, 467, 456, 439, 422, 411. **HR-MS** (ESI): Calcd. for $C_{22}H_{42}NaO_2Si_2$ $[M+Na]^+$ 417.2616 *m/z*; found 417.2617 *m/z*.

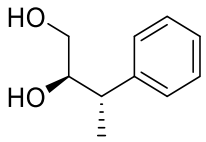
(2R,3R)-2-((tert-Butyldimethylsilyl)oxy)-3-phenylbutan-1-ol (16)



To a stirred solution of **15A** (233 mg, 0.59 mmol, 1.00 eq.) in a mixture of MeOH/DCM 1:1 (12 mL) was added camphorsulfonic acid (27.4 mg, 0.12 mmol, 0.20 eq.) at 0 °C. The mixture was stirred at 0 °C for 5 h. The reaction was quenched with sat. aq. $NaHCO_3$ solution (4 mL) and the mixture was extracted with DCM (3x ca. 15 mL). The combined organic layers were dried over

$MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 3:1 to 1:5) provided **16** (99.3 mg, 60%) as a colorless oil along with **15** (24.1 mg, 24 %) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 9:1, UV, CPS): R_f = 0.22. **$[\alpha]_D^{20}$** = +17.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.32 – 7.17 (m, 5H), 3.78 (dt, *J* = 6.8, 4.2 Hz, 1H), 3.62 (dd, *J* = 11.3, 4.4 Hz, 1H), 3.50 (dd, *J* = 11.3, 3.9 Hz, 1H), 3.00 (p, *J* = 7.1 Hz, 1H), 1.28 (d, *J* = 7.2 Hz, 3H), 0.83 (s, 9H), -0.02 (s, 3H), -0.26 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 144.1, 128.5, 128.3, 126.5, 77.5, 64.2, 43.0, 26.0, 17.0, -4.5, -5.1. **IR** (thin film): υ (cm^{-1}) = 2955, 2929, 2884, 2857, 1495, 1471, 1463, 1454, 1389, 1362, 1254, 1104, 1056, 1021, 938, 866, 834, 812, 775, 700, 674, 411. **HR-MS** (ESI): Calcd. for $C_{16}H_{28}NaO_2Si$ $[M+Na]^+$ 303.1751 *m/z*; found 303.1750 *m/z*.

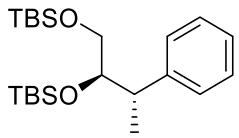
(2R,3S)-3-Phenylbutan-1,2-diol (17)



To a stirred solution of Me_3Al (2M in toluene, 10.0 mL, 20.0 mmol, 3.00 eq.) was slowly added a solution of **14** (1.00 g, 6.66 mmol, 1.00 eq.) in toluene (33 mL) at 0 °C. The mixture was stirred at 0 °C for 2 h. The reaction was quenched at 0 °C with sat. aq. Rochelle's salt solution (25 mL) and the mixture diluted with EtOAc (30 mL). The resulting biphasic mixture was stirred at rt for 1 h. The aqueous layer was extracted with Et_2O (3x 50mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 1:3) provided **17** (*d.r.* = 3.4:1, 853 mg, 77%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 1:3, UV, $KMnO_4$). R_f = 0.33. **^{1}H-NMR** (400 MHz, $CDCl_3$) δ 7.37 – 7.15 (m, 5H), 3.83 – 3.74 (m, 1H), 3.50 – 3.44 (m, 1H), 3.37 (m, 1H), 2.85 – 2.74 (m, 1H), 1.37 (d, *J* = 7.0 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$) δ 143.7, 130.6, 128.9, 128.8, 128.3, 128.1, 127.7, 126.8, 76.8, 65.2, 43.0, 18.0. **IR** (thin film): (cm^{-1}) =

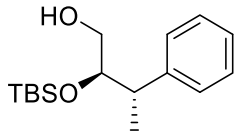
3361, 2965, 2931, 2879, 1494, 1453, 1087, 1061, 1015, 763, 701. **HR-MS** (ESI): Calcd. for $C_{10}H_{14}NaO_2$ $[M+Na]^+$ 189.0886 *m/z*; found 189.0891 *m/z*.

(R)-2,2,3,3,8,8,9,9-Octamethyl-5-((S)-1-phenylethyl)-4,7-dioxa-3,8-disiladecane (17A)



17A was prepared from **17** according to the procedure described for **15A**. FC (hexane/EtOAc 95:5) provided **17A** (*d.r.* = 7.73:1, 9.02 g, 88%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 95:5, UV, CPS): R_f = 0.71. **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.28 – 7.14 (m, 5H), 3.73 (dt, *J* = 6.4, 5.0 Hz, 1H), 3.52 – 3.35 (m, 2H), 3.06 (qd, *J* = 7.0, 4.4 Hz, 1H), 1.26 (d, *J* = 7.1 Hz, 3H), 0.89 (d, *J* = 1.7 Hz, 9H), 0.85 (s, 9H), 0.02 (s, 3H), 0.01 (s, 3H), -0.08 (s, 3H), -0.30 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 145.6, 129.3, 128.3, 128.1, 127.8, 126.1, 77.9, 65.4, 41.2, 26.1, 18.3, 14.1, -4.2, -5.2, -5.3. **IR** (thin film): υ (cm^{-1}) = 2955, 2929, 2885, 2857, 1494, 1472, 1463, 1388, 1362, 1254, 1133, 1088, 1020, 1006, 956, 939, 878, 834, 813, 775, 699, 665. **HR-MS** (ESI): Calcd. for $C_{22}H_{42}NaO_2Si_2$ $[M+Na]^+$ 417.2616 *m/z*; found 417.2628 *m/z*.

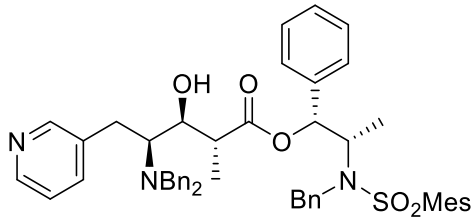
(2R,3S)-2-((tert-Butyldimethylsilyl)oxy)-3-phenylbutan-1-ol (18)



18 was prepared from **17A** according to the procedure described for **16**. FC (hexane/EtOAc 9:1) provided **18** (374 mg, 48%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 9:1, UV, $KMnO_4$): R_f = 0.25. **$[\alpha]_D^{20}$** = +18.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.32 – 7.26 (m, 2H), 7.25 – 7.17 (m, 3H), 3.74 (dt, *J* = 7.7, 3.7 Hz, 1H), 3.45 (dt, *J* = 11.3, 4.0 Hz, 1H), 3.30 (ddd,

J = 11.4, 8.4, 3.5 Hz, 1H), 3.01 (m 1H), 1.71 (dd, *J* = 8.4, 4.2 Hz, 1H), 1.31 (d, *J* = 7.1 Hz, 3H), 0.93 (s, 9H), 0.08 (s, 3H), 0.04 (s, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 144.3, 128.6, 128.3, 128.1, 126.6, 77.8, 64.6, 42.5, 26.1, 26.0, 17.9, -4.2, -4.6. **IR** (thin film): υ (cm^{-1}) = 2955, 2929, 2884, 2857, 1494, 1471, 1463, 1389, 1362, 1254, 1129, 1107, 1050, 1017, 983, 937, 865, 834, 812, 775, 700, 676. **HR-MS** (ESI): Calcd. for $C_{16}H_{28}NaO_2Si$ $[M+Na]^+$ 303.1751 *m/z*; found 303.1752 *m/z*.

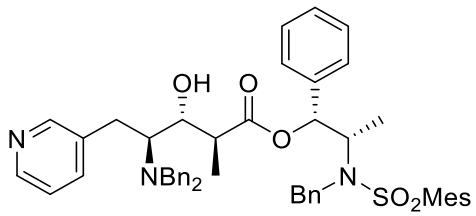
(1R,2S)-2-((N-Benzyl-2,4,6-trimethylphenyl)sulfonamido)-1-phenylpropyl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (23)



To a solution of **22** (1.10 g, 2.29 mmol, 1.50 eq.) in anhydrous DCM (5 mL) was added Et_3N (0.80 mL, 5.73 mmol, 3.75 eq.) at rt. The reaction mixture was cooled to –78 °C and dicyclohexylboron trifluoromethanesulfonate (1.64 g, 5.04 mmol, 3.30 eq.) in hexane (5.0 mL) was added dropwise during 15 min. The resulting solution was stirred at –78 °C for 3 h (colorless/light yellow solution). A solution of **20** (505 mg, 1.53 mmol, 1.00 eq.) in DCM (5.0 mL) was added dropwise during 25 min (syringe pump, 0.05 mL/min) and the solution was stirred at –78 °C for 3 h (orange solution). The mixture was allowed to warm very slowly to 0 °C (around 1 h) and stirred at 0 °C for 1 h (the acetone of the cooling bath was transferred into another less isolating beaker while the dry ice was left behind. Thereby, the temperature rose only very slowly. At –5 °C, the acetone bath was replaced by a 0 °C water/ice bath). The reaction was then quenched with phosphate buffer pH 7 (9 mL). The mixture was diluted with MeOH (30 mL) and 30% H_2O_2

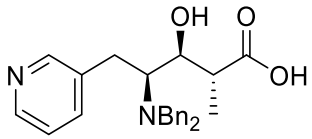
(3 mL) was added carefully. The mixture was stirred at rt for 18 h and then concentrated *in vacuo*. The residue was taken up in DCM and H_2O and the aqueous layer was extracted with DCM (3 x 25 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 3:2) provided **23** (559 mg, 0.69 mmol, 45%) as a white foam and bis-*epi*-**23** (223 mg, 0.28 mmol, 18%) as a white foam. **TLC** (SiO_2, hexane/EtOAc 1:1, UV): R_f = 0.35. **$[\alpha]_D^{20}$** = +23.0° (*c* 1.33, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.46 (br. s, 1H), 8.40 (d, *J* = 4.1 Hz, 1H), 7.46 (dt, *J* = 7.9, 2.0 Hz, 1H), 7.36 – 7.23 (m, 7H), 7.23 – 7.01 (m, 12H), 6.84 – 6.80 (m, 4H), 5.77 (d, *J* = 3.9 Hz, 1H), 4.68 (d, *J* = 16.5 Hz, 1H), 4.50 (d, *J* = 16.6 Hz, 1H), 4.18 (d, *J* = 13.4 Hz, 2H), 4.08 (qd, *J* = 7.0, 3.9 Hz, 1H), 3.56 (br. s, 1H), 3.42 (d, *J* = 13.4 Hz, 2H), 3.36 – 3.27 (m, 1H), 3.12 – 2.98 (m, 3H), 2.85 (ddd, *J* = 9.6, 5.6, 4.1 Hz, 1H), 2.44 (s, 6H), 2.23 (s, 3H), 1.10 (d, *J* = 7.0 Hz, 3H), 0.42 (d, *J* = 7.1 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.0, 150.6, 147.6, 142.6, 140.3, 139.8, 138.4, 138.4, 136.8, 135.7, 133.6, 132.2, 129.2, 128.5, 128.3, 128.0, 127.6, 127.3, 127.1, 125.9, 123.6, 78.4, 73.0, 59.0, 56.9, 55.7, 48.3, 42.7, 26.8, 23.0, 21.0, 13.1, 13.0. **IR** (thin film): υ (cm^{-1}) = 3667, 3018, 2977, 1731, 1604, 1496, 1454, 1379, 1321, 1215, 1152, 1077, 1055, 930, 855, 752, 701, 667. **HR-MS** (ESI): Calcd for $C_{50}H_{56}N_3O_5S$ $[M+H]^+$ 810.3935 *m/z*, found 810.3924 *m/z*.

(1R,2S)-2-((N-Benzyl-2,4,6-trimethylphenyl)sulfonamido)-1-phenylpropyl (2S,3R,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (bis-*epi*-23)



TLC (SiO_2, hexane/EtOAc 1:1, UV): R_f = 0.42. **$[\alpha]_D^{20}$** = +8.6° (*c* 1.26, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.46 (br. s, 1H), 8.35 (br. s, 1H), 7.25 – 7.03 (m, 20H), 6.98 – 6.93 (m, 2H), 6.84 (s, 2H), 5.91 (d, *J* = 4.4 Hz, 1H), 4.65 (d, *J* = 16.3 Hz, 1H), 4.39 (d, *J* = 16.3 Hz, 1H), 4.26 – 4.12 (m, 2H), 3.84 (d, *J* = 14.2 Hz, 2H), 3.53 (d, *J* = 14.2 Hz, 2H), 3.08 – 2.88 (m, 3H), 2.76 (dd, *J* = 13.8, 4.1 Hz, 1H), 2.60 – 2.51 (m, 1H), 2.50 (s, 6H), 2.26 (s, 3H), 1.25 (d, *J* = 7.0 Hz, 3H), 0.96 (d, *J* = 7.2 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.6, 151.0, 147.2, 142.8, 140.3, 139.8, 138.0, 137.9, 137.1, 136.3, 133.3, 132.2, 128.6, 128.6, 128.5, 128.3, 128.2, 127.7, 127.4, 127.0, 126.4, 123.1, 78.6, 72.2, 60.4, 57.1, 54.5, 48.6, 44.5, 29.1, 23.1, 21.0, 14.6, 14.0. **IR** (thin film): υ (cm^{-1}) = 3026, 2978, 2939, 2927, 2366, 2337, 1737, 1604, 1496, 1454, 1380, 1320, 1216, 1152, 1028, 930, 928, 926, 856, 755, 698, 667. **HR-MS** (ESI): Calcd for $C_{50}H_{56}N_3O_5S$ $[M+H]^+$ 810.3935 *m/z*, found 810.3920 *m/z*.

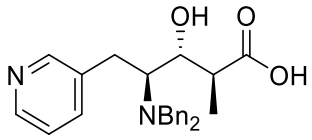
(2R,3S,4S)-4-(Dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoic acid (24)



To a solution of **23** (176 mg, 217 µmol, 1.00 eq.) in MeOH/THF/H_2O 3:2:2 (5.2 mL) was added $LiOH \cdot H_2O$ (45.6 mg, 1.09 mmol, 5.00 eq.) at rt. The clear solution was stirred at rt for 24 h and diluted with Et_2O (7mL). The aqueous layer was acidified to pH 2 (aq. 1M HCl) and the cleaved auxiliary was extracted, leaving the product in the aqueous layer. The latter was adjusted to pH 7 (sat. aq. $NaHCO_3$) and the product was extracted with $CHCl_3$ (4 x 15 mL). The organic layer was dried over $MgSO_4$, filtered, and concentrated *in vacuo* to deliver **24** (84.1 mg, 96%) as a viscous yellow resin. **TLC** (SiO_2, hexane/EtOAc 3:7, UV): R_f = 0.08. **$[\alpha]_D^{20}$** = +35.6° (*c* 0.54, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.48 (s, 1H), 8.40 (d, *J* = 3.3 Hz, 1H), 7.53 (d, *J* = 7.9 Hz,

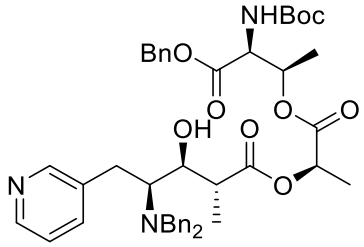
1H), 7.29 – 7.04 (m, 11H), 4.05 (d, *J* = 12.8 Hz, 2H), 3.46 (t, *J* = 5.6 Hz, 1H), 3.39 (d, *J* = 13.4 Hz, 2H), 3.04 – 2.89 (m, 3H), 2.80 – 2.64 (m, 1H), 0.84 (d, *J* = 7.1 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 177.7, 149.5, 146.3, 138.8, 137.8, 136.3, 129.2, 128.5, 127.4, 123.8, 73.0, 61.0, 55.1, 42.2, 28.6, 14.6. **IR** (thin film): υ (cm^{-1}) = 3411, 3062, 3027,2973, 2936, 2804, 1713, 1494, 1454, 1423, 1376, 1302, 1266, 1196, 1129, 1090, 1075, 1049, 1027, 1007, 983, 751, 700. **HR-MS** (ESI): Calcd for $C_{25}H_{29}N_2O_3$ $[M+H]^+$, 405.2173 *m/z*; Found, 405.2173 *m/z*.

(2S,3R,4S)-4-(Dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoic acid (bis-*epi*-24)



Bis-*epi*-**24** was prepared from bis-*epi*-**23** according to the procedure described for **24**. Bis-*epi*-**24** (91.9 mg, 83%) was obtained as a yellow foam. **TLC** (SiO_2, DCM/MeOH, 9:1, UV): R_f = 0.29. **[α]$_D^{20}$** = –8.7° (*c* 1.16, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.40 (d, *J* = 4.6 Hz, 1H), 8.30 (s, 1H), 7.64 (d, *J* = 6.7 Hz, 1H), 7.39 – 7.25 (m, 2H), 7.23 – 7.14 (m, 7H), 7.14 – 7.06 (m, 2H), 3.96 – 3.85 (m, 1H), 3.79 (d, *J* = 14.0 Hz, 2H), 3.68 (d, *J* = 14.0 Hz, 2H), 3.16 – 3.01 (m, 2H), 2.92 (dd, *J* = 14.1, 5.1 Hz, 1H), 2.84 – 2.75 (m, 1H), 1.25 (d, *J* = 7.3 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 180.0, 148.1, 144.3, 139.7, 139.5, 138.8, 128.8, 128.4, 127.1, 123.7, 73.7, 62.7, 54.7, 42.7, 30.0, 16.1. **IR** (thin film): υ (cm^{-1}) = 3422, 3060, 3026, 2972, 2933, 2880, 2834, 2803, 1704, 1600, 1581, 1493, 1453, 1424, 1362, 1261, 1213, 1119, 1073, 1045, 1027, 974, 931, 747, 699. **HR-MS** (ESI): Calcd. for $C_{25}H_{29}N_2O_3$ $[M+H]^+$ 405.2173 *m/z*; found 405.2169 *m/z*.

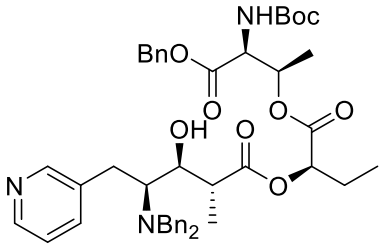
(R)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-oxopropan-2-yl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25a)



To a stirred solution of **24** (41.5 mg, 0.103 mmol, 1.00 eq.) in THF (0.60 mL) were added Et_3N (42.0 µL, 0.302 mmol, 2.93 eq.) and 2,4,6-trichloro benzoyl chloride (28.0 µL, 0.179 mmol, 1.74 eq.). After 5 min, a solution of **8a** (43.5 mg, 0.114 mmol, 1.11 eq.) in toluene (0.50 mL) and DMAP (18.3 mg, 0.149 mmol, 1.45 eq.) was added. The reaction was stirred at –78 °C for 1 h and then gradually allowed to warm to –35 °C (over 1 h) and stirred at –35 °C for 41.5 h. The mixture was then gradually allowed to warm to 0 °C (over 2.5 h) and stirred at 0 °C for 1 h. The reaction was quenched by addition of sat. aq. $NaHCO_3$ (2 mL) and the aqueous layer was extracted with EtOAc (3 x 10 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 2:3) provided **25a** (50.3 mg, 64%) as a pale yellow oil. **TLC** (SiO_2, hexane/EtOAc 2:3, UV) R_f = 0.43. **[α]$_D^{20}$** = +47.9° (*c* 0.43, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.57 (d, *J* = 1.9 Hz, 1H), 8.47 (dd, *J* = 4.9, 1.6 Hz, 1H), 7.63 (dt, *J* = 7.8, 1.7 Hz, 1H), 7.39 – 7.26 (m, 13H), 7.26 – 7.17 (m, 3H), 5.64 (d, *J* = 9.9 Hz, 1H), 5.52 (qd, *J* = 6.3, 2.5 Hz, 1H), 5.10 (d, *J* = 12.1 Hz, 1H), 5.08 (d, *J* = 12.0 Hz, 1H), 4.86 (q, *J* = 7.1 Hz, 1H), 4.51 (dd, *J* = 9.9, 2.4 Hz, 1H), 4.36 – 4.17 (m, 2H), 3.96 – 3.85 (m, 1H), 3.51 – 3.36 (m, 3H), 3.28 – 3.07 (m, 3H), 2.90 – 2.81 (m, 1H), 1.43 (s, 9H), 1.35 (d, *J* = 7.1 Hz, 3H), 1.24 (d, *J* = 6.4 Hz, 3H), 0.35 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.5, 170.3, 170.0, 156.0, 150.7, 147.5, 140.0, 137.1, 135.8, 134.9, 129.4, 128.6, 128.6, 128.3, 127.1, 123.5, 80.3, 73.9, 72.1, 68.7, 67.9,

59.4, 57.1, 55.9, 42.1, 28.3, 27.3, 16.8, 16.6, 13.0. **IR** (thin film): υ (cm^{-1}) = 3084, 3061, 3027, 2978, 2936, 2879, 2803, 1734, 1717, 1655, 1575, 1496, 1455, 1424, 1380, 1367, 1347, 1316, 1250, 1213, 1162, 1130, 1088, 1062, 1028, 1003, 988, 949, 913, 864, 841, 750, 745, 734, 699. **HR-MS** (ESI): Calcd. for $C_{44}H_{54}N_3O_9$ $[M+H]^+$ 768.3855 *m/z*; found 768.3844 *m/z*.

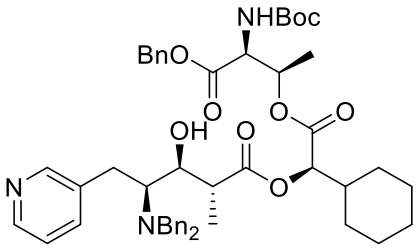
(R)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-oxo-butan-2-yl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25b)



25b was prepared from **24** and **8b** according to the procedure described for **25a**. FC (hexane/EtOAc 3:1 to 1:1) provided **25b** (164 mg, 71%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 9:1, UV): R_f = 0.50. **$[\alpha]_D^{20}$** = +56.0° (*c* 0.98, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.57 (d, *J* = 1.7 Hz, 1H), 8.47 (dd, *J* = 4.8, 1.5 Hz, 1H), 7.63 (dt, *J* = 7.8, 1.8 Hz, 1H), 7.38 – 7.27 (m, 13H), 7.27 – 7.19 (m, 3H), 5.67 (d, *J* = 9.9 Hz, 1H), 5.54 (qd, *J* = 6.0, 2.6 Hz, 1H), 5.15 – 5.03 (m, 2H), 4.76 (dd, *J* = 7.5, 4.6 Hz, 1H), 4.51 (dd, *J* = 10.0, 2.4 Hz, 1H), 4.37 – 4.19 (m, 2H), 3.91 (br. s, 1H), 3.50 – 3.40 (m, 3H), 3.29 – 3.11 (m, 3H), 2.88 – 2.79 (m, 1H), 1.86 – 1.64 (m, 3H), 1.43 (s, 9H), 1.23 (d, *J* = 6.4 Hz, 3H), 0.95 (t, *J* = 7.4 Hz, 3H), 0.32 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.9, 170.5, 169.6, 156.1, 150.9, 147.7, 140.2, 137.2, 135.9, 135.00, 129.6, 128.8, 128.7, 128.7, 128.5, 127.3, 123.7, 80.4, 74.0, 73.44, 72.2, 68.1, 59.5, 57.3, 56.1, 42.3, 28.4, 27.4, 24.6, 16.8, 13.1, 9.6. **IR** (thin film): υ (cm^{-1}) = 3452, 2978, 2931,

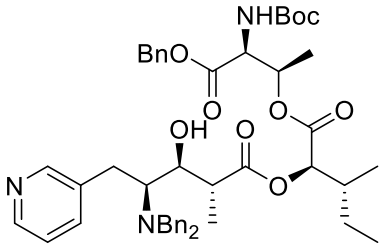
2802, 1739, 1715, 1496, 1454, 1367, 1248, 1215, 1161, 1128, 1087, 1062, 982, 752, 699. **HR-MS** (ESI): Calcd. for $C_{45}H_{56}N_3O_9$ $[M+H]^+$ 782.4011 *m/z*; found 782.3994 *m/z*.

(R)-2-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-cyclohexyl-2-oxoethyl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25c)



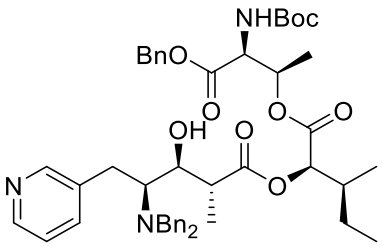
25c was prepared from **24** and **8c** according to the procedure described for **25a**. FC (hexane/EtOAc 1:1) provided **25c** (56.6 mg, 65%) as a pale yellow oil. **TLC** (SiO_2; hexane/EtOAc 3:2, UV): R_f = 0.23. $[\alpha]_D^{20}$ = +53.8° (*c* 1.04, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.49 (br. s, 1H), 8.44 – 8.37 (m, 1H), 7.63 – 7.56 (m, 1H), 7.34 – 7.21 (m, 12H), 7.21 – 7.13 (m, 4H), 5.66 (d, *J* = 10.0 Hz, 1H), 5.50 (qd, *J* = 6.4, 2.5 Hz, 1H), 5.06 (d, *J* = 12.2 Hz, 1H), 4.96 (d, *J* = 12.2 Hz, 1H), 4.61 (d, *J* = 3.9 Hz, 1H), 4.44 (dd, *J* = 10.0, 2.5 Hz, 1H), 4.29 – 4.12 (m, 2H), 3.85 (d, J = 3.9 Hz, 1H), 3.42 – 3.32 (m, 3H), 3.27 – 3.03 (m, 3H), 2.75 (dt, *J* = 10.0, 4.0 Hz, 1H), 1.88 – 1.75 (m, 1H), 1.66 – 1.41 (m, 5H), 1.35 (s, 9H), 1.25 – 0.97 (m, 8H), 0.22 (d, *J* = 6.7 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.9, 170.5, 169.1, 156.2, 150.5, 147.3, 140.2, 137.6, 136.1, 134.9, 129.6, 128.8, 128.7, 128.6, 128.5, 127.3, 123.8, 80.3, 76.7, 74.0, 72.3, 68.1, 59.5, 57.3, 56.1, 42.2, 39.8, 29.3, 28.4, 27.4, 27.4, 26.2, 26.1, 26.0, 16.9, 13.1. **IR** (thin film): υ (cm^{-1}) = 3451, 3086, 3064, 3029, 2978, 2932, 2856, 2802, 1739, 1718, 1496, 1453, 1423, 1367, 1345, 1315, 1251, 1217, 1163, 1134, 1103, 1085, 1062, 1028, 985, 944, 866, 842, 752, 731, 699 cm-1. **HR-MS** (ESI): Calcd. for $C_{49}H_{62}N_3O_9$ $[M+H]^+$ 836.4481 *m/z*; found, 836.4481 *m/z*.

(2R,3R)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-3-methyl-1-oxopentan-2-yl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25d)



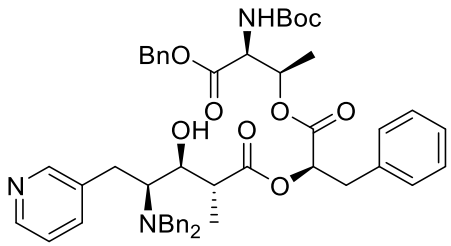
25d was prepared from **24** and **8d** according to the procedure described for **25a**. FC (hexane/EtOAc 4:1 to 1:1) provided **25d** (13.4 mg, 21%) as a yellow oil. **TLC** (SiO_2, MeOH/DCM 95:5, UV): R_f = 0.43. **$[\alpha]_D^{20}$** = +40.1° (*c* 0.43, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.57 (d, *J* = 1.8 Hz, 1H), 8.47 (dd, *J* = 4.8, 1.5 Hz, 1H), 7.63 (dt, *J* = 7.8, 1.8 Hz, 1H), 7.35 – 7.29 (m, 10H), 7.24 – 7.15 (m, 5H), 7.11 – 7.09 (m, 1H), 5.73 (d, *J* = 10.0 Hz, 1H), 5.58 (qd, *J* = 6.3, 2.3 Hz, 1H), 5.15 (d, *J* = 12.0 Hz, 1H), 5.09 (dd, *J* = 31.3, 12.1 Hz, 2H), 4.75 (d, *J* = 4.0 Hz, 1H), 4.51 (dd, *J* = 9.9, 2.4 Hz, 1H), 4.28 (dd, *J* = 17.3, 7.6 Hz, 2H), 3.93 (m, 1H), 3.45 (d, *J* = 13.2 Hz, 1H), 3.43 – 3.35 (m, 2H), 3.32 – 3.12 (m, 3H), 2.82 (dd, *J* = 5.6, 4.0 Hz, 1H), 2.01 – 1.91 (m, 1H), 1.42 (s, 9H), 1.35 – 1.28 (m, 2H), 1.23 (d, *J* = 6.4 Hz, 3H), 0.95 (d, *J* = 7.0 Hz, 3H), 0.92 (t, *J* = 7.5 Hz, 3H), 0.29 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 151.1, 150.9, 147.7, 137.2, 129.6, 128.8, 128.7, 128.5, 127.3, 123.7, 76.5, 74.1, 72.2, 68.1, 59.4, 57.4, 56.1, 42.3, 36.8, 28.4, 24.5, 16.9, 15.6, 13.2, 11.7. Three of the carbonyl carbons were not detectable. **IR** (thin film): υ (cm^{-1}) = 3464, 3019, 2969, 2933, 1742, 1495, 1455, 1381, 1367, 1253, 1217, 1164, 1131, 1062, 774, 752. **HR-MS** (ESI): Calcd. for $C_{47}H_{60}N_3O_9$ $[M+Na]^+$ 810.4324 *m/z*; found 810.4329 *m/z*.

(2R,3S)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-3-methyl-1-oxopentan-2-yl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25e)



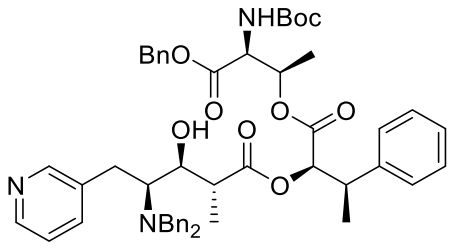
25e was prepared from **24** and **8e** according to the procedure described for **25a**. FC (hexane/EtOAc 4:1 to 1:1) provided **25e** (14.3 mg, 18%) as a yellow oil. **TLC** (SiO_2, DCM/MeOH 95:5, UV): R_f = 0.29. **[α]$_D^{20}$** = +51.1° (*c* 0.73, $CHCl_3$). **^{1}H-NMR** (500 MHz, $(CD_3)_2SO$): δ (ppm) = 8.54 (d, *J* = 1.7 Hz, 1H), 8.40 (dd, *J* = 4.8, 1.4 Hz, 1H), 7.69 (dt, *J* = 7.9, 2.0 Hz, 1H), 7.37 – 7.30 (m, 14H), 7.25 – 7.21 (m, 2H), 5.26 – 5.20 (m, 1H), 5.12 – 5.02 (m, 3H), 4.76 (d, *J* = 3.2 Hz, 1H), 4.39 (dd, *J* = 9.2, 3.7 Hz, 1H), 4.14 (s, 1H), 3.47 (d, *J* = 13.3 Hz, 2H), 3.31 (d, *J* = 9.7 Hz, 1H), 3.14 (dd, *J* = 13.0, 2.9 Hz, 1H), 2.99 (d, *J* = 26.9 Hz, 2H), 2.56 (dt, *J* = 10.5, 3.1 Hz, 1H), 1.83 (qd, *J* = 7.0, 3.2 Hz, 1H), 1.39 (s, 9H), 1.27 – 1.23 (m, 2H), 1.14 (d, *J* = 6.3 Hz, 3H), 0.79 (t, *J* = 7.4 Hz, 3H), 0.68 (d, *J* = 6.9 Hz, 3H), 0.08 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (126 MHz, $(CD_3)_2SO$): δ (ppm) = 174.7, 170.1, 168.9, 156.3, 151.0, 147.6, 140.7, 137.1, 136.5, 135.9, 129.6, 128.8, 128.6, 128.2, 127.3, 123.8, 79.2, 74.0, 73.0, 71.1, 66.9, 59.3, 57.4, 55.6, 43.4, 36.0, 28.6, 26.9, 25.9, 16.8, 14.3, 13.3, 11.9. **IR** (thin film): υ (cm^{-1}) = 3521, 3037, 2842, 2763, 1796, 1423, 1382, 1397, 1225, 1216, 1159, 1132, 1073, 776. **HR-MS** (ESI): Calcd. for $C_{47}H_{60}N_3O_9$ $[M+H]^+$ 810.4324 *m/z*; found 810.4321 *m/z*.

(R)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-oxo-3-phenylpropan-2-yl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25f)



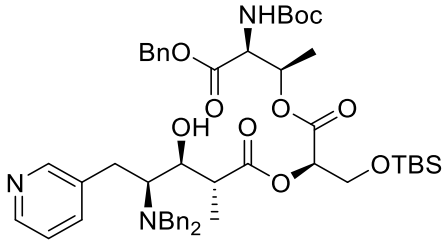
25f was prepared from **24** and **8f** according to the procedure described for **25a**. FC (hexane/EtOAc 4:1 to 0:1) provided **25f** (55.0 mg, 66%) as a pale yellow oil. **TLC** (SiO_2, hexane/EtOAc 4:1, UV): R_f = 0.40. **$[\alpha]_D^{20}$** = +48.8° (*c* 1.14, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.55 (d, *J* = 1.7 Hz, 1H), 8.47 (dd, *J* = 4.7, 1.2 Hz 1H), 7.59 (d, *J* = 7.8 Hz, 1H), 7.37 – 7.28 (m, 13H), 7.27 – 7.24 (m, 3H), 7.23 – 7.16 (m, 4H), 5.60 (d, *J* = 9.9 Hz, 1H), 5.52 (qd, *J* = 6.0, 2.6 Hz, 1H), 5.10 (s, 2H), 5.03 (dd, *J* = 9.0, 3.8 Hz, 1H), 4.49 (dd, *J* = 9.9, 2.3 Hz, 1H), 4.34 – 4.19 (m, 2H), 3.64 (s, 1H), 3.45 (d, *J* = 13.3 Hz, 2H), 3.41 – 3.34 (m, 1H), 3.25 – 3.11 (m, 3H), 3.07 (dd, *J* = 14.4, 3.7 Hz, 1H), 2.91 (dd, *J* = 14.3, 9.1 Hz, 1H), 2.82 – 2.75 (m, 1H), 1.41 (s, 9H), 1.12 (d, *J* = 6.4 Hz, 3H), 0.17 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.2, 170.5, 168.8, 156.1, 150.8, 147.7, 140.2, 137.1, 135.9, 135.9, 135.0, 129.5, 129.5, 128.7, 128.7, 128.7, 128.6, 128.5, 127.3, 127.2, 123.6, 80.3, 73.8, 72.8, 72.4, 68.1, 59.2, 57.3, 56.1, 42.5, 37.2, 28.4, 27.2, 16.6, 13.0. **IR** (thin film): υ (cm^{-1}) = 3422, 3030, 2979, 2931, 2806, 1740, 1715, 1496, 1454, 1366, 1312, 1274, 1247, 1160, 1082, 1028, 865, 751, 698. **HR-MS** (ESI): Calcd. for $C_{50}H_{58}N_3O_9$ $[M+H]^+$ 844.4168 *m/z*; found 844.4153 *m/z*.

(2R,3R)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-oxo-3-phenylbutan-2-yl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25g)



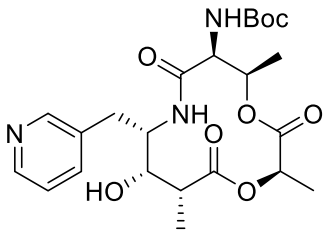
25g was prepared from **24** and **8g** according to the procedure described for **25a**. FC (hexane/EtOAc 1:1) provided **25g** (38.2 mg, 41%) as a beige foam. **TLC** (SiO_2, hexane/EtOAc 1:1, UV, CPS): R_f = 0.31. **[α]$_D^{20}$** = +50.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.59 – 8.41 (m, 2H), 7.68 – 7.56 (m, 1H), 7.44 – 7.28 (m, 15H), 7.25 – 7.18 (m, 6H), 5.61 – 5.52 (m, 1H), 5.41 (m, 1H), 5.13 – 4.98 (m, 2H), 4.98 – 4.88 (m, 1H), 4.40 (d, *J* = 9.9 Hz, 1H), 4.26 (m, 2H), 3.48 – 3.14 (m, 7H), 2.81 (s, 1H), 1.37 (s, 9H), 1.33 (d, *J* = 5.1 Hz, 3H), 0.83 (d, *J* = 4.4 Hz, 3H), 0.26 (d, *J* = 6.5 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.7, 170.6, 168.4, 156.1, 150.8, 147.7, 140.7, 140.1, 137.2, 135.9, 135.0, 129.5, 128.7, 128.7, 128.5, 128.4, 127.3, 127.3, 127.2, 123.7, 80.2, 76.4, 73.7, 72.0, 68.1, 59.3, 57.2, 56.1, 42.6, 41.4, 28.5, 28.4, 27.3, 18.2, 16.3, 13.3. **IR** (thin film): υ (cm^{-1}) = 3029, 2977, 2934, 1739, 1716, 1496, 1454, 1380, 1366, 1347, 1314, 1266, 1218, 1163, 1112, 1088, 1061, 1027, 984, 945, 752, 699. **HR-MS** (ESI): Calcd. for $C_{51}H_{60}N_3O_9$ $[M+H]^+$ 858.4324 *m/z*; found 858.4313 *m/z*.

(6R,9R,10S)-10-((Benzyloxy)carbonyl)-2,2,3,3,9,14,14-heptamethyl-7,12-dioxo-4,8,13-trioxa-11-aza-3-silapentadecan-6-yl (2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (25i)



25i was prepared from **24** and **8i** according to the procedure described for **25a**. FC (hexane/EtOAc 3:2) provided **25i** (38.1 mg, 72%) as a yellow oil. **TLC** (SiO_2, hexane/EtOAc 3:2, UV): R_f = 0.26. **[α]$_D^{20}$** = +47.3° (*c* 1.48, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.53 (d, *J* = 1.8 Hz, 1H), 8.44 (dd, *J* = 4.8, 1.5 Hz, 1H), 7.62 (d, *J* = 7.8 Hz, 1H), 7.39 – 7.24 (m, 10H), 7.24 – 7.12 (m, 6H), 5.70 (d, *J* = 10.0 Hz, 1H), 5.52 (qd, *J* = 6.4, 2.6 Hz, 1H), 5.06 (s, 2H), 4.90 (dd, *J* = 5.4, 2.9 Hz, 1H), 4.48 (dd, *J* = 10.0, 2.4 Hz, 1H), 4.36 – 4.18 (m, 2H), 3.89 (qd, *J* = 11.3, 4.2 Hz, 3H), 3.43 (d, *J* = 13.3 Hz, 3H), 3.30 – 3.11 (m, 3H), 2.79 (dd, *J* = 9.9, 7.3 Hz, 1H), 1.37 (s, 9H), 1.19 (d, *J* = 6.4 Hz, 3H), 0.84 (s, 9H), 0.26 (d, *J* = 6.8 Hz, 3H), 0.04 (d, *J* = 7.0 Hz, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.7, 170.5, 167.3, 156.1, 150.4, 147.2, 140.0, 137.4, 136.0, 134.9, 129.4, 128.6, 128.5, 128.5, 128.3, 127.1, 123.6, 80.1, 73.7, 73.7, 72.4, 68.0, 62.8, 59.1, 57.2, 56.0, 42.6, 28.3, 26.9, 25.7, 16.7, 13.0, -5.3, -5.4. **IR** (thin film): υ (cm^{-1}) = 2954, 2931, 2857, 1742, 1718, 1497, 1455, 1366, 1303, 1253, 1216, 1162, 1133, 1088, 1062, 1027, 1006, 948, 837, 780, 752, 700. **HR-MS** (ESI): Calcd. for $C_{50}H_{68}N_3O_{10}Si$ $[M+H]^+$ 898.4668 *m/z*; found 898.4672 *m/z*.

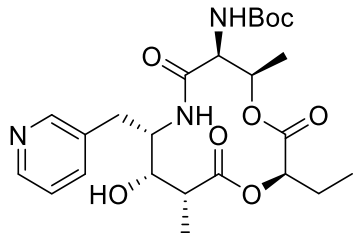
tert-Butyl ((2R,5R,6S,9S,10S,11R)-10-hydroxy-2,5,11-trimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate **(26a)**



To a solution of **25a** (60.4 mg, 0.079 mmol, 1.00 eq.) in degassed MeOH (1.8 mL) was added Pd/C (10%, 32.6 mg, 0.031 mmol, 0.39 eq.) and the argon atmosphere was exchanged with a H_2 atmosphere (1 bar). The reaction was stirred at rt for 3 h 15 min and was filtered over celite and concentrated *in vacuo*. The ensuing amino acid (45.5 mg, crude) was used in the following step without further purification. To a stirred solution of HATU (103 mg, 0.271 mmol, 1.76 eq.) and DIPEA (70.0 µL, 0.405 mmol, 2.62 eq.) in DCM/DMF (100:1, 70 mL) was added the above amino acid (76.7 mg, 0.154 mmol, 1.00 eq) in DCM/DMF (100:1, 47 mL) dropwise over 4 h (syringe pump). The reaction mixture was stirred at rt for 13 h and the reaction was then quenched by addition of sat. aq. $NaHCO_3$ (80 mL). The aqueous layer was extracted with DCM (4 x 100 mL) and the combined organic layers were washed with H_2O (4 x 100 mL), dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 4:1 to 0:1) provided **26a** (21.9 mg, 34% over 2 steps, containing tetramethyl urea impurities) as a white foam. **TLC** (SiO_2, EtOAc, UV) R_f = 0.19. **[α]$_D^{20}$** = –22.6° (*c* 0.43, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$) δ (ppm) = 8.53 (s, 1H), 8.47 (d, *J* = 4.5 Hz, 1H), 7.63 (d, *J* = 7.8 Hz, 1H), 7.22 (dd, *J* = 7.6, 4.8 Hz, 1H), 6.33 (br. s, 1H), 5.32 – 5.22 (m, 2H), 5.15 (q, *J* = 7.0 Hz, 1H), 4.44 (br. s, 1H), 4.32 – 4.22 (m, 1H), 4.04 – 3.94 (m, 1H), 3.54 (s, 1H), 3.06 – 2.90 (m, 2H), 2.45 (qd, *J* = 7.5, 1.0 Hz, 1H), 1.50 (d, *J* = 7.1 Hz, 3H), 1.44 (s, 9H), 1.37 (d, *J* = 7.4 Hz, 3H), 1.29 (d, *J* = 6.5 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$) δ (ppm) = 178.5, 168.4, 154.8, 150.9, 148.2, 137.3, 133.6, 123.6, 80.6, 72.9, 69.6, 57.1, 55.3, 40.9, 38.7, 35.7, 28.4,

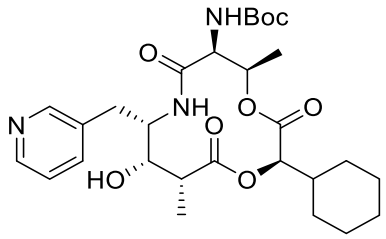
17.7, 16.5, 14.0. **IR** (thin film): υ (cm^{-1}) = 3483, 3430, 2246, 3202, 2979, 2939, 2875, 1745, 1716, 1671, 1597, 1578, 1498, 1453, 1427, 1381, 1368, 1320, 1250, 1166, 1065, 1048, 1027, 967, 913, 860, 753, 715. **HR-MS** (ESI): Calcd. for $C_{23}H_{34}N_3O_8$ $[M+H]^+$ 480.2340 *m/z*; found 480.2343 *m/z*.

tert-Butyl ((2R,5R,6S,9S,10S,11R)-2-ethyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26b)



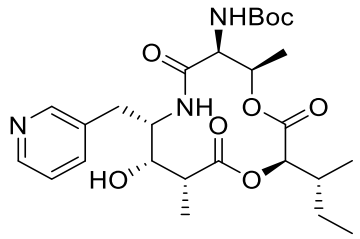
26b was prepared from **25b** according to the procedure described for **26a**. FC (hexane/EtOAc 4:1 to 1:0) provided **26b** (6.60 mg, 23% over 2 steps, containing some tetramethyl urea impurities) as a pale yellow film. **TLC** (SiO_2, hexane/EtOAc 3:2, UV): R_f = 0.33. **[α]D^{20}** = –27.4° (*c* 0.37, MeOH). **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 8.48 – 8.43 (m, 1H), 8.37 (d, *J* = 3.9 Hz, 1H), 7.73 (d, *J* = 7.8 Hz, 1H), 7.33 (dd, *J* = 7.6, 5.0 Hz, 1H), 5.21 – 5.12 (m, 1H), 4.86 – 4.78 (m, 1H), 4.28 – 4.20 (m, 1H), 4.14 – 4.03 (m, 1H), 3.63 (br. s, 1H), 3.07 – 2.91 (m, 2H), 2.56 (qd, *J* = 7.4, 1.0 Hz, 1H), 1.93 – 1.78 (m, 2H), 1.45 (s, 9H), 1.37 (d, *J* = 7.3 Hz, 3H), 1.29 (d, *J* = 6.5 Hz, 3H), 0.98 (t, *J* = 7.4 Hz, 3H). **^{13}C-NMR** (101 MHz, CD_3OD): δ (ppm) = 178.3, 170.9, 169.8, 157.1, 151.0, 148.0, 139.2, 136.3, 125.0, 81.0, 75.7, 75.6, 71.0, 57.7, 56.8, 42.4, 36.4, 28.7, 25.1, 17.9, 14.9, 9.6. **IR** (thin film): υ (cm^{-1}) = 3356, 2974, 2935, 2878, 1744, 1716, 1669, 1496, 1457, 1383, 1367, 1245, 1167, 1091, 1046, 1028, 960, 865, 711. **HR-MS** (ESI): Calcd. for $C_{24}H_{36}N_3O_8$ $[M+H]^+$ 494.2497 *m/z*; found 494.2497 *m/z*.

tert-Butyl ((2R,5R,6S,9S,10S,11R)-2-cyclohexyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26c)



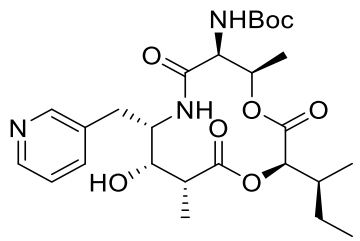
26c was prepared from **25c** according to the procedure described for **26a**. FC (hexane/EtOAc 1:9 to 0:1) provided **26c** (27.4 mg, 42% over 2 steps) as a yellow oil. **TLC** (SiO_2; EtOAc, UV): R_f = 0.35. **[α]$_D^{20}$** = –14.28° (*c* 0.98, $CHCl_3$). **^{1}H-NMR** (500 MHz, CD_3OD): δ (ppm) = 8.46 (s, 1H), 8.41 – 8.33 (m, 1H), 7.73 (d, *J* = 7.7 Hz, 1H), 7.33 (dd, *J* = 7.9, 4.9 Hz, 1H), 5.25 – 5.13 (m, 1H), 4.68 (d, *J* = 4.9 Hz, 1H), 4.27 (d, *J* = 5.8 Hz, 1H), 4.10 (t, *J* = 6.8 Hz, 1H), 3.66 – 3.61 (m, 1H), 2.99 (ddd, *J* = 30.6, 13.5, 7.5 Hz, 2H), 2.62 – 2.52 (m, 1H), 1.94 – 1.85 (m, 1H), 1.80 – 1.72 (m, 2H), 1.70 – 1.61 (m, 3H), 1.45 (s, 9H), 1.37 (d, *J* = 7.3 Hz, 3H), 1.30 (d, *J* = 6.5 Hz, 3H), 1.27 – 1.09 (m, 5H). **^{13}C-NMR** (126 MHz, CD_3OD): δ (ppm) = 178.4, 170.8, 169.2, 157.0, 151.0, 148.0, 139.2, 136.2, 125.0, 81.0, 78.7, 75.6, 71.0, 57.6, 56.8, 42.3, 40.3, 36.4, 29.8, 29.0, 28.7, 27.0, 26.9, 26.8, 18.0, 15.1. **IR** (thin film): υ (cm^{-1}) = 2975, 2930, 2855, 1744, 1715, 1698, 1671, 1451, 1405, 1393, 1367, 1261, 1168, 1050, 1026, 994, 857, 769 cm^{-1}. **HR-MS** (ESI): Calcd. for $C_{28}H_{42}N_3O_8$ $[M+H]^+$ 548.2966 *m/z*; found, 548.2957 *m/z*.

tert-Butyl ((2R,5R,6S,9S,10S,11R)-2-((R)-sec-butyl)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26d)



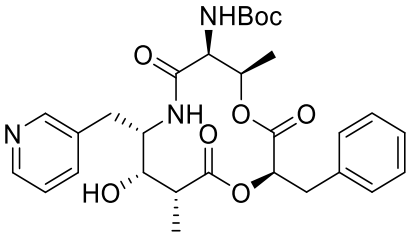
26d was prepared from **25d** according to the procedure described for **26a**. FC (hexane/EtOAc 5:95 to 0:1) provided **26d** (6.8 mg, 62% over 2 steps) as a colorless film. **TLC** (SiO_2, hexane/EtOAc 1:9, UV): R_f = 0.39. **$[\alpha]_D^{20}$** = +19.7° (*c* 0.10, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.55 (d, *J* = 1.7 Hz, 1H), 8.48 (dd, *J* = 4.8, 1.4 Hz, 1H), 7.63 (d, *J* = 7.7 Hz, 1H), 7.23 (dd, *J* = 7.6, 5.1 Hz, 1H), 6.10 (d, *J* = 8.9 Hz, 1H), 5.36 – 5.32 (m, 1H), 5.21 (d, *J* = 8.9 Hz, 1H), 4.94 (d, *J* = 4.2 Hz, 1H), 4.27 – 4.17 (m, 1H), 4.05 – 3.91 (m, 1H), 3.56 – 3.41 (m, 1H), 3.08 – 2.90 (m, 2H), 2.55 – 2.43 (m, 1H), 2.16 – 2.03 (m, 1H), 1.45 (s, 9H), 1.38 (d, *J* = 7.4 Hz, 3H), 1.39 – 1.30 (m, 2H), 1.29 (d, *J* = 6.6 Hz, 3H), 0.96 (d, *J* = 6.9 Hz, 3H), 0.89 (t, *J* = 7.2 Hz, 3H). **IR** (thin film): υ (cm^{-1}) = 2925, 2856, 2320, 2118, 2103, 1735, 1719, 1673, 1502, 1453, 1369, 1263, 803, 776, 692, 545. **HR-MS** (ESI): *m/z* Calcd. for $C_{26}H_{40}N_3O_8$ $[M+H]^+$ 522.2810 *m/z*; found 522.2807 *m/z*.

tert-Butyl ((2R,5R,6S,9S,10S,11R)-2-((S)-sec-butyl)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26e)



26e was prepared from **25e** according to the procedure described for **26a**. FC (hexane/EtOAc 1:9 to 0:1) provided **26e** (16.4 mg, 49% over 2 steps) as a colorless film. **TLC** (SiO_2, EtOAc, UV): R_f = 0.46. **$[\alpha]_D^{20}$** = +27.2° (*c* 0.47, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.55 (d, *J* = 1.8 Hz, 1H), 8.48 (dd, *J* = 4.8, 1.6 Hz, 1H), 7.63 (d, *J* = 7.8 Hz, 1H), 7.23 (dd, *J* = 7.7, 4.8 Hz, 1H), 6.12 (d, *J* = 8.5 Hz, 1H), 5.40 – 5.30 (m, 1H), 5.22 (d, *J* = 8.2 Hz, 1H), 5.07 (d, *J* = 3.2 Hz, 1H), 4.26 – 4.18 (m, 1H), 4.05 – 3.94 (m, 1H), 3.61 – 3.40 (m, 1H), 3.05 – 2.91 (m, 2H), 2.50 (q, *J* = 8.1, 7.4 Hz, 1H), 2.20 – 2.05 (m, 1H), 1.45 (s, 9H), 1.38 (d, *J* = 7.5 Hz, 3H), 1.37 – 1.25 (m, 2H), 1.29 (d, *J* = 6.5 Hz, 3H), 0.94 – 0.83 (m, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 150.9, 148.2, 137.3, 123.6, 75.5, 72.8, 69.2, 57.1, 55.4, 40.9, 36.5, 28.4, 27.3, 26.0, 17.8, 14.2, 14.2, 11.6. **HR-MS** (ESI): Calcd. for $C_{26}H_{40}N_3O_8$ $[M+H]^+$ 522.2810 *m/z*; found 522.2806 *m/z*.

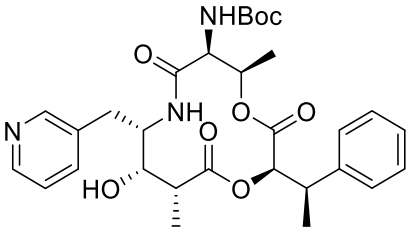
tert-Butyl ((2R,5R,6S,9S,10S,11R)-2-benzyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26f)



26f was prepared from **25f** according to the procedure described for **26a**. FC (hexane/EtOAc 8:2 to 1:0) provided **26f** (8.30 mg, 26% over 2 steps, containing tetramethyl urea impurities) as a yellow film. **TLC** (SiO_2, EtOAc, UV): R_f = 0.30. **$[\alpha]_D^{20}$** = –23.2° (*c* 0.35, MeOH). **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 8.46 – 8.42 (m, 1H), 8.36 (d, *J* = 3.8 Hz, 1H), 7.71 (d, *J* = 7.8 Hz, 1H), 7.34 – 7.19 (m, 6H), 5.19 – 5.07 (m, 2H), 4.22 (d, *J* = 5.6 Hz, 1H), 4.08 – 4.01 (m, 1H), 3.59 (br. s, 1H), 3.20 (dd, *J* = 14.3, 4.4 Hz, 1H), 3.09 (dd, *J* = 14.3, 8.5 Hz, 1H), 3.03 – 2.89 (m, 2H), 2.51 (qd, *J* = 7.4, 1.3 Hz, 1H), 1.45 (s, 9H), 1.21 (d, *J* = 7.3 Hz, 3H), 1.15 (d, *J* = 6.5 Hz, 3H). **^{13}C-**

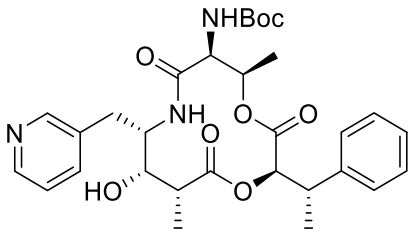
NMR (101 MHz, CD_3OD): δ (ppm) = 178.0, 170.9, 169.1, 157.1, 151.0, 148.0, 139.2, 137.1, 136.3, 130.6, 129.4, 128.1, 125.0, 81.0, 75.5, 75.0, 71.1, 57.6, 56.8, 42.4, 37.5, 36.4, 28.7, 17.7, 14.6. **IR** (thin film): υ (cm^{-1}) = 3334, 2974, 2934, 1747, 1717, 1669, 1507, 1456, 1386, 1367, 1247, 1163, 1063, 1023, 700. **HR-MS** (ESI): Calcd. for $C_{29}H_{38}N_3O_8$ $[M+H]^+$ 556.2653 *m/z*; found 556.2652 *m/z*.

tert-Butyl ((2R,5R,6S,9S,10S,11R)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-2-((R)-1-phenylethyl)-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26g)



26g was prepared from **25g** according to the procedure described for **26a**. FC (EtOAc) provided **26g** (24.2 mg, 52% over 2 steps) as a slightly yellow oil. **TLC** (SiO_2, hexane/EtOAc 1:9, UV, $KMnO_4$): R_f = 0.23. **[α]$_D^{20}$** = –4.0° (*c* 0.50, MeOH). **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 8.48 – 8.40 (m, 1H), 8.35 (dd, *J* = 5.0, 1.6 Hz, 1H), 7.70 (d, *J* = 7.9 Hz, 1H), 7.36 – 7.15 (m, 6H), 5.03 (d, *J* = 6.3 Hz, 2H), 4.15 (d, *J* = 5.7 Hz, 1H), 4.02 (t, *J* = 7.5 Hz, 1H), 3.61 (s, 1H), 3.35 (qd, *J* = 7.3, 6.3 Hz, 1H), 2.98 (m, 2H), 2.53 (qd, *J* = 7.4, 1.2 Hz, 1H), 1.44 (s, 9H), 1.36 (d, *J* = 7.2 Hz, 3H), 1.27 (d, *J* = 7.5 Hz, 3H), 0.90 (d, *J* = 6.5 Hz, 3H). **^{13}C-NMR** (101 MHz, CD_3OD): δ (ppm) = 178.2, 170.9, 168.8, 151.0, 147.9, 142.4, 139.3, 136.3, 129.3, 128.2, 125.0, 78.5, 75.7, 70.9, 57.7, 56.8, 42.4, 42.3, 42.0, 36.1, 28.6, 18.3, 17.8, 14.4. **IR** (thin film): υ (cm^{-1}) = 3397, 2977, 2930, 1744, 1415, 1673, 1496, 1455, 1388, 1369, 1268, 1165, 1064, 845, 702, 559. **HR-MS** (ESI): Calcd. for $C_{30}H_{40}N_3O_8$ $[M+H]^+$ 570.2810 *m/z*; found 570.2809 *m/z*.

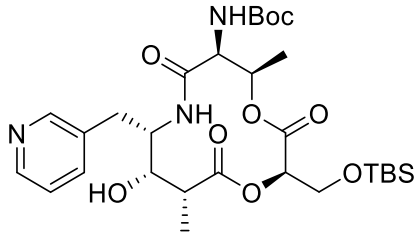
tert-Butyl ((2R,5R,6S,9S,10S,11R)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-2-((S)-1-phenylethyl)-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26h)



To a stirred solution of **31** (33.4 mg, 47.5 µmol, 1.00 eq.) in MeOH (3.0 mL) was added Pd/C (10%, 10.1 mg, 9.5 µmol) at rt. The reaction mixture was put under a hydrogen atmosphere (balloon) and stirred for 1 h. To the mixture was added more Pd/C (10%, 10.1 mg, 9.5 µmol) and stirring under hydrogen (balloon) was continued for 1 h. The reaction was filtered through a pad of celite and concentrated *in vacuo*. The ensuing amino acid (27.9 mg, quant., crude) was obtained as a slight yellow oil and was used in the following step without further purification. To a stirred solution of HATU (30.7 mg, 80.8 µmol, 1.70 eq.) and DIPEA (21.5 µL, 0.12 mmol, 2.60 eq.) in DMF (0.18 mL) and DCM (18 mL) was added at rt a solution of the above amino acid (27.9 mg, 47.5 µmol, 1.00 eq.) in DMF (0.18 mL) and DCM (18 mL) over 4 h. The mixture was stirred at rt for 48 h. The reaction was then quenched with sat. aq. $NaHCO_3$ solution (5 mL) and the mixture was extracted with DCM (3 x 20 mL). The combined organic layers were dried over $MgSO_4$, filtered and concentrated *in vacuo*. FC (EtOAc) provided **26h** (8.3 mg, 31% over 2 steps, containing tetramethyl urea impurities) as a slightly yellow oil. **TLC** (SiO_2, hexane/EtOAc 9:1, UV, $KMnO_4$): R_f = 0.14. **$[\alpha]_D^{20}$** = –20.0° (*c* 0.50, MeOH). **^{1}H-NMR** (500 MHz, CD_3OD): δ (ppm) = 8.47 – 8.42 (m, 1H), 8.36 (dd, *J* = 5.0, 1.6 Hz, 1H), 7.72 (dt, *J* = 8.0, 1.9 Hz, 1H), 7.34 – 7.17 (m, 6H), 5.12 (t, *J* = 6.4 Hz, 1H), 4.99 (d, *J* = 5.0 Hz, 1H), 4.24 (d, *J* = 6.0 Hz, 1H), 4.13 – 4.05 (m, 1H), 3.62 – 3.58 (m, 1H), 3.41 (qd, *J* = 7.1, 4.9 Hz, 1H), 3.04 – 2.89 (m, 2H), 2.58 (qd, *J* = 7.3, 1.4 Hz, 1H), 1.45 (s, 9H), 1.35 (d, *J* = 7.1 Hz, 3H), 1.23 (dd, *J* = 6.9, 4.2 Hz, 6H). **^{13}C-NMR**

(126 MHz, CD_3OD): δ (ppm) = 178.0, 170.8, 168.6, 157.0, 151.0, 148.0, 142.4, 139.2, 136.2, 129.5, 129.3, 128.9, 128.3, 128.2, 125.0, 81.0, 78.6, 75.5, 71.1, 57.6, 56.7, 42.3, 41.9, 36.4, 33.0, 28.7, 17.7, 15.8, 14.8. **IR** (thin film): υ (cm^{-1}) = 3334, 2978, 2928, 2854, 1743, 1717, 1669, 1496, 1455, 1425, 1387, 1367, 1249, 1166, 1046, 1025, 716, 700, 445, 432. **HR-MS** (ESI): Calcd. for $C_{30}H_{40}N_3O_8$ $[M+H]^+$ 570.2810 *m/z*; found 570.2820 *m/z*.

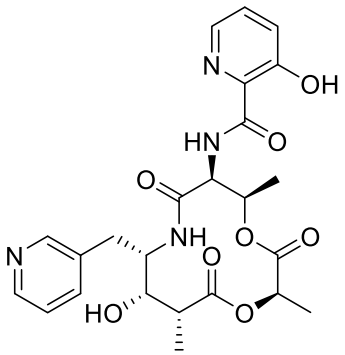
tert-Butyl ((2R,5R,6S,9S,10S,11R)-2-(((tert-butyldimethylsilyl)oxy)methyl)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (26i)



26i was prepared from **25i** according to the procedure described for **26a**. FC (hexane/EtOAc 1:9) provided **26i** (11.4 mg, 57% over 2 steps) as an orange oil. **TLC** (SiO_2, hexane/EtOAc 1:9, UV): R_f = 0.22. **$[\alpha]_D^{20}$** = −9.3° (*c* 0.76, MeOH). **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 8.46 (d, *J* = 1.6 Hz, 1H), 8.37 (dd, *J* = 4.9, 1.5 Hz, 1H), 7.74 (dt, *J* = 7.8, 1.7 Hz, 1H), 7.33 (dd, *J* = 7.6, 4.9 Hz, 1H), 5.25 – 5.11 (m, 1H), 5.03 (t, *J* = 3.5 Hz, 1H), 4.26 (d, *J* = 6.2 Hz, 1H), 4.17 – 4.04 (m, 2H), 3.96 (dd, *J* = 11.5, 3.0 Hz, 1H), 3.64 (s, 1H), 2.98 (qd, *J* = 13.6, 7.4 Hz, 2H), 2.57 (qd, *J* = 7.2, 1.1 Hz, 1H), 1.46 (s, 10H), 1.38 (d, *J* = 7.4 Hz, 3H), 1.27 (d, *J* = 6.5 Hz, 3H), 0.88 (s, 9H), 0.08 (s, 3H), 0.07 (s, 3H). **^{13}C-NMR** (101 MHz, CD_3OD): δ (ppm) = 178.0, 168.1, 151.1, 148.0, 139.3, 136.3, 125.0, 75.7, 75.4, 71.0, 63.6, 57.5, 56.5, 42.6, 36.5, 28.7, 26.3, 19.2, 17.8, 15.0, -5.2, -5.3. **IR** (thin film): υ (cm^{-1}) = 3377, 2932, 2856, 1746, 1718, 1675, 1496, 1387, 1367, 1253, 1166,

1046, 1026, 838, 779, 559, 526. **HR-MS** (ESI): Calcd. for $C_{29}H_{48}N_3O_9Si$ $[M+H]^+$ 610.3160 *m/z*; found 610.3158 *m/z*.

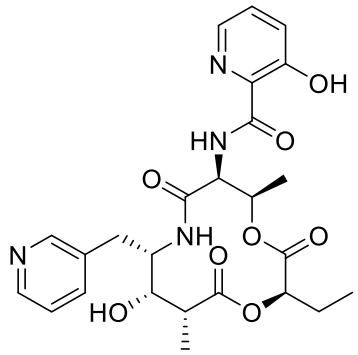
3-Hydroxy-N-((2R,5R,6S,9S,10S,11R)-10-hydroxy-2,5,11-trimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)picolinamide (28a)



To a stirred solution of **26a** (13.1 mg, 0.027 mmol, 1.00 eq.) in DCM (1.6 mL) at 0 °C was added TFA (0.21 mL, 2.73 mmol, 100 eq.). The solution was allowed to warm to rt and stirred for 3.5 h. The solvent was then evaporated *in vacuo* to deliver the free amine (as the TFA salt) (18.4 mg, crude), which was used in the following step without further purification. To a stirred solution of HATU (15.2 mg, 0.040 mmol, 2.50 eq.) and DIPEA (16.0 µL, 0.090 mmol, 5.50 eq.) in MeCN (0.25 mL) were added 3-hydroxypicolinic acid **27** (4.60 mg, 0.033 mmol, 2.00 eq.) and a solution of the above TFA salt (8.10 mg, 0.016 mmol, 1.00 eq.) in MeCN (1.2 mL) at rt. The solution was stirred at rt for 20 h and then diluted with DCM (2 mL) and $NaHCO_3$ (1 mL). The aqueous layer was extracted with DCM (4 x 10 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (EtOAc/MeOH 9:1) provided **28a** as a green oil. Further purification by RP-HPLC (10 to 30% MeCN/0.1% TFA in 0.1% aq. TFA) gave **28a** (3.28 mg, 55% over 2 steps) as a colorless film. **TLC** (SiO_2, EtOAc/MeOH 9:1, UV) R_f = 0.32. **$[\alpha]_D^{20}$** = –43.5° (*c* 0.23, MeOH). **^{1}H-NMR** (500 MHz, CD_3OD): δ (ppm) = 8.76 (s, 1H), 8.53 (d, *J* = 4.7 Hz,

1H), 8.41 (d, *J* = 7.1 Hz, 1H), 8.22 – 8.14 (m, 2H), 7.72 (t, *J* = 5.9 Hz, 1H), 7.52 (dd, *J* = 8.5, 4.4 Hz, 1H), 7.42 (d, *J* = 8.5 Hz, 1H), 5.31 (p, *J* = 6.4 Hz, 1H), 5.04 (q, *J* = 6.9 Hz, 1H), 4.69 (d, *J* = 6.1 Hz, 1H), 4.33 – 4.22 (m, 1H), 3.81 (s, 1H), 3.22 (d, *J* = 6.5 Hz, 2H), 2.70 – 2.62 (m, 1H), 1.49 (d, *J* = 7.1 Hz, 3H), 1.44 (d, *J* = 7.3 Hz, 3H), 1.29 (d, *J* = 6.4 Hz, 3H). **^{13}C-NMR** (126 MHz, CD_3OD): δ (ppm) = 178.1, 170.4, 170.2, 169.4, 159.2, 148.1, 144.2, 141.2, 140.5, 131.6, 130.8, 127.7, 127.52, 76.3, 71.1, 70.1, 57.0, 54.8, 42.4, 36.7, 17.8, 16.6, 14.8. **IR** (thin film): υ (cm^{-1}) = 3399, 2920, 2852, 1738, 1653, 1522, 1451, 1385, 1268, 1182, 1141, 1063, 1035, 841, 558. **HR-MS** (ESI): Calcd. for $C_{24}H_{29}N_4O_8$ $[M+H]^+$ 501.1980 *m/z*; found 501.1975 *m/z*.

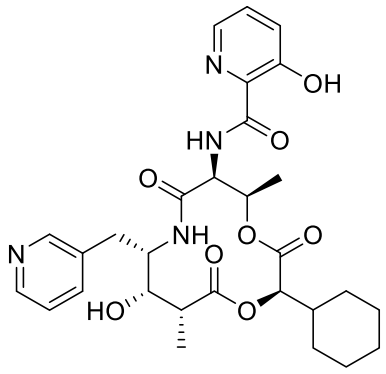
N-((2R,5R,6S,9S,10S,11R)-2-Ethyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)-3-hydroxypicolinamide (28b)



28b was prepared from **26b** according to the procedure described for **28a**. FC (DCM/MeOH, 98:2 to 95:5) provided a pale yellow oil. Further purification by RP-HPLC (20 to 40% MeCN/0.1% TFA in 0.1% aq. TFA) gave **28b** (4.07 mg, 45% over 2 steps) as a colorless film. **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.43. **$[\alpha]_D^{20}$** = –10.0° (*c* 0.20, $CHCl_3$). **^{1}H NMR** (500 MHz, $CDCl_3$): δ (ppm) = 8.83 (s, 1H), 8.52 (d, *J* = 5.2 Hz, 1H), 8.40 – 8.32 (m, 1H), 8.27 – 8.20 (m, 1H), 8.10 (d, *J* = 3.9 Hz, 1H), 7.59 (t, *J* = 6.8 Hz, 1H), 7.39 (dd, *J* = 8.5, 4.3 Hz, 1H), 7.32 (d, *J* = 8.3 Hz, 1H), 6.93 (br. s, 1H), 5.40 (p, *J* = 6.5 Hz, 1H), 5.01 (t, *J* = 6.1 Hz, 1H), 4.70 (t, *J* = 7.0 Hz, 1H),

4.24 (q, *J* = 7.7 Hz, 1H), 3.75 (s, 1H), 3.30 – 3.13 (m, 2H), 2.68 – 2.60 (m, 1H), 2.02 – 1.83 (m, 2H), 1.47 (d, *J* = 7.4 Hz, 3H), 1.32 (d, *J* = 6.5 Hz, 3H), 0.99 (t, *J* = 7.4 Hz, 3H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ (ppm) = 178.3, 168.2, 168.0, 168.0, 158.0, 145.2, 143.9, 140.6, 140.0, 138.1, 130.5, 129.5, 126.8, 125.9, 74.5, 74.4, 68.8, 56.1, 53.6, 41.1, 36.0, 24.2, 17.7, 14.4, 9.5. **IR** (thin film): υ (cm^{-1}) = 3361, 2927, 1745, 1730, 1669, 1650, 1517, 1449, 1380, 1289, 1259, 1171, 1137, 1095, 1061, 795, 749, 719. **HR-MS** (ESI): Calcd. for $C_{25}H_{31}N_4O_8$ $[M+H]^+$ 515.2136 *m/z*; found 515.2135 *m/z*.

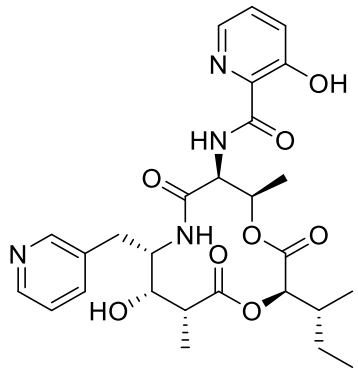
N-((2R,5R,6S,9S,10S,11R)-2-Cyclohexyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)-3-hydroxypicolinamide (28c)



28c was prepared from **26c** according to the procedure described for **28a**. FC (DCM/MeOH 95:5) provided **28c** (35.9 mg, 90%) as a pale yellow oil. Further purification by RP-HPLC (20 to 60% MeCN/0.1% TFA in 0.1% aq. TFA) gave **28c** (16.2 mg, 41% over 2 steps) as a white lyophilisate. **TLC** (SiO_2, EtOAc, UV): R_f = 0.21. **$[\alpha]_D^{20}$** = −24.9° (*c* 0.07, MeOH). **^{1}H-NMR** (500 MHz, $(CD_3)_2SO$): δ (ppm) = 11.81 (s, 1H), 8.42 (br. s, 1H), 8.37 (d, *J* = 1.8 Hz, 1H), 8.20 (dd, *J* = 4.8, 1.6 Hz, 1H), 8.16 (d, *J* = 4.0 Hz, 1H), 8.08 (d, *J* = 9.4 Hz, 1H), 7.61 – 7.51 (m, 2H), 7.45 (d, *J* = 8.3 Hz, 1H), 7.07 (dd, *J* = 7.7, 4.8 Hz, 1H), 5.28 (p, *J* = 6.4 Hz, 1H), 4.73 (d, *J* = 8.8 Hz, 1H), 4.71 – 4.65 (m, 2H), 4.11 – 4.03 (m, 1H), 3.61 (d, *J* = 8.9 Hz, 1H), 2.86 (dd, *J* = 13.4, 6.1 Hz, 1H),

2.76 (dd, *J* = 13.4, 8.7 Hz, 1H), 2.59 (q, *J* = 7.1 Hz, 1H), 1.86 – 1.76 (m, 1H), 1.68 (dd, *J* = 12.6, 1.7 Hz, 2H), 1.60 (d, *J* = 11.6 Hz, 3H), 1.27 (d, *J* = 7.3 Hz, 3H), 1.23 – 1.01 (m, 8H). **^{13}C-NMR** (126 MHz, $(CD_3)_2SO$): δ (ppm) = 176.2, 168.1, 168.1, 167.5, 157.9, 150.8, 147.6, 140.4, 137.0, 134.6, 130.9, 130.1, 126.9, 123.4, 77.1, 73.8, 68.8, 56.1, 53.3, 41.4, 38.9, 35.2, 28.5, 27.9, 26.0, 25.8, 25.6, 17.4, 15.1. **IR** (thin film): υ (cm^{-1}) = 2934, 1747, 1652, 1523, 1451, 1296, 1259, 1168, 1057, 1033, 1008, 685, 470. **HR-MS** (ESI): Calcd. for $C_{29}H_{37}N_4O_8$ $[M+H]^+$ 569.2606 *m/z*; found 569.2608 *m/z*.

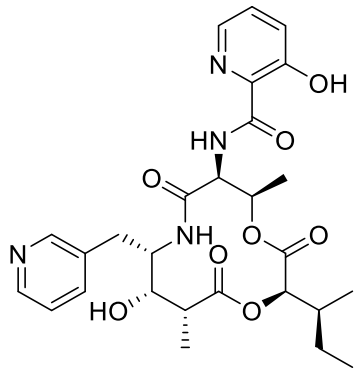
N-((2R,5R,6S,9S,10S,11R)-2-((R)-sec-Butyl)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)-3-hydroxypicolinamide (28d)



28d was prepared from **26d** according to the procedure described for **28a**. FC (DCM/MeOH 97.5:2.5 to 9:1) provided **28d** (5.40 mg, 49%) as a yellow oil. Part of this material was purified by RP-HPLC (30 to 100% MeCN/0.1% TFA in 0.1% aq. TFA) to provide a pure sample for biological testing (recovery yield not determined). **TLC** (DCM/MeOH 9:1): R_f = 0.26. **$[\alpha]_D^{20}$** = +13.0° (*c* 0.023, MeOH). **^{1}H-NMR** (500 MHz, $(CD_3)_2SO$): δ (ppm) = 11.89 (s, 1H), 8.37 (d, *J* = 2.2 Hz, 1H), 8.20 (dd, *J* = 4.8, 1.7 Hz, 1H), 8.18 (d, *J* = 4.4 Hz, 1H), 8.10 (d, *J* = 9.4 Hz, 1H), 7.61 – 7.53 (m, 2H), 7.46 (d, *J* = 8.5 Hz, 1H), 7.07 (dd, *J* = 7.8, 4.8 Hz, 1H), 5.27 (t, *J* = 6.4 Hz, 1H), 4.78 – 4.66 (m, 3H), 4.08 (q, *J* = 8.2 Hz, 1H), 3.62 (d, *J* = 9.5 Hz, 1H), 2.86 (dd, *J* = 13.4, 6.0 Hz, 1H),

2.77 (dd, *J* = 13.4, 8.7 Hz, 1H), 2.65 – 2.56 (m, 1H), 1.95 – 1.85 (m, 1H), 1.51 – 1.40 (m, 1H), 1.27 (d, *J* = 7.3 Hz, 3H), 1.21 (d, *J* = 6.5 Hz, 3H), 0.89 (d, *J* = 6.9 Hz, 3H), 0.86 (t, *J* = 7.5 Hz, 3H). **^{13}C-NMR** (126 MHz, $(CD_3)_2SO$): δ (ppm) = 168.4, 167.8, 167.4, 161.2, 151.0, 147.9, 145.2, 140.7, 137.3, 134.8, 131.7, 131.4, 130.4, 123.7, 77.1, 74.1, 69.2, 56.4, 53.6, 41.7, 36.1, 35.5, 24.9, 17.6, 15.3, 15.3, 11.7. **IR** (thin film): υ (cm^{-1}) = 2924, 2855, 1742, 1712, 1653, 1518, 1457, 1377, 1289, 1263, 842, 772, 731, 686, 558. **HR-MS** (ESI): Calcd. for $C_{27}H_{34}N_4O_8$ $[M+Na]^+$ 543.2449 *m/z*; found 543.2448 *m/z*.

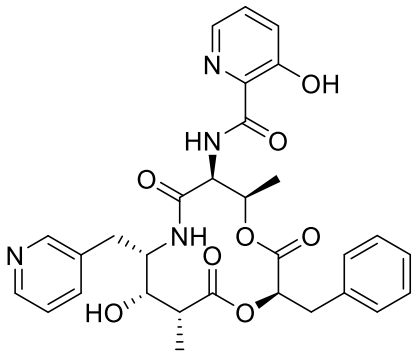
N-((2R,5R,6S,9S,10S,11R)-2-((S)-sec-Butyl)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)-3-hydroxypicolinamide (28e)



28e was prepared from **26e** according to the procedure described for **28a**. FC (DCM/MeOH 97.5:2.5 to 9:1) provided **28e** (6.7 mg, 66%) as a colorless film. Part of this material was purified by RP-HPLC (30 to 100% MeCN/0.1% TFA in 0.1% aq. TFA) to provide a pure sample for biological testing (recovery yield not determined). **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.26. **$[\alpha]_D^{20}$** = +8.7° (*c* 0.012, MeOH). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 11.78 (br. s, 1H), 8.37 (d, *J* = 2.1 Hz, 1H), 8.20 (dd, *J* = 4.8, 1.7 Hz, 1H), 8.16 (d, *J* = 4.4 Hz, 1H), 8.09 (d, *J* = 9.5 Hz, 1H), 7.60 – 7.53 (m, 2H), 7.45 (d, *J* = 8.5 Hz, 1H), 7.07 (dd, *J* = 7.8, 4.8 Hz, 1H), 5.29 (p, *J* = 6.4 Hz, 1H), 4.86 (d, *J* = 4.4 Hz, 1H), 4.77 – 4.67 (m, 2H), 4.08 (q, *J* = 8.9 Hz, 1H), 3.61 (d, *J* = 9.1

Hz, 1H), 2.87 (dd, *J* = 13.5, 6.2 Hz, 1H), 2.76 (dd, *J* = 13.4, 8.6 Hz, 1H), 2.60 (q, *J* = 7.7 Hz, 1H), 1.94 (p, *J* = 6.6 Hz, 1H), 1.40 (dt, *J* = 13.8, 6.9 Hz, 1H), 1.27 (d, *J* = 7.3 Hz, 3H), 1.20 (d, *J* = 6.5 Hz, 3H), 0.90 – 0.83 (m, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 175.7, 168.0, 167.6, 167.0, 150.3, 147.2, 139.9, 136.6, 134.1, 130.4, 129.7, 126.4, 123.0, 119.6, 75.0, 73.3, 68.2, 55.6, 52.7, 41.0, 35.8, 34.8, 25.0, 17.0, 14.4, 14.2, 11.2. **IR** (thin film): υ (cm^{-1}) = 3368, 2966, 2931, 1742, 1652, 1524, 1451, 1385, 1294, 1255, 1169, 1063, 815, 784, 712, 662. **HR-MS** (ESI): Calcd. for $C_{27}H_{34}N_4O_8$ $[M+H]^+$ 543.2449 *m/z*; found 543.2441 *m/z*.

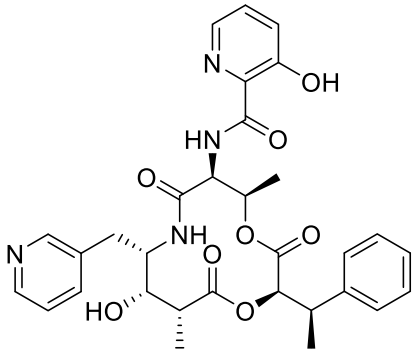
N-((2R,5R,6S,9S,10S,11R)-2-Benzyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)-3-hydroxypicolinamide (28f)



28f was prepared from **26f** according to the procedure described for **28a**. FC (DCM/MeOH, 98:2 to 95:5) provided **28f** as a pale yellow oil. Further purification by RP-HPLC (20 to 60% MeCN/0.1% TFA in 0.1% aq. TFA) gave **28f** (5.36 mg, 65% over 2 steps) as a white lyophilisate. **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.53. **[α]$_D^{20}$** = –7.4° (*c* 0.27, $CHCl_3$). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ (ppm) = 8.81 (s, 1H), 8.50 (d, *J* = 5.1 Hz, 1H), 8.31 (d, *J* = 6.7 Hz, 1H), 8.25 – 8.20 (m, 1H), 8.10 (d, *J* = 4.2 Hz, 1H), 7.63 – 7.55 (m, 1H), 7.40 – 7.36 (m, 1H), 7.34 – 7.30 (m, 1H), 7.29 – 7.25 (m, 2H), 7.25 – 7.22 (m, 1H), 7.21 – 7.18 (m, 2H), 7.06 – 6.91 (m, 1H), 5.37 – 5.32 (m, 1H), 5.30 (dd, *J* = 8.8, 4.3 Hz, 1H), 4.75 – 4.65 (m, 1H), 4.28 – 4.19 (m, 1H), 3.72 (br. s,

1H), 3.29 – 3.07 (m, 4H), 2.59 (q, *J* = 7.2 Hz, 1H), 1.28 (d, *J* = 7.4 Hz, 3H), 1.18 (d, *J* = 5.6 Hz, 3H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ (ppm) = 177.7, 168.1, 167.4, 157.9, 145.4, 143.7, 140.4, 139.9, 138.2, 135.4, 130.5, 129.5, 129.4, 128.7, 127.4, 126.8, 125.9, 74.3, 73.6, 69.0, 56.0, 53.5, 41.1, 36.7, 36.0, 17.4, 14.1. **IR** (thin film): υ (cm^{-1}) = 3361, 3064, 2935, 1745, 1669, 1650, 1517, 1456, 1380, 1255, 1186, 1133, 1061, 799, 741. **HR-MS** (ESI): Calcd. for $C_{30}H_{33}N_4O_8$ $[M+H]^+$ 577.2293 *m/z*; found 577.2290 *m/z*.

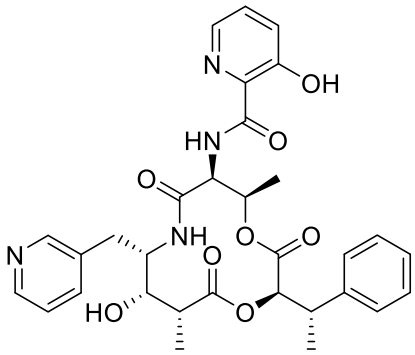
3-Hydroxy-N-((2R,5R,6S,9S,10S,11R)-10-Hydroxy-5,11-dimethyl-3,7,12-trioxo-2-((R)-1-phenylethyl)-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)picolinamide (28g)



28g was prepared from **26g** according to the procedure described for **28a**. FC (DCM/MeOH 95:5) followed by RP-HPLC (20 to 85% MeCN/0.1% TFA in 0.1% aq. TFA) provided **28g** (17.0 mg, 36% over 2 steps) as a white lyophilisate. **TLC** (SiO_2, DCM/MeOH 95:5, UV, $KMnO_4$): R_f = 0.26. **[α]$_D^{20}$** = –8.0° (*c* 0.50, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.83 (s, 1H), 8.56 – 8.45 (m, 1H), 8.22 (dd, *J* = 8.2, 1.8 Hz, 2H), 8.09 (dd, *J* = 4.3, 1.5 Hz, 1H), 7.59 (dd, *J* = 7.9, 5.5 Hz, 1H), 7.42 – 7.30 (m, 2H), 7.25 – 7.18 (m, 4H), 6.77 (d, *J* = 9.1 Hz, 1H), 5.26 (q, *J* = 6.5 Hz, 1H), 5.20 (d, *J* = 5.7 Hz, 1H), 4.61 (dd, *J* = 8.3, 6.1 Hz, 1H), 4.28 – 4.16 (m, 1H), 3.74 (s, 1H), 3.50 – 3.39 (m, 1H), 3.28 – 3.11 (m, 2H), 2.68 – 2.54 (m, 1H), 1.41 (dd, *J* = 7.3, 4.9 Hz, 6H), 0.85 (d, *J* = 6.5 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 178.1, 168.0, 166.9, 162.5, 162.1,

158.0, 145.5, 143.6, 140.3, 139.7, 138.3, 130.3, 129.5, 128.5, 128.3, 127.5, 125.9, 77.2, 74.5, 68.3, 56.0, 53.5, 41.1, 40.9, 36.0, 18.0, 17.6, 13.7. **IR** (thin film): υ (cm^{-1}) = 1743, 1671, 1648, 1602, 1522, 1450, 1387, 1296, 1266, 1172, 1137, 1062, 796, 753, 720, 699, 660, 621, 572, 544, 521. **HR-MS** (ESI): Calcd. for $C_{31}H_{35}N_4O_8$ $[M+H]^+$ 591.2449 *m/z*; found 591.2446 *m/z*.

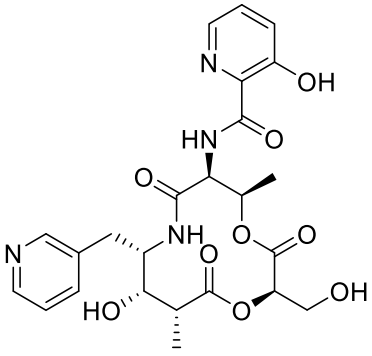
3-Hydroxy-N-((2R,5R,6S,9S,10S,11R)-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-2-((S)-1-phenylethyl)-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)picolinamide (28h)



28h was prepared from **26h** according to the procedure described for **28a**. FC (DCM/MeOH 95:5) followed by RP-HPLC (20 to 85% MeCN/0.1% TFA in 0.1% aq. TFA) provided **28h** (3.75 mg, 43% over 2 steps) as a white lyophilisate. **TLC** (SiO_2, DCM/MeOH 9:1, UV, CPS): R_f = 0.47. **[α]D^{20}** = –23.3° (*c* 0.30, MeOH). **^{1}H-NMR** (500 MHz, CD_3OD): δ (ppm) = 8.75 (d, *J* = 4.6 Hz, 1H), 8.52 (d, *J* = 5.9 Hz, 1H), 8.39 (d, *J* = 7.7 Hz, 1H), 8.18 (dd, *J* = 4.3, 1.3 Hz, 1H), 8.16 – 8.12 (m, 1H), 7.70 (dd, *J* = 8.0, 5.6 Hz, 1H), 7.52 (dd, *J* = 8.6, 4.4 Hz, 1H), 7.41 (dd, *J* = 8.5, 1.4 Hz, 1H), 7.30 (d, *J* = 4.3 Hz, 4H), 7.27 – 7.21 (m, 1H), 5.29 (p, *J* = 6.5 Hz, 1H), 5.08 (d, *J* = 4.8 Hz, 1H), 4.66 (dd, *J* = 6.3, 2.3 Hz, 1H), 4.34 – 4.22 (m, 1H), 3.82 – 3.74 (m, 1H), 3.45 (qd, *J* = 7.1, 4.8 Hz, 1H), 3.24 – 3.09 (m, 2H), 2.72 – 2.64 (m, 1H), 1.38 (d, *J* = 7.1 Hz, 3H), 1.30 (d, *J* = 7.3 Hz, 3H), 1.23 (d, *J* = 6.5 Hz, 3H). **^{13}C-NMR** (126 MHz, CD_3OD): δ (ppm) = 176.6, 168.8, 167.9, 167.1, 157.8, 141.0, 139.8, 130.3, 129.4, 129.3, 128.1, 128.0, 127.5, 127.4, 127.0, 126.8, 126.3,

126.0, 77.1, 74.8, 68.7, 55.5, 53.2, 40.9, 40.6, 35.4, 16.3, 14.3, 13.4. **IR** (thin film): υ (cm^{-1}) = 1742, 1672, 1526, 1451, 1388, 1298, 1262, 1175, 1138, 1061, 702, 484, 472, 418. **HR-MS** (ESI): Calcd. for $C_{31}H_{35}N_4O_8$ $[M+H]^+$ 591.2449 *m/z*; found 591.2450 *m/z*.

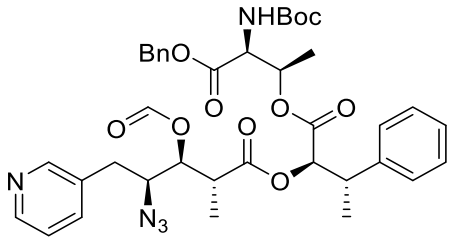
3-Hydroxy-N-((2R,5R,6S,9S,10S,11R)-10-hydroxy-2-(hydroxymethyl)-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)picolinamide (28i)



28i was prepared from **26i** according to the procedure described for **28a**. FC (DCM/MeOH, 98:2 to 95:5) provided **28i** (10.7 mg, quant.) as a colorless film. Further purification by RP-HPLC (5 to 50% MeCN in H_2O) gave **28i** (2.90 mg, 30% yield over 2 steps) as a white lyophilisate. **TLC** (SiO_2, DCM/MeOH 9:1, UV): R_f = 0.27. **$[\alpha]_D^{20}$** = −15.9° (*c* 0.47, MeOH). **^{1}H-NMR** (500 MHz, $(CD_3)_2SO$): δ (ppm) = 11.80 (br. s, 1H), 8.45 (br. s, 1H), 8.38 (d, *J* = 2.1 Hz, 1H), 8.21 (dd, *J* = 4.7, 0.9 Hz, 1H), 8.16 (d, *J* = 4.0 Hz, 1H), 8.06 (d, *J* = 9.3 Hz, 1H), 7.60 – 7.53 (m, 2H), 7.44 (d, *J* = 8.5 Hz, 1H), 7.09 (dd, *J* = 7.7, 4.8 Hz, 1H), 5.32 – 5.23 (m, 2H), 4.95 (dd, *J* = 4.8, 3.8 Hz, 1H), 4.79 (d, *J* = 8.6 Hz, 1H), 4.76 – 4.70 (m, 1H), 4.10 – 4.03 (m, 1H), 3.84 – 3.76 (m, 1H), 3.76 – 3.68 (m, 1H), 3.62 (d, *J* = 9.0 Hz, 1H), 2.87 (dd, *J* = 13.4, 6.3 Hz, 1H), 2.77 (dd, *J* = 13.4, 8.3 Hz, 1H), 2.59 (qd, *J* = 7.0, 0.9 Hz, 1H), 1.28 (d, *J* = 7.3 Hz, 3H), 1.18 (d, *J* = 6.4 Hz, 3H). **^{13}C-NMR** (126 MHz, $(CD_3)_2SO$): δ (ppm) = 176.0, 168.1, 167.6, 167.4, 150.8, 147.7, 140.3, 137.1, 134.6, 131.0, 130.1, 126.9, 123.5, 74.8, 73.5, 68.6, 60.9, 56.2, 53.1, 41.6, 35.3, 17.2, 14.8. **IR** (thin film):

υ (cm^{-1}) = 3354, 1676, 1542, 1455, 1203, 1136, 844, 800, 779, 722, 558, 458, 433. **HR-MS** (ESI): Calcd. for $C_{24}H_{29}N_4O_9$ $[M+H]^+$ 517.1922 *m/z*; found 517.1922 *m/z*.

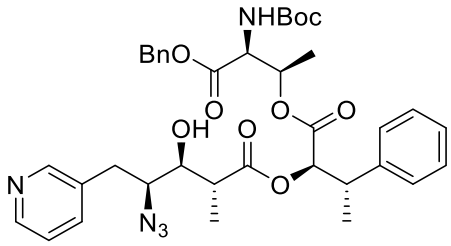
(2R,3S)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-oxo-3-phenylbutan-2-yl (2R,3S,4S)-4-azido-3-(formyloxy)-2-methyl-5-(pyridin-3-yl)pentanoate (30)



To a stirred solution of **29** (63.9 mg, 230 µmol, 1.00 eq.), DMAP (2.8 mg, 23.0 µmol, 0.10 eq.) and **8h** (130 mg, 0.28 mmol, 1.20 eq.) in DCM (2.3 mL) was added DCC (56.9 mg, 276 µmol, 1.20 eq.) at 0 °C. The mixture was allowed to warm to rt and stirred for 5 h. The reaction mixture was diluted with hexane (3 mL), filtered through a pad of celite and concentrated *in vacuo*. FC (hexane/EtOAc 1:1) provides **30** (132 mg, 78%) as a colorless semisolid. **TLC** (SiO_2, hexane/EtOAc 1:5, UV, $KMnO_4$): R_f = 0.45. **[α]$_D^{20}$** = –9.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.56 – 8.49 (m, 2H), 8.11 (s, 1H), 7.60 (dt, *J* = 7.9, 2.0 Hz, 1H), 7.39 – 7.27 (m, 9H), 7.23 (dd, *J* = 7.1, 2.9 Hz, 2H), 5.50 – 5.42 (m, 1H), 5.37 (ddd, *J* = 6.7, 4.4, 0.9 Hz, 1H), 5.22 – 5.07 (m, 4H), 4.49 (dd, *J* = 9.9, 2.8 Hz, 1H), 3.73 (dt, *J* = 10.2, 4.0 Hz, 1H), 3.33 – 3.25 (m, 1H), 3.14 – 3.05 (m, 1H), 3.00 (dd, *J* = 14.3, 3.9 Hz, 1H), 2.79 (dd, *J* = 14.3, 10.3 Hz, 1H), 1.46 (s, 9H), 1.30 (d, *J* = 6.9 Hz, 6H), 1.17 (d, *J* = 7.1 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 172.1, 170.0, 167.7, 160.0, 155.9, 150.4, 148.5, 141.1, 137.4, 137.1, 135.0, 132.5, 128.8, 128.7, 128.6, 128.5, 127.8, 127.4, 123.8, 80.6, 76.9, 74.8, 72.6, 67.9, 62.7, 57.2, 41.4, 40.6, 34.7, 28.4, 16.9, 15.1, 13.2. **IR** (thin film): υ (cm^{-1}) = 2979, 2934, 2115, 1742, 1498, 1455, 1384, 1367, 1346,

1314, 1260, 1212, 1164, 1086, 1062, 756, 701. **HR-MS** (ESI): Calcd. for $C_{38}H_{46}N_5O_{10}$ $[M+H]^+$ 732.3239 *m/z*; found 732.3229 *m/z*.

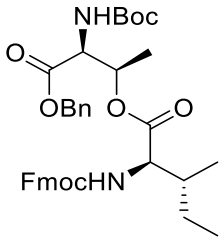
(2R,3S)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-oxo-3-phenylbutan-2-yl (2R,3S,4S)-4-azido-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (31)



To a stirred solution of **30** (47.3 mg, 64.6 µmol, 1.00 eq.) in MeOH (1.5 mL) was added $KHCO_3$ (7.1 mg, 71.1 µmol, 1.10 eq.). The mixture was stirred at rt for 65 min, the reaction was quenched with sat. aq. NaCl solution (5 mL) and the mixture was extracted with EtOAc (3 x 10 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 1:1) provided **31** (40.4 mg, 89%) as a slight yellow oil. **TLC** (SiO_2, hexane/EtOAc 1:3, UV, $KMnO_4$): R_f = 0.37. **$[\alpha]_D^{20}$** = +13.0° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.74 – 8.41 (m, 2H), 7.75 (d, *J* = 7.9 Hz, 1H), 7.44 – 7.27 (m, 10H), 7.26 – 7.21 (m, 1H), 5.61 – 5.49 (m, 2H), 5.20 – 5.01 (m, 3H), 4.54 (dd, *J* = 9.9, 2.7 Hz, 1H), 3.86 (d, *J* = 5.4 Hz, 1H), 3.75 – 3.66 (m, 1H), 3.45 – 3.32 (m, 2H), 3.25 – 3.09 (m, 2H), 2.92 (dq, *J* = 8.7, 7.0 Hz, 1H), 1.47 (s, 9H), 1.34 (d, *J* = 7.2 Hz, 3H), 1.29 (d, *J* = 6.4 Hz, 3H), 0.96 (d, *J* = 7.1 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.7, 170.5, 168.4, 156.2, 150.4, 148.3, 141.3, 137.2, 133.3, 128.7, 128.6, 128.6, 128.2, 127.8, 127.8, 127.4, 123.8, 80.5, 76.7, 74.2, 72.6, 68.2, 62.9, 57.3, 43.6, 40.9, 34.1, 34.1, 28.4, 28.4, 28.4, 28.3, 28.2, 25.7, 25.1, 16.9, 16.9, 14.8, 13.4. **IR** (thin film): υ (cm^{-1}) =

2978, 2932, 2112, 1743, 1715, 1508, 1499, 1455, 1382, 1367, 1339, 1314, 1260, 1217, 1165, 1086, 1060, 992, 754, 700. **HR-MS** (ESI): Calcd. for $C_{37}H_{46}N_5O_9$ $[M+H]^+$ 704.3290 *m/z*; found 704.3290 *m/z*.

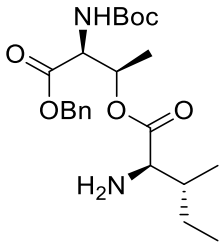
(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl (((9H-fluoren-9-yl)methoxy)carbonyl)-D-isoleucinate (32A)



To a stirred solution of Fmoc-D-Ile-OH (**32**) (1.40 g, 3.96 mmol, 1.00 eq.) and Et_3N (1.10 mL, 7.92 mmol, 2.00 eq.) in THF (20 mL) was added 2,4,6-trichlorobenzoyl chloride (712 µL, 4.56 mmol, 1.15 eq.). As the mixture became turbid (5 min), a solution of **6** (1.23 g, 3.96 mmol, 1.00 eq.) in toluene (20 mL) and DMAP (72.6 mg, 594 µmol, 0.15 eq.) were added at rt. The white slurry was stirred at rt for 1 h. Sat. aq. NH_4Cl (10 mL) was added and the aqueous layer was extracted with EtOAc (3 x 15 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 4:1) provided **32A** (2.51 g, 98%) as a white foam. **TLC** (SiO_2, hexane/EtOAc 4:1, UV): R_f = 0.24. **$[\alpha]_D^{20}$** = +24.9° (*c* 1.30, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.78 (d, *J* = 0.5 Hz, 1H), 7.76 (d, *J* = 0.6 Hz, 1H), 7.64 – 7.58 (m, 2H), 7.44 – 7.29 (m, 9H), 5.53 – 5.42 (m, 1H), 5.24 – 5.19 (m, 1H, NH), 5.12 (dd, *J* = 30.9, 12.2 Hz, 2H), 4.50 (dd, *J* = 9.5, 1.8 Hz, 1H), 4.47 – 4.36 (m, 2H), 4.31 – 4.20 (m, 2H), 1.89 – 1.75 (m, 1H), 1.64 (s, 1H), 1.46 (s, 9H), 1.36 – 1.29 (m, 4H), 1.17 – 1.02 (m, 1H), 0.97 – 0.86 (m, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 170.9, 169.9, 156.1, 155.8, 143.9, 143.8, 141.3 (2 C), 134.9, 128.7 (2 C), 128.6 (2 C), 128.3, 127.7 (2 C), 127.1 (2 C), 125.1 (2 C), 120.0, 120.0, 80.5, 72.2,

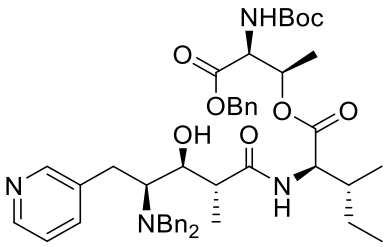
67.7, 67.1, 58.7, 57.1, 47.2, 37.6, 28.3 (3 C), 24.6, 17.0, 15.4, 11.5. **IR** (thin film): υ (cm^{-1}) = 2969, 1712, 1504, 1452, 1384, 1367, 1316, 1248, 1211, 1162, 1085, 1063, 996, 756, 739, 698. **HR-MS** (ESI): Calcd. for $C_{37}H_{44}N_2NaO_8$ $[M+Na]^+$ 667.2990 *m/z*; found 667.2988 *m/z*.

(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl D-isoleucinate (33)



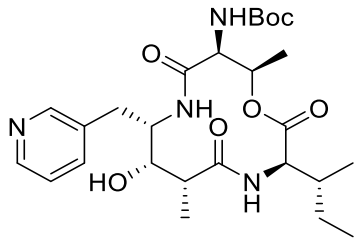
32A (800 mg, 1.24 mmol, 1.00 eq.) was dissolved in MeCN (14 mL) and piperidine (3.5 mL). The solution was stirred at rt for 30 min and then concentrated *in vacuo* very quickly to remove most of the solvent. FC (hexane/EtOAc 1:0 to 1:4) provided **33** (337 mg, 64%) as a yellow oil. **TLC** (SiO_2, hexane/EtOAc 1:4, UV): R_f = 0.35. **[α]D^{20}** = +26.6° (*c* 1.00, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 7.39 – 7.30 (m, 5H), 5.44 (qd, *J* = 6.3, 2.5 Hz, 1H), 5.24 – 5.04 (m, 3H), 4.48 (dd, *J* = 9.7, 2.4 Hz, 1H), 3.20 (d, *J* = 5.1 Hz, 1H), 1.71 – 1.59 (m, 1H), 1.45 (s, 9H), 1.36 – 1.32 (m, 1H), 1.30 (d, *J* = 6.4 Hz, 3H), 1.18 – 1.05 (m, 1H), 0.91 – 0.84 (m, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.3, 170.0, 155.8, 135.0, 128.7 (3 C), 128.6, 128.4, 80.4, 71.2, 67.6, 59.5, 57.2, 38.6, 28.3 (3 C), 24.2, 17.0, 15.7, 11.6. **IR** (thin film): υ (cm^{-1}) = 2968, 2935, 1717, 1502, 1457, 1368, 1345, 1314, 1247, 1215, 1163, 1085, 1063, 993, 699. **HR-MS** (ESI): Calcd. for $C_{22}H_{35}N_2O_6$ $[M+H]^+$ 423.2490 *m/z*; found 423.2493 *m/z*.

(2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl ((2R,3S,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoyl)-D-isoleucinate (34)



To a solution of **24** (30.0 mg, 74.2 µmol, 1.00 eq.) and **33** (34.5 mg, 81.6 µmol, 1.10 eq.) in MeCN (1 mL) and THF (0.1 mL) were added HATU (62.0 mg, 163 µmol, 2.2 eq.) and DIPEA (41.1 µL, 237 µmol, 3.20 eq.) at 0 °C. The mixture was stirred for 48 h was slowly allowed to warm to rt. The mixture was diluted with EtOAc (1.5 mL) and sat. aq. $NaHCO_3$ (2 mL). The aqueous layer was extracted with EtOAc (3 x 5 mL) and the combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (hexane/EtOAc 2:3) provided **34** (44.8 mg, 75%) as a colorless film. **TLC** (SiO_2, hexane/EtOAc 2:3, UV): R_f = 0.22. **$[\alpha]_D^{20}$** = +25.5° (*c* 1.00, MeOH). **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 8.41 (d, *J* = 1.7 Hz, 1H), 8.35 (dd, *J* = 4.9, 1.6 Hz, 1H), 7.65 – 7.59 (m, 1H), 7.37 – 7.24 (m, 14H), 7.23 – 7.16 (m, 2H), 5.42 (qd, *J* = 6.3, 3.0 Hz, 1H), 4.42 (d, *J* = 2.9 Hz, 1H), 4.24 (d, *J* = 5.9 Hz, 1H), 4.14 (d, *J* = 13.3 Hz, 2H), 3.58 (d, *J* = 13.4 Hz, 3H), 3.11 – 2.99 (m, 2H), 2.95 – 2.85 (m, 2H), 1.83 – 1.71 (m, 1H), 1.45 (s, 9H), 1.37 – 1.31 (m, 1H), 1.23 (d, *J* = 6.4 Hz, 3H), 1.15 – 1.03 (m, 1H), 0.85 (t, *J* = 7.4 Hz, 3H), 0.80 (d, *J* = 6.8 Hz, 3H), 0.65 (d, *J* = 6.9 Hz, 3H). **^{13}C-NMR** (101 MHz, CD_3OD): δ (ppm) = 178.9, 172.0, 171.5, 158.3, 151.1, 147.5, 141.5, 139.3 (2 C), 138.5, 136.8, 130.5 (6 C), 129.6, 129.4, 129.3 (6 C), 128.1, 125.2, 81.1, 79.5, 75.4, 72.6, 68.5, 61.8, 58.8, 58.3, 56.5, 44.3, 37.8, 29.5, 28.8, 26.1, 17.2, 16.0, 15.7, 11.8. **IR** (thin film): υ (cm^{-1}) = 2970, 2935, 2362, 2355, 1742, 1719, 1667, 1501, 1455, 1368, 1250, 1165, 1134, 1063, 838, 751, 701, 424, 415; **HR-MS** (ESI): Calcd. for $C_{47}H_{61}N_4O_8$ $[M+H]^+$ 809.4484 *m/z*; found 809.4465 *m/z*.

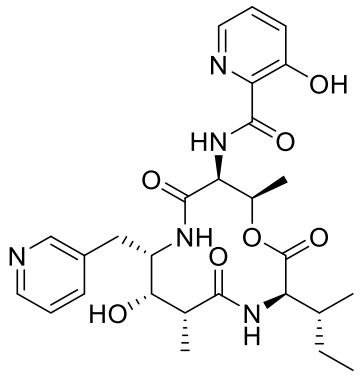
tert-Butyl ((3R,6R,7S,8S,11S,12R)-3-((R)-sec-butyl)-7-hydroxy-6,12-dimethyl-2,5,10-trioxo-8-(pyridin-3-ylmethyl)-1-oxa-4,9-diazacyclododecan-11-yl)carbamate (34A)



34 (17.1 mg, 21.1 µmol, 1.00 eq.) was dissolved in MeOH (1 mL) and Pd/C (10%, 9.00 mg, 8.45 µmol, 0.40 eq.) was added under argon. The atmosphere was exchanged with H_2 (1 bar) and the mixture was stirred at rt for 2.75 h. The suspension was filtered over celite, the filter cake was washed with MeOH and the filtrate was concentrated *in vacuo* to deliver the ensuing amino acid (11.0 mg, 96%, crude) as a white solid. To a suspension of DIPEA (36.3 µL, 210 µmol, 2.60 eq.) and HATU (52.2 mg, 137 µmol, 1.70 eq.) in DCM (30 mL), the above amino acid (43.5 mg, 80.8 µmol, 1.00 eq.) was added in 1% DMF/DCM (15 ml) at rt over 1.5 h (pale yellow color develops). The solution was stirred at rt for 18 h. Sat. aq. $NaHCO_3$ (10 mL) was added and the aqueous layer was extracted with DCM (3 x 15 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated *in vacuo*. FC (DCM/MeOH 95:5) provided **34A** (34.9 mg, 83%) as a yellow solid. **TLC** (SiO_2, DCM/MeOH 95:5, UV): R_f = 0.24. **[α]D^{20}** = −25.1° (*c* 1.40, MeOH). **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 8.46 (d, *J* = 1.6 Hz, 1H), 8.36 (dd, *J* = 4.9, 1.4 Hz, 1H), 7.72 (d, *J* = 7.8 Hz, 1H), 7.33 (dd, *J* = 7.7, 5.0 Hz, 1H), 5.18 – 5.07 (m, 1H), 4.29 (d, *J* = 5.8 Hz, 1H), 4.08 (t, *J* = 7.0 Hz, 1H), 4.01 (d, *J* = 7.6 Hz, 1H), 3.59 (s, 1H), 2.98 (dd, *J* = 13.5, 7.2 Hz, 1H), 2.91 (dd, *J* = 13.5, 7.7 Hz, 1H), 2.45 (qd, *J* = 7.0, 0.7 Hz, 1H), 1.89 – 1.77 (m, 1H), 1.59 – 1.50 (m, 1H), 1.46 (s, 9H), 1.33 (d, *J* = 5.7 Hz, 3H), 1.31 (d, *J* = 5.0 Hz, 3H), 1.29 – 1.18 (m, 1H), 0.95 – 0.88 (m, 6H). **^{13}C-NMR** (101 MHz, CD_3OD): δ (ppm) = 180.5, 171.3, 170.9, 157.2, 151.1, 148.0, 139.3, 136.4, 125.0, 81.0, 76.5, 70.8, 59.7, 57.8, 56.5, 41.8, 37.0, 36.4, 28.7, 26.8, 19.1,

15.9, 15.0, 11.3. **IR** (thin film): υ (cm^{-1}) = 3339, 2970, 2934, 1737, 1718, 1664, 1535, 1496, 1457, 1388, 1368, 1317, 1244, 1167, 1064, 1049, 1027, 844, 716, 557. **HR-MS** (ESI): Calcd. for $C_{26}H_{41}N_4O_7$ $[M+H]^+$ 521.2970 *m/z*; found 521.2969 *m/z*.

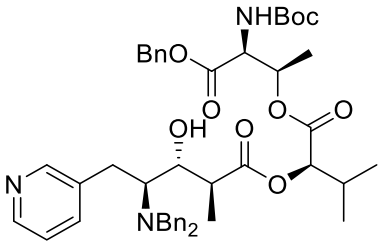
N-((3R,6R,7S,8S,11S,12R)-3-((R)-sec-Butyl)-7-hydroxy-6,12-dimethyl-2,5,10-trioxo-8-(pyridin-3-ylmethyl)-1-oxa-4,9-diazacyclododecan-11-yl)-3-hydroxypicolinamide (35)



35 was prepared from **34A** according to the procedure described for **28a**. FC (DCM/MeOH 95:5) followed by RP-HPLC (10 to 50% MeCN/0.1% TFA in 0.1% aq. TFA) provided **35** (5.1 mg, 25% over 2 steps) as a white lyophilisate. **TLC** (SiO_2, DCM/MeOH 95:5, UV): R_f = 0.21. **$[\alpha]_D^{20}$** = −12.4° (*c* 0.15, MeOH). **^{1}H-NMR** (500 MHz, $(CD_3)_2SO$): δ (ppm) = 8.69 (s, 1H), 8.65 (d, *J* = 7.0 Hz, 1H), 8.56 (d, *J* = 4.7 Hz, 1H), 8.24 (d, *J* = 7.5 Hz, 1H), 8.20 (dd, *J* = 4.4, 1.2 Hz, 1H), 8.14 (d, *J* = 8.6 Hz, 1H), 8.02 (d, *J* = 8.2 Hz, 1H), 7.66 (t, *J* = 6.6 Hz, 1H), 7.60 (dd, *J* = 8.5, 4.4 Hz, 1H), 7.47 (dd, *J* = 8.5, 1.1 Hz, 1H), 5.22 (p, *J* = 6.4 Hz, 1H), 4.65 (dd, *J* = 8.2, 5.9 Hz, 1H), 4.14 (tdd, *J* = 9.9, 4.5, 1.3 Hz, 1H), 3.90 (t, *J* = 7.3 Hz, 1H), 3.66 (s, 1H), 3.04 (dd, *J* = 13.2, 4.5 Hz, 1H), 2.96 (dd, *J* = 13.2, 10.2 Hz, 1H), 2.57 – 2.52 (m, 1H), 1.79 – 1.69 (m, 1H), 1.51 – 1.41 (m, 1H), 1.27 (d, *J* = 7.2 Hz, 3H), 1.25 – 1.17 (m, 1H), 1.16 (d, *J* = 6.5 Hz, 3H), 0.87 – 0.79 (m, 6H). **^{13}C-NMR** (126 MHz, $(CD_3)_2SO$): δ (ppm) = 178.7, 170.1, 167.9, 167.8, 158.9 (q, *J* = 34.4 Hz, TFA), 157.6, 145.7, 143.9, 141.1, 140.7, 138.8, 130.6, 130.3, 126.9, 126.2, 116.8 (d, *J* = 293.0 Hz, TFA),

76.2, 68.5, 58.5, 56.0, 53.5, 39.8, 35.8, 35.0, 25.6, 19.0, 15.7, 15.1, 11.3. **IR** (thin film): υ (cm^{-1}) = 3361, 3289, 2365, 1970, 1742, 1673, 1651, 1526, 1452, 1199, 1138, 801, 723, 682, 651, 567, 458. **HR-MS** (ESI): Calcd. for $C_{27}H_{36}N_5O_7$ $[M+H]^+$ 542.2609 *m/z*; found 542.2609 *m/z*.

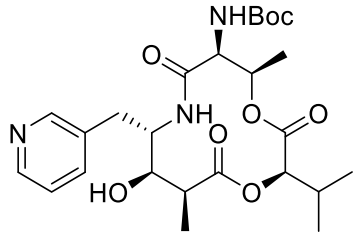
(R)-1-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-3-methyl-1-oxobutan-2-yl (2S,3R,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (37)



37 was prepared from bis-*epi*-**24** and **36**[24] according to the procedure described for **25a**. FC (hexane/EtOAc 3:2) provided **37** (82.6 mg, 52%) as a pale yellow oil. **TLC** (SiO_2, hexane/EtOAc, 1:1, UV): R_f = 0.29. **$[\alpha]_D^{20}$** = +11.9° (*c* 0.51, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.49 (dd, *J* = 4.8, 1.6 Hz, 1H), 8.42 (d, *J* = 1.8 Hz, 1H), 7.40 – 7.30 (m, 7H), 7.23 – 7.13 (m, 7H), 7.12 – 7.07 (m, 4H), 5.57 – 5.50 (m, 1H), 5.45 (d, *J* = 9.7 Hz, 1H), 5.18 (d, *J* = 12.1 Hz, 1H), 5.10 (d, *J* = 12.1 Hz, 1H), 4.85 (d, *J* = 3.7 Hz, 1H), 4.53 (dd, *J* = 9.7, 2.3 Hz, 1H), 4.27 – 4.20 (m, 1H), 3.97 (d, *J* = 14.3 Hz, 2H), 3.58 (d, *J* = 14.3 Hz, 2H), 3.50 (d, *J* = 4.6 Hz, 1H), 3.08 (dd, *J* = 14.2, 9.3 Hz, 1H), 2.99 – 2.92 (m, 1H), 2.86 (dd, *J* = 14.2, 3.8 Hz, 1H), 2.69 – 2.59 (m, 1H), 2.27 – 2.15 (m, 2H), 1.45 (s, 9H), 1.33 (d, *J* = 6.4 Hz, 3H), 1.02 (dd, *J* = 9.7, 7.1 Hz, 6H), 0.95 (d, *J* = 6.9 Hz, 6H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.1, 169.9, 169.3, 156.0, 151.0, 147.3, 139.8, 137.1, 136.4, 134.9, 128.8, 128.7, 128.6, 128.5, 128.4, 128.3, 127.0, 123.1, 80.5, 76.3, 72.9, 72.0, 67.9, 60.2, 57.1, 54.5, 45.2, 30.1, 29.0, 28.4, 19.0, 16.9, 14.8. **IR** (thin film): υ (cm^{-1}) = 3026, 2976,

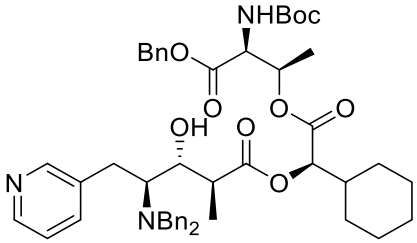
2935, 2802, 1739, 1723, 1495, 1455, 1367, 1314, 1252, 1212, 1166, 1128, 1084, 1023, 985, 749, 699. **HR-MS** (ESI): Calcd. for $C_{46}H_{58}N_3O_9$ $[M+H]^+$ 796.4168 *m/z*; found 796.4159 *m/z*.

tert-Butyl ((2R,5R,6S,9S,10R,11S)-10-hydroxy-2-isopropyl-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (37A)



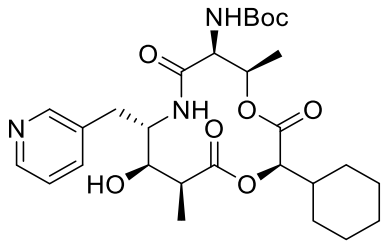
37A was prepared from **37** according to the procedure described for **26a**. FC (hexane/EtOAc 5:95) provided **37A** (13.0 mg, 33% over 2 steps) as a pale yellow film. **TLC** (SiO_2, hexane/EtOAc, 0.5:10): R_f = 0.28. **$[\alpha]_D^{20}$** = –10.4 (*c* 0.37, MeOH). **^{1}H-NMR** (500 MHz, CD_3OD): δ (ppm) = 8.43 (s, 1H), 8.34 (d, *J* = 3.6 Hz, 1H), 7.69 (d, *J* = 7.8 Hz, 1H), 7.27 (dd, *J* = 7.5, 5.0 Hz, 1H), 7.21 (d, *J* = 9.4 Hz, 1H exchanged), 6.22 (d, *J* = 8.5 Hz, 1H exchanged), 5.23 – 5.13 (m, 1H), 4.88 – 4.78 (m, 1H), 4.41 (d, *J* = 8.4 Hz, 1H), 4.13 (d, *J* = 5.4 Hz, 1H), 4.06 (br. s, 1H), 3.33 – 3.28 (m, 1H), 2.90 (qd, *J* = 7.1, 2.2 Hz, 1H), 2.74 (br. s, 1H), 2.30 – 2.18 (m, 1H), 1.45 (s, 9H), 1.27 (d, *J* = 7.3 Hz, 3H), 1.20 (d, *J* = 6.5 Hz, 3H), 0.99 (dd, *J* = 9.6, 6.9 Hz, 6H). **^{13}C-NMR** (126 MHz, CD_3OD): δ (ppm) = 175.3, 169.7, 169.2, 156.8, 151.0, 147.7, 139.5, 136.1, 124.7, 80.9, 78.5, 73.0, 70.5, 56.7, 53.3, 45.3, 36.4, 31.2, 28.7, 18.9, 17.6, 14.9, 8.7. **IR** (thin film): υ (cm^{-1}) = 3357, 2969, 2924, 2851, 1744, 1716, 1670, 1521, 1496, 1456, 1390, 1367, 1290, 1246, 1163, 1067, 1044, 1024, 989, 863, 713. **HR-MS** (ESI): Calcd. for $C_{25}H_{38}N_3O_8$ $[M+H]^+$ 508.2653 *m/z*; found 508.2655 *m/z*.

(R)-2-(((2R,3S)-4-(Benzyloxy)-3-((tert-butoxycarbonyl)amino)-4-oxobutan-2-yl)oxy)-1-cyclohexyl-2-oxoethyl (2S,3R,4S)-4-(dibenzylamino)-3-hydroxy-2-methyl-5-(pyridin-3-yl)pentanoate (38)



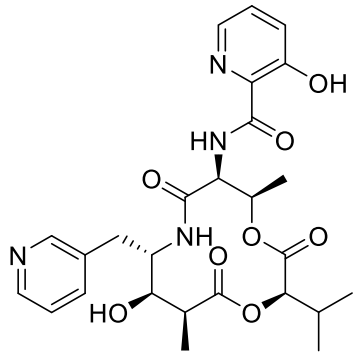
38 was prepared from bis-*epi*-**24** and **8c** according to the procedure described for **25a**. FC (hexane/EtOAc 3:1) provided **38** (39.9 mg, 51%) as a colorless oil. **TLC** (SiO_2, hexane/EtOAc 3:1, UV): R_f = 0.15. **$[\alpha]_D^{20}$** = +12.5° (*c* 0.64, $CHCl_3$). **^{1}H-NMR** (400 MHz, $CDCl_3$): δ (ppm) = 8.49 (dd, *J* = 4.9, 1.6 Hz, 1H), 8.42 (d, *J* = 2.2 Hz, 1H), 7.43 – 7.30 (m, 6H), 7.24 – 7.14 (m, 7H), 7.14 – 7.06 (m, 4H), 5.54 (qd, *J* = 6.4, 2.4 Hz, 1H), 5.47 (d, *J* = 9.7 Hz, 1H), 5.18 (d, *J* = 12.3 Hz, 1H), 5.09 (d, *J* = 12.2 Hz, 1H), 4.84 (d, *J* = 4.0 Hz, 1H), 4.53 (dd, *J* = 9.7, 2.4 Hz, 1H), 4.27 – 4.19 (m, 1H), 3.97 (d, *J* = 14.2 Hz, 2H), 3.59 (d, *J* = 14.2 Hz, 2H), 3.52 (br. s, 1H), 3.10 (dd, *J* = 14.2, 9.2 Hz, 1H), 3.00 – 2.93 (m, 1H), 2.87 (dd, *J* = 14.3, 4.0 Hz, 1H), 2.65 (p, *J* = 7.2 Hz, 1H), 1.94 – 1.59 (m, 5H), 1.52 (dd, *J* = 12.2, 3.3 Hz, 1H), 1.45 (s, 9H), 1.33 (d, *J* = 6.4 Hz, 3H), 1.31 – 1.10 (m, 5H), 1.05 (d, *J* = 7.1 Hz, 3H). **^{13}C-NMR** (101 MHz, $CDCl_3$): δ (ppm) = 174.2, 169.8, 169.3, 156.0, 151.1, 147.3, 139.8, 137.1, 136.5, 135.0, 128.8, 128.7, 128.6, 128.4, 128.3, 127.0, 123.1, 80.5, 76.2, 72.9, 72.1, 67.8, 60.3, 57.2, 54.5, 45.2, 39.5, 29.3, 29.0, 28.4, 27.5, 26.1, 26.0, 25.9, 17.0, 14.8. **IR** (thin film): υ (cm^{-1}) = 2977, 2929, 2854, 1742, 1495, 1454, 1424, 1381, 1367, 1314, 1258, 1209, 1164, 1085, 1065, 992, 740, 698 cm^{-1}. **HR-MS** (ESI): Calcd. for $C_{49}H_{62}N_3O_9$ $[M+H]^+$ 836.4481 *m/z*; found 836.4481 *m/z*.

tert-Butyl ((2R,5R,6S,9S,10R,11S)-2-cyclohexyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)carbamate (38A)



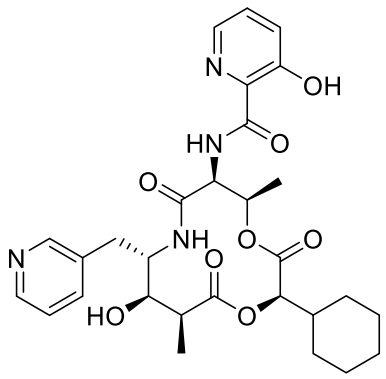
38A was prepared from **38** according to the procedure described for **26a**. FC (EtOAc) provided **38A** (9.0 mg, 42% over 2 steps) as a colorless film. **TLC** (SiO_2, EtOAc, UV, $KMnO_4$): R_f = 0.20. **[α]$_D^{20}$** = +29.6° (*c* 0.98, $CHCl_3$). **^{1}H-NMR** (400 MHz, CD_3OD): δ (ppm) = 8.46 – 8.39 (m, 1H), 8.35 (dd, *J* = 5.0, 1.6 Hz, 1H), 7.70 (dt, *J* = 8.0, 1.9 Hz, 1H), 7.30 (dd, *J* = 7.8, 4.9 Hz, 1H), 5.19 – 5.08 (m, 1H), 4.82 (br. s, 1H), 4.46 – 4.33 (m, 1H), 4.08 (d, *J* = 5.5 Hz, 1H), 4.01 (br. s, 1H), 3.29 (br. s, 1H), 2.92 – 2.84 (m, 1H), 2.75 (br. s, 1H), 1.98 – 1.88 (m, 1H), 1.81 – 1.74 (m, 2H), 1.72 – 1.63 (m, 3H), 1.45 (s, 9H), 1.38 – 1.10 (m, 11H). **^{13}C-NMR** (101 MHz, CD_3OD): δ (ppm) = 175.3, 170.0, 169.3, 156.9, 150.8, 147.5, 139.9, 136.5, 124.9, 81.0, 78.3, 73.1, 70.7, 57.0, 53.5, 45.4, 40.6, 36.2, 30.1, 28.9, 28.7, 27.1, 27.1, 26.9, 15.0, 8.7. **IR** (thin film): υ (cm^{-1}) = 3429, 3359, 2979, 2928, 2856, 1745, 1719, 1675, 1497, 1451, 1433, 1391, 1367, 1318, 1291, 1251, 1166, 1106, 1086, 1047, 1025. **HR-MS** (ESI): Calcd. for $C_{28}H_{42}N_3O_8$ $[M+H]^+$ 548.2966 *m/z*; found 548.2965 *m/z*.

3-Hydroxy-N-((2R,5R,6S,9S,10R,11S)-10-hydroxy-2-isopropyl-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)picolinamide (39)



39 was prepared from **37A** according to the procedure described for **28a**. FC (DCM/MeOH 9:1) provided **39** (10 mg) as a yellow oil. Further purification by RP-HPLC (20 to 65% MeCN/0.1% TFA in 0.1% aq. TFA) gave **39** (3.30 mg, 37% over 2 steps) as a white lyophilisate. **TLC** (SiO_2, DCM/MeOH, 9:1, UV) R_f = 0.40. **[α]D^{20}** = –28.2° (*c* 0.07, MeOH). **^{1}H-NMR** (500 MHz, $CDCl_3$): δ (ppm) = 11.62 (s, 1H), 8.50 (d, *J* = 3.6 Hz, 1H), 8.48 (s, 1H), 8.32 (br. s, 1H), 8.09 – 8.06 (m, 1H), 7.54 (d, *J* = 7.8 Hz, 1H), 7.39 – 7.32 (m, 1H), 7.32 – 7.28 (m, 1H), 7.18 – 7.12 (m, 1H), 6.09 (br. s, 1H), 5.52 – 5.44 (m, 1H), 4.97 (d, *J* = 3.9 Hz, 1H), 4.55 (dd, *J* = 8.0, 5.6 Hz, 1H), 4.50 (br. s, 1H), 4.12 (br. s, 1H), 3.07 (dd, *J* = 14.8, 3.7 Hz, 1H), 3.03 – 2.91 (m, 2H), 2.40 – 2.32 (m, 1H), 1.36 (d, *J* = 7.2 Hz, 3H), 1.31 (d, *J* = 6.6 Hz, 3H), 1.03 (d, *J* = 6.9 Hz, 3H), 1.00 (d, *J* = 6.8 Hz, 3H). **^{13}C-NMR** (126 MHz, $CDCl_3$): δ (ppm) = 176.3, 168.4, 167.2, 166.9, 157.9, 150.5, 147.8, 140.1, 137.3, 133.1, 130.9, 129.2, 126.2, 123.7, 77.8, 68.4, 54.0, 43.7 (HMBC), 34.8, 30.3, 18.9, 17.1, 14.5 **IR** (thin film): υ (cm^{-1}) = 3363, 2965, 2930, 2855, 1746, 1726, 1651, 1598, 1521, 1449, 1389, 1294, 1252, 1045, 997, 810, 716, 658. **HR-MS** (ESI): Calcd. for $C_{26}H_{33}N_4O_8$ $[M+H]^+$ 529.2293 *m/z*; found 529.2290 *m/z*.

N-((2R,5R,6S,9S,10R,11S)-2-Cyclohexyl-10-hydroxy-5,11-dimethyl-3,7,12-trioxo-9-(pyridin-3-ylmethyl)-1,4-dioxa-8-azacyclododecan-6-yl)-3-hydroxypicolinamide (40)



40 was prepared from **38A** according to the procedure described for **28a**. FC (DCM/MeOH 95:5) provided **40** (6.2 mg) as a colorless oil. Further purification by RP-HPLC (20 to 60% MeCN/0.1% TFA in 0.1% aq. TFA) provided **40** (3.7 mg, 51% over 2 steps) as a white lyophilisate. **TLC** (SiO_2, DCM/MeOH 95:5, UV): R_f = 0.16. **$[\alpha]_D^{20}$** = –8.1° (*c* 0.37, MeOH). **^{1}H-NMR** (500 MHz, CD_3OD): δ (ppm) = 8.70 (d, *J* = 2.1 Hz, 1H), 8.47 – 8.41 (m, 1H), 8.40 – 8.35 (m, 1H), 8.15 (dd, *J* = 4.4, 1.4 Hz, 1H), 7.72 – 7.59 (m, 2H), 7.52 (dd, *J* = 8.5, 4.4 Hz, 1H), 7.41 (dd, *J* = 8.5, 1.3 Hz, 1H), 5.35 (p, *J* = 6.4 Hz, 1H), 4.96 – 4.90 (br. s, 1 H), 4.51 (d, *J* = 6.0 Hz, 2H), 4.15 (br. s, 1H), 3.62 – 3.53 (m, 1H), 2.98 – 2.82 (m, 2H), 2.00 – 1.92 (m, 1H), 1.83 – 1.76 (m, 2H), 1.74 – 1.65 (m, 3H), 1.39 – 1.13 (m, 11H). **^{13}C-NMR** (126 MHz, CD_3OD): δ (ppm) = 175.1, 169.4, 169.1, 159.1, 148.1, 144.4, 141.2, 141.2, 140.7, 131.7, 130.8, 127.6, 127.4, 78.3, 73.2, 69.7, 55.0, 53.1, 45.4, 40.8, 36.9, 30.1, 28.9, 27.1, 27.0, 15.1, 8.6. **IR** (thin film): υ (cm^{-1}) = 3366, 2932, 1733, 1675, 1559, 1523, 1452, 1262, 1200, 1140, 1050, 885. **HR-MS** (ESI): Calcd for $C_{29}H_{37}N_4O_8$ $[M+H]^+$ 569.2606 *m/z*; found 569.2605 *m/z*.

MIC determinations. MIC's were determined in a resazurin reduction microplate assay (REMA)[40] as described previously[9]. Briefly, frozen stocks of H37Rv, H37Rv:pMV261, and H37Rv:pMVInhA (all frozen at OD_{600} of 1) were diluted 1 in 2500 (v/v) into Middlebrook 7H9 media supplemented 10% ADC, 0.2 % glycerol and 0.05 % Tween 80 (25 μg/mL kanamycin was also used for maintenance of the pMV261 and pMVInhA contain strains). Diluted bacterial suspensions were added to a 96 well plate (100 μL) in the presence of serial dilutions of the compounds of interest (maximum DMSO concentration of 1 %). Plates were sealed with a PCR film and incubated (7 days, 37 °C). Bacterial viability was determined using the redox indicator, resazurin (0.025% w/v, 16 hr), and measured by fluorescence (Ex: 560 nm, Em: 590 nm) using a Tecan Infinite M200 microplate reader. The minimal inhibitory concentration (MIC) was determined as the lowest concentration of compound where the resazurin fluorescence was at background level (equal to no bacteria control). Experiments were carried out at least in duplicate and average values are reported.

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Author Contributions

K.-H.A., B.S., M.K., O.P.H., and P.E. designed the research; M.K., B.S., O.P.H., P.E., and R.C.H. designed experiments; B.S., M.K., O.P.H., P.E., S.H., G.v.C., R.C.H., and A.V. carried out experiments; S.T.C. and K.H.A. provided supervision. M.K. and K.-H.A. wrote the article.

M.K., P.E., B.S., and O.P.H. contributed equally to this work.

Funding Sources

This work was supported by the European Community Framework Programme 7, grant agreement no. 260872 (O.P.H, R.C.H, S.T.C, K.H.A) and by the Swiss National Science Foundation (M.K., project 200021_149253; P.E., project 200020_175744). Financial support was also provided by ETH Zurich through base funding to K.H.A. (B.S., S.H.).

ACKNOWLEDGMENT

We gratefully acknowledge financial support by the European Community Framework Programme 7 (grant agreement no. 260872) and the Swiss National Science Foundation (projects 200021_149253 and 200020_175744). We are indebted to Dr. Bernhard Pfeiffer (ETHZ) for NMR support and to Dr. Xiangyang Zhang, Louis Bertschi, Rolf Häfliger, and Oswald Greter (ETHZ) for HR-MS spectra. Kurt Hauenstein is acknowledged for crucial technical support. We are grateful to Nils Trapp for providing the X-ray crystal structures of cpds. *ent*-**23** and **26c.**

ABBREVIATIONS

CSA, camphorsulfonic acid; DCC, *N*,*N'*-dicyclohexylcarbodiimide; DCM, dichloromethane; DIPEA, *N*,*N*-diisopropylethylamine; DMAP, 4-dimethylaminopyridine; DMF, *N*,*N*-dimethylformamide; DMSO, dimethylsulfoxide; HATU, *O*-(7-azabenzotriazol-1-yl)-*N*,*N*,*N*′,*N*′-tetramethyluronium hexafluorophosphate; *c*-Hex_2BOTf, dicyclohexylboron trifluoromethanesulfonate; HPLC, high performance liquid chromatography; 3-HPA, 3-hydroxypicolinic acid; IC_{50}, half maximum inhibitory concentration; L-(+)-DET, L-(+)-diethyl tartrate; MeCN, acetonitrile; MIC, minimum inhibitory concentration; *Mtb*, *Mycobacterium tuberculosis*; Pd/C, palladium on charcoal; *p*-TsOH, *para*-toluenesulfonic acid; REMA, resazurin reduction microplate assay; RP, reversed phase; rt, room temperature; TBSCl, *tert*-butyldimethylsilyl chloride; TBSOTf, trifluoromethanesulfonic acid *tert*-butyldimethylsilyl ester; TFA, trifluoroacetic acid; THF, tetrahydrofuran; and TTIP, titanium isopropoxide.

ASSOCIATED CONTENT

Supporting Information. Experimental protocols for the synthesis of acid **29** (incl. copies of ^{1}H- and ^{13}C-NMR spectra of acid **29** and all intermediates); ^{1}H- and ^{13}C-NMR spectra of final products and intermediates; HPLC traces for all final products, except **29e**; MIC values of cpds. **1**, **2**, **28c**, **28e**, and **35**, in the absence of Tween80; information on X-ray crystal structures of bis-*epi*-**23** and **26c**. CSV file with molecular formula strings of all new compounds and associated biological data (where applicable). The following files are available free of charge.

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