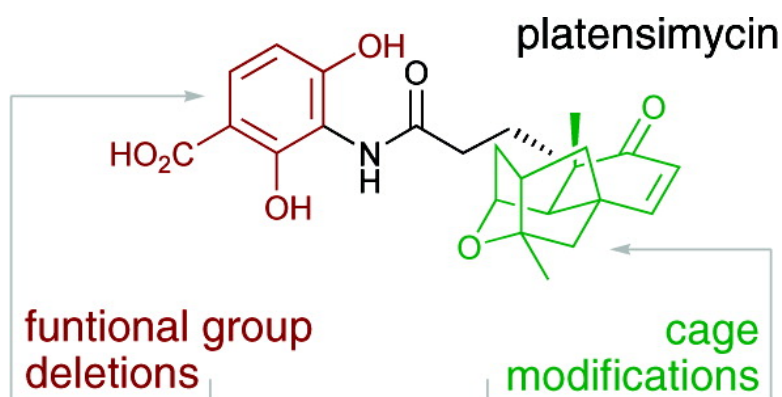Article

Design, Synthesis, and Biological Evaluation of Platensimycin Analogues with Varying Degrees of Molecular Complexity

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Design, Synthesis, and Biological Evaluation of Platensimycin Analogues with Varying Degrees of Molecular Complexity

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Abstract: The molecular design, chemical synthesis, and biological evaluation of two distinct series of platensimycin analogues with varying degrees of complexity are described. The first series of compounds probes the biological importance of the benzoic acid subunit of the molecule, while the second series explores the tetracyclic cage domain. The biological data obtained reveal that, while the substituted benzoic acid domain of platensimycin is a highly conserved structural motif within the active compounds with strict functional group requirements, the cage domain of the molecule can tolerate considerable structural modifications without losing biological action. These findings refine our present understanding of the platensimycin pharmacophore and establish certain structure−activity relationships from which the next generation of designed analogues of this new antibiotic may emerge.

Introduction

The disclosure of the novel antibiotic platensimycin (**1**, Figure 1)[1] by the Merck group of Wang, Soisson, and Singh et al. has attracted considerable attention from within the scientific community, as manifested by a series of publications describing its chemical synthesis.[2] The intense interest in this natural product can be attributed to its unique molecular architecture and extraordinary antibacterial activity against Gram-positive bacteria, including multiple drug-resistant strains such as methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant *Enterococcus faecalis* (VREF). Platensimycin
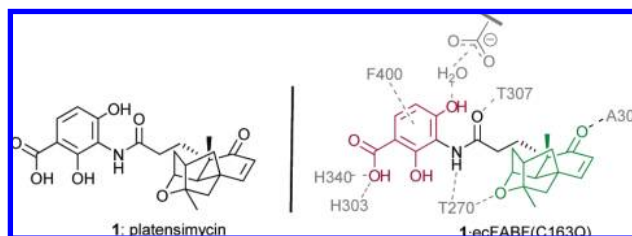
Figure 1. Molecular structure of platensimycin (**1**) and interactions of platensimycin with its mutated target enzyme ecFABF(C163Q) [**1**·ecFABF-(C163Q)].

exhibits its impressive antibacterial activity through a novel mode of action that involves selective interference with the catalytic machinery responsible for the biosynthesis of bacterial fatty acids by selective inhibition of the elongation-condensing enzyme FabF.[1a] X-ray crystallographic analysis of platensimycin in complex with a mutant version of the FabF enzyme [ecFABF(C163Q)] designed to mimic the acyl−enzyme intermediate reveals two distinct binding domains (Figure 1): (I) the highly polar benzoic acid unit which docks to the malonate pocket of the condensing enzyme, and (II) the lipophilic cage that is positioned in the mouth of the enzyme. Within binding domain I, the carboxylate of the benzoic acid interacts with the active-site histidine residues (H303 and H340), an interaction that is likely to have some ionic character. Moreover, the aromatic ring of the benzoic acid is involved in an edge-to-face π-interaction with F400, which is oriented in the position it is believed to occupy in the acyl−enzyme intermediate. Within binding domain II, the hydrophilic ketolide motif, which is partially exposed to solvent, has significant van der Waals interactions with the protein surface, covering some 122 Å^2 of

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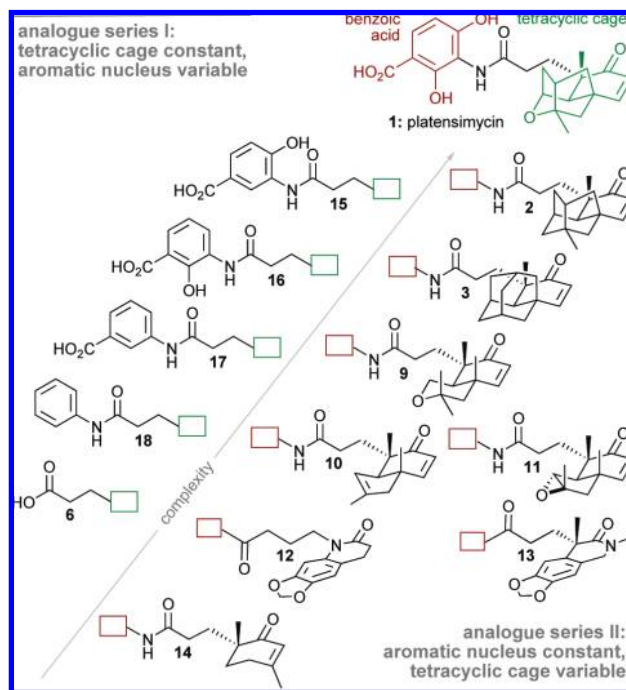‖ Now at the University at Cambridge, United Kingdom.

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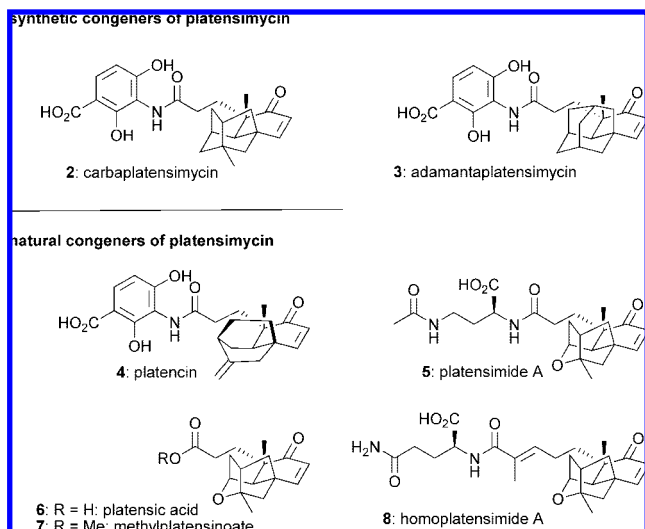
Figure 2. Structures of synthetic (**2** and **3**) and natural (**4**−**8**) platensimycin congeners.

Figure 3. Platensimycin (**1**) and surrogates (**2**, **3**, **6**, **9**−**18**) incorporating varying degrees of molecular complexity.

the protein surface. The cage also makes hydrogen bonds involving its ether and carbonyl oxygen atoms with T270 and A309, respectively. The amide linker, connecting the two binding domains, is involved in two hydrogen bonds to the T307 side chain and the amide backbone at T270.

Despite its possessing this outstanding biological profile, the full medicinal potential of platensimycin as a breakthrough anti-infective agent has not yet been realized, possibly due to a non-ideal pharmacokinetic profile.[3] In order to translate this exceptional *in vitro* potency to *in vivo* systems, fine-tuning of the platensimycin lead structure may be necessary, a task that will require an in-depth understanding of its pharmacophore. Toward this goal, we have recently investigated the bioactive platensimycin isosteres carba-[4] and adamantaplatensimycin[5] (**2** and **3**, Figure 2), which we prepared through total synthesis. Further indications regarding the essential structural features necessary for platensimycin-like antibacterial activity have been reported recently by the Merck team of Singh et al., who isolated and biologically evaluated a number of naturally occurring congeners of platensimycin, namely platencin (**4**),[6] platensimide A (**5**),[7a] platensic acid (**6**),[7a] methylplatensinoate (**7**),[7a] and homoplatensimide A (**8**).[7b] Moreover, Singh et al. reported the semi-synthesis of various other platensimycin derivatives; however, the detailed biological data for these compounds have not been disclosed.[7c] We now wish to report the design, synthesis, and biological evaluation of several platensimycin analogues incor-

porating varying degrees of molecular complexity. This detailed investigation refines our present understanding of the contributory role played by each characteristic domain within platensimycin and establishes certain structure−activity relationships within the platensimycin structural motif from which the next generation of designed analogues may emerge.

Results and Discussion

1. Design of Platensimycin Analogues. As shown in Figure 1, the molecular structure of platensimycin (**1**) comprises two structurally distinct domains, a substituted benzoic acid moiety and a lipophilic cage, that are connected through an amide linker. In order to determine the importance of the polar aromatic subunit and its substitution pattern for biological activity, we aimed to synthesize members of analogue series I (Figure 3). Within this series, the tetracyclic cage domain remains intact while increasingly complex aromatic surrogates (compounds **6**, **18** to **15**) probe the specific interactions of this domain with the targeted enzyme through single, double, and triple functional group deletions.

In analogue series II, the aromatic nucleus is kept constant while the tetracyclic region is probed by appending cage mimics that approach in complexity and similarity the structure of the natural product (compounds **14** to **9** and **3** to **2**, Figure 3). We anticipated structures within analogue series I to be more sensitive to changes than structures within analogue series II, owing to the higher level of interactions of the aromatic substituents with the receptor, as opposed to those of the cage domain.[1a]

2. Synthesis of Analogue Series I. The first member of analogue series I (Figure 3), platensic acid (**6**), is a key intermediate in our asymmetric synthesis of platensimycin[2b] and was obtained enantiomerically enriched from an (*S,S*)-pseu-doephedrine amide, as described in our previous communication.[2b] Access to aniline derivatives **15**−**18** relied on late-stage divergence from this carboxylic acid (Scheme 1). Thus,

(3) The high *in vitro* potency of platensimycin could only be translated to *in vivo* systems if it was administered by continuous infusion; see refs 1a, 1c, and 7.

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Scheme 1. Synthesis of Aniline Analogues **15–18**[a]
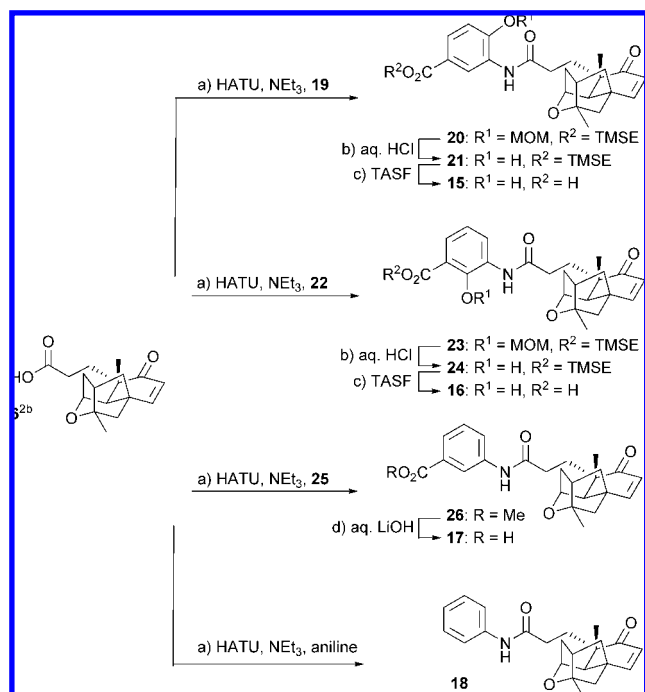
[a] Reagents and conditions: (a) HATU (4.0 equiv), NEt$_3$ (5.0 equiv), **19** (for **20**) or **22** (for **23**) or **25** (for **26**) or aniline (for **18**) (4.0 equiv), DMF, 25 °C, 12 h, 82% (for **20**), 90% (for **23**), 83% (for **26**), or 86% (for **18**); (b) aq. HCl (2.0 M, 30 equiv), THF, 45 °C, 6 h (for **21**) or 24 h (for **24**), 83% (for **21**) or 90% (for **24**); (c) TASF (5.0 equiv), DMF, 40 °C, 24 h (for **15**) or 2 h (for **16**), 94% (for **15**), 99% (for **16**); (d) aq. LiOH (2.0 M, 30 equiv), THF, 45 °C, 4 h, 69%. Abbreviations: DMF = *N,N*-dimethylformamide; Et = ethyl; Me = methyl; MOM = methoxymethyl; HATU = *O*-(7-azabenzotriazol-1-yl)-*N,N,N′,N′*-tetramethyluronium hexafluorophosphate; TASF = tris(dimethylamino)sulfonium difluorotrimethylsilicate; THF = tetrahydrofuran.

Scheme 2. Construction of Aromatic Fragments **19**, **22**, and **25**[a]
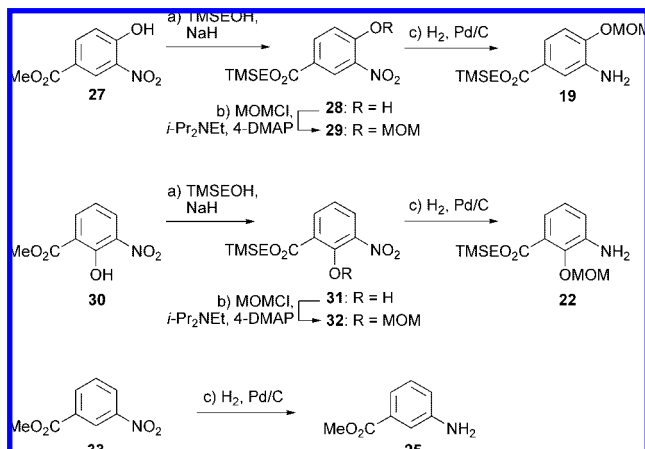
[a] Reagents and conditions: (a) TMSE-OH (38 equiv), NaH (60% dispersion in mineral oil, 19 equiv), benzene, 10 min, 25 °C, 95% (for **28**) or 99% (for **31**); (b) MOMCl (1.2 equiv), *i*-Pr$_2$NEt (1.2 equiv), 4-DMAP (0.1 equiv), 25 °C, 30 min, 91% (for **29**), 96% (for **32**); (c) H$_2$ (1.1 atm), 10% Pd/C (0.2 equiv), MeOH, 25 °C, 12 h (for **19**) or 1 h (for **22** and **25**), 85% (for **19**), 90% (for **22**), or 92% (for **25**). Abbreviations: 4-DMAP = 4-dimethylaminopyridine; *i*-Pr = isopropyl; TMSE = 2-(trimethylsilyl)ethyl.

HATU-mediated amide coupling of **6** with amines **19**, **22**, and **25** (see Scheme 2), as well as aniline itself, gave the corresponding amides **20**, **23**, **26**, and **18** in 82, 90, 83, and 86% yield, respectively. As required, MOM-protecting group

Scheme 3. Synthesis of Cage Analogue **14**[a]
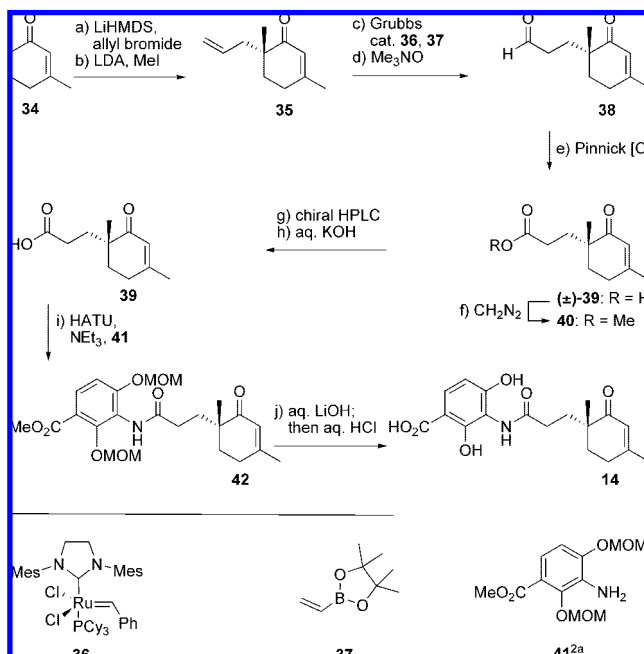
[a] Reagents and conditions: (a) LiHMDS (1.0 M in toluene, 1.2 equiv), THF, −78 °C, 30 min; then allyl bromide (1.1 equiv), THF, −78 → 0 °C, 1 h, 99%; (b) LDA (1.0 M in THF, 1.1 equiv), THF, −78 °C, 30 min; then MeI (1.1 equiv), THF, −78 → 25 °C, 12 h, 95%; (c) **37** (2.0 equiv), Grubbs catalyst **36** (0.04 equiv), benzene, reflux, 30 min, 85% (*E:Z* ca. 5:1); (d) Me$_3$NO (5.0 equiv), THF, 65 °C, 1 h, 92%; (e) NaClO$_2$ (2.0 equiv), 2-methyl-2-butene (11 equiv), NaH$_2$PO$_4$ (5.0 equiv), *t*-BuOH:water (4:1), 25 °C, 30 min, 99%; (f) CH$_2$N$_2$ (excess), CH$_2$Cl$_2$, 0 °C, 10 min, 100%; (g) chiralcel OD-H column, 4% isopropyl alcohol/hexanes; (h) aq. KOH (2.0 M, 30 equiv), MeOH, 25 °C, 12 h, 83%; (i) **41** (1.2 equiv), HATU (1.2 equiv), NEt$_3$ (6.0 equiv), DMF, 25 °C, 12 h, 65%; (j) aq. LiOH (2.0 M, 30 equiv), THF, 45 °C, 3 h; then aq. HCl (2.0 M, 60 equiv), THF, 45 °C, 12 h, 81%. Abbreviations: *t*-Bu = *tert*-butyl; Cy = cyclohexyl; LDA = lithium diisopropylamide; Mes = mesityl; Ph = phenyl; HMDS = hexamethyldisilazide.

removal (aqueous HCl, THF, 45 °C) followed by deprotection of the ester functionality using fluoride (TASF, DMF, 40 °C) or basic conditions (aqueous LiOH, THF, 45 °C) revealed platensimycin derivatives **15–18** in excellent yields, as indicated in Scheme 1.

The required aniline fragments **19**, **22**, and **25** were swiftly prepared from commercially available methyl esters **27**, **30**, and **33**, as depicted in Scheme 2. Thus, starting from **27** and **30**, ester exchange under basic conditions (TMSE-OH, NaH, benzene, 25 °C) gave phenols **28** (95% yield) and **31** (99% yield), which were MOM-protected (MOMCl, *i*-Pr$_2$NEt, 4-D-MAP) to furnish fully protected intermediates **29** (91% yield) and **32** (96% yield). Catalytic hydrogenation [H$_2$ (1.1 atm), 10% Pd/C, MeOH, 25 °C] of the nitro functionality proceeded smoothly to provide the requisite aniline fragments **19**, **22**, and **25** in 85, 90, and 92% yield, respectively.

3. Synthesis of Analogue Series II. 3.1. Cyclohexenenone Analogue 14. The first member of analogue series II, cyclohexenone **14**, was conveniently prepared from enone **34** (Scheme 3). In analogy to our previously reported syntheses of platensimycin,[2a,b] sequential double alkylation [(a) LiHMDS, allyl bromide, 99% yield; (b) LDA, MeI, 95% yield] of **34** gave diene **35**. Chemoselective cross-metathesis using excess vinyl

boronate **37** and Grubbs II catalyst (**36**)[8,9] then gave the corresponding vinyl pinacol boronates as a ca. 5:1 mixture of *E* and *Z* isomers (85% combined yield). Subsequent oxidative cleavage of this mixture of boronates with Me$_3$NO[9a,10] (THF, 65 °C) furnished aldehyde **38** (92% yield), which was converted to racemic carboxylic acid (±)-**39** by a Pinnick oxidation[11] (NaClO$_2$, 99% yield). Formation of the corresponding methyl ester (**40**) with diazomethane (100% yield) allowed for the separation of this racemate by HPLC using a chiral column. Each of the enantiomers so obtained was saponified with aqueous KOH to reveal acid **39** (and its enantiomer *ent*-**39**) in 83% overall yield and >99% ee. Since we did not know which enantiomer corresponded to the natural configuration of platensimycin, we carried both **39** and *ent*-**39** to the final product, hoping to evaluate both isomers for biological activity. Finally, amide coupling (HATU, DMF, 25 °C) of **39** (and *ent*-**39**) with the required, fully protected aniline fragment **41**[2a,12] gave advanced intermediate **42** (and *ent*-**42**) (65% yield), which upon deprotection (aqueous LiOH, 45 °C; then HCl, 45 °C, 81% overall yield) led to the desired platensimycin cage analogue **14** (and *ent*-**14**) (Scheme 3).

3.2. Aromatic Analogue 12. Synthesis of achiral aromatic platensimycin derivative **12** commenced from commercially available 6-nitropiperonal (**43**) (Scheme 4), which was subjected to a Wittig olefination with phosphorane **44** to give enoates **45** (77% yield, *E*:*Z* ca. 3:2). Catalytic hydrogenation [H$_2$ (1.1 atm), 10% Pd/C] followed by base-promoted (aqueous KOH) cyclization furnished lactam **46** in 83% overall yield, which was *N*-alkylated with commercially available methyl 4-bromobutyrate (**47**) (NaH, DMF) to give intermediate **48** in 86% yield. Basic hydrolysis of methyl ester **48** (aqueous LiOH, THF, 45 °C, 94% yield) revealed carboxylic acid **49**, which was coupled with known aniline **41**[2a] (HATU, NEt$_3$, DMF) to give amide **50** in 74% yield. As described previously,[2a] basic hydrolysis of the methyl ester in **50** (aqueous LiOH, THF, 45 °C), followed by removal of both MOM protecting groups (aqueous HCl, THF, 45 °C), gave derivative **12** in 81% yield.

3.3. Aromatic Analogue 13. Aromatic platensimycin derivative **13** was accessed from commercially available bromopiperonal (**51**) (Scheme 5). Thus, Wittig reaction with the *in situ*-generated ylide of (methoxymethyl)triphenyl phosphonium chloride (*n*-BuLi, THF, 0 °C), followed by acid-promoted cleavage of the resulting methyl vinyl ether (aqueous HCl, acetone, reflux), gave one-carbon homologated aldehyde **52** in 58% overall yield. The corresponding methyl ester **53** was obtained via consecutive Pinnick oxidation[11] (NaClO$_2$) and acidic esterification (H$_2$SO$_4$, MeOH, reflux, 53% over two steps).

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(12) In the syntheses of cage analogues **10** and **11**, we used the second-generation aniline fragment **71** (developed during our total synthesis of platencin),[18] as it allowed for mild unmasking of the substituted benzoic acid domain (see Scheme 6). For a protecting-group-free approach to this key amide coupling, see: Hayashida, J.; Rawal, V. H. *Angew. Chem., Int. Ed.* **2008**, *47*, 4373. We expect that most, if not all, amide couplings descibed in this article (and carried out before these innovations occurred) could be carried out with minimal or no protecting groups on the aniline fragments.

Scheme 4. Synthesis of Cage Analogue **12**[a]
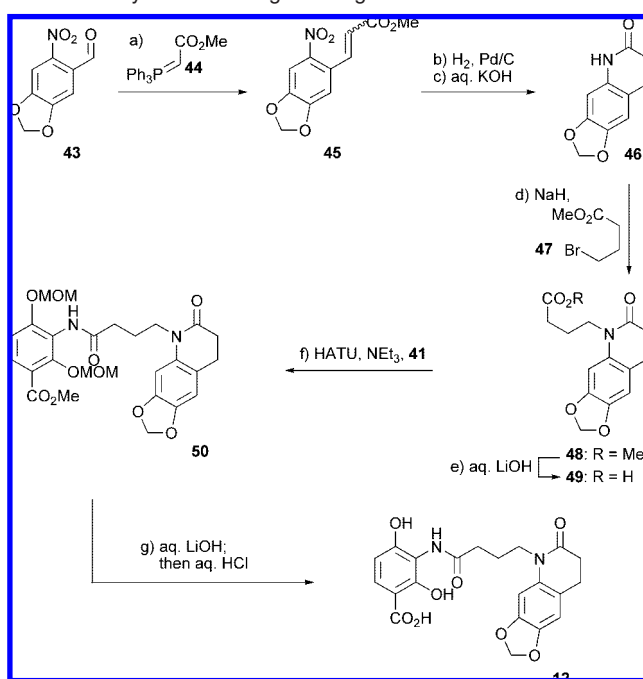
[a] Reagents and conditions: (a) **44** (1.5 equiv), CH$_2$Cl$_2$, 25 °C, 12 h, 77% (*E*:*Z* ca. 3:2); (b) H$_2$ (1.1 atm), 10% Pd/C (0.1 equiv), MeOH:EtOAc (1:1), 25 °C, 1 h; (c) aq. KOH (2.0 M, 30 equiv), MeOH, 83% over two steps; (d) NaH (1.5 equiv), DMF, 0 °C, 1 h; then **47** (3.0 equiv), 25 °C, 12 h, 86%; (e) aq. LiOH (1.0 M, 5.9 equiv), THF, 45 °C, 94%; (f) **41** (2.0 equiv), HATU (4.0 equiv), NEt$_3$ (6.0 equiv), DMF, 25 °C, 12 h, 74%; (g) aq. LiOH (2.0 M, 15 equiv), THF, 45 °C, 6 h; then aq. HCl (2.0 M, 30 equiv), THF, 45 °C, 12 h, 81%. Abbreviations: Ac = acetyl.

Methylation of the potassium enolate (KHMDS, MeI, 92% yield) gave **54**, which underwent an Ullmann-type reaction[13] with CuCN that installed the nitrile functionality on the aromatic ring (DMF, reflux). Catalytic hydrogenation [H$_2$ (1.1 atm), 10% Pd/C, MeOH] of the resulting nitrile functionality then afforded the corresponding primary amine, which spontaneously cyclized to give lactam **55** (39% overall yield). *N*-Methylation of **55** (LiHMDS, MeI, 84% yield) furnished advanced intermediate **56**, which was then alkylated with alkyl iodide **57** (prepared in one step from acrolein via a literature protocol)[14] to give dioxolane **58** (74% yield). Conversion of the latter compound to carboxylic acid **59** (aqueous HCl then NaClO$_2$, 80% overall yield), followed by HATU-mediated amide coupling with aniline **41**,[2a] led to amide **60** (87% yield), which was swiftly deprotected using the above-described protocol[2a] (aqueous LiOH, THF, 45 °C; then aqueous HCl, THF) to afford targeted analogue **13** as a racemate in 76% overall yield.

3.4. Cyclopentene and Epoxide Analogues 10 and 11. The chemical synthesis of cage surrogates **10** and **11** commenced from commercially available enone **61** (also available from 1,3-cyclohexanedione through literature procedures).[15] Thus, double alkylation [(a) LDA, 3-bromo-2-methylpropene, 88% yield; (b) LDA, MeI, 84% yield] led to α-substituted enone **62** which, upon one-pot reduction and acid-catalyzed hydrolysis/elimination (DIBAL-H then aqueous HCl), furnished rearranged enone **63** in 95% overall yield (Scheme 6). Dienone formation was

(13) A modification of the procedure reported in the following was used: Le, T. N.; Cho, W.-J. *Chem. Pharm. Bull.* **2006**, *54*, 476.

(14) Larson, G. L.; Klesse, R. *J. Org. Chem.* **1985**, *50*, 3627.

(15) House, H. O.; Gannon, W. F. *Org. Synth. Coll. Vol. V* **1973**, 539.

Scheme 5. Synthesis of Cage Analogue **13**[a]

Scheme 6. Synthesis of Cage Analogues **10** and **11**[a]

[a] Reagents and conditions: (a) Ph₃PCH₂OMeCl (1.3 equiv), *n*-BuLi (2.5 M in hexanes, 1.2 equiv), THF, 0 °C, 30 min; (b) aq. HCl (1.0 M, 2.7 equiv), acetone, reflux, 3 h, 58% over two steps; (c) 2-methyl-2-butene (20 equiv), NaH₂PO₄ (3.0 equiv), NaClO₂ (2.8 equiv), THF:water (9:1), 25 °C, 30 min; (d) 10% aq. H₂SO₄ (0.2 equiv), MeOH, reflux, 12 h, 53% over two steps; (e) KHMDS (0.5 M in toluene, 1.1 equiv), THF, −78 °C, 30 min; then MeI (3.0 equiv), −78 → 25 °C, 12 h, 92%; (f) CuCN (1.1 equiv), DMF, reflux, 3 h; (g) H₂ (1.1 atm), 10% Pd/C (0.04 equiv), MeOH, 25 °C, 1 h, 39% over two steps; (h) LiHMDS (1.0 M in toluene, 1.1 equiv), THF, −78 °C, 30 min; then MeI (3.0 equiv), −78 → 25 °C, 12 h, 84%; (i) KHMDS (0.5 M in toluene, 2.0 equiv), −78 °C, 30 min; then **57** (5.0 equiv), −78 → 25 °C, 12 h, 74%; (j) aq. HCl (1.0 M, 14 equiv), THF, 45 °C, 5 h; (k) NaClO₂ (2.0 equiv), 2-methyl-2-butene (30 equiv), NaH₂PO₄ (3.0 equiv), *t*-BuOH:water (5:1), 25 °C, 30 min, 80% over two steps; (l) **41** (2.3 equiv), HATU (4.6 equiv), NEt₃ (7.0 equiv), DMF, 25 °C, 12 h, 87%; (m) aq. LiOH (2.0 M, 25 equiv), THF, 45 °C, 3 h; then aq. HCl (2.0 M, 50 equiv), THF, 45 °C, 12 h, 76%.

achieved through the formation of the intermediate silyl enol ether (NEt₃, TMSOTf), which was cleanly oxidized with IBX·MPO[16] to give **64** (92% overall yield). Subsequent tandem Michael addition−enolate alkylation (CuBr·Me₂S, vinyl magnesium bromide; then MeI, 75% yield) led to triene **65**, which was subjected to ring-closing metathesis using Grubbs II catalyst[8] (CH₂Cl₂, reflux) to yield fused bicycle **66** (81% yield). Allylation of enone **66** (KHMDS, allyl iodide, 88% yield) afforded allyl derivative **67**, which underwent selective cross-metathesis with boronate **37** under the influence of Grubbs II catalyts[8,9] to give the corresponding vinyl pinacol boronates (82% yield, *E:Z* ca. 5:1). A two-step oxidation of these boronates[9a,10,11] (Me₃NO, 79% yield; NaClO₂, 95% yield),

(16) Nicolaou, K. C.; Gray, D. L. F.; Montagnon, T.; Harrison, S. T. *Angew. Chem., Int. Ed.* **2002**, *41*, 996.

[a] Reagents and conditions: (a) LDA (1.0 M in THF, 1.1 equiv), THF, −78 °C, 30 min; then 3-bromo-2-methylpropene (1.0 equiv), THF, −78 → 0 °C, 1 h, 88%; (b) LDA (1.0 M in THF, 1.1 equiv), THF, −78 °C, 30 min; then MeI (1.0 equiv), THF, −78 → 25 °C, 1 h, 84%; (c) DIBAL-H (1.0 M in toluene, 1.2 equiv), MeOH, −20 °C, 1 h; then aq. HCl (2.0 M, 5.1 equiv), −20 → 25 °C, 12 h, 95%; (d) TMSOTf (1.2 equiv), NEt₃ (1.2 equiv), CH₂Cl₂, 0 °C, 30 min; (e) IBX (1.4 equiv), MPO (1.4 equiv), DMSO, 1 h, 25 °C, 92% over two steps; (f) CuBr·Me₂S (2.0 equiv), vinyl magnesium bromide (4.4 equiv), −78 °C, THF, 2 h; then **64**, THF, −78 °C, 2 h; then MeI (8.0 equiv), HMPA, −78 → 25 °C, 12 h, 75%; (g) Grubbs catalyst **36** (0.1 equiv), CH₂Cl₂, reflux, 2 days, 81%; (h) KHMDS (4.0 equiv), THF:HMPA (4:1), −78 °C, 30 min; then allyl iodide (8.0 equiv), −78 → −10 °C, 1 h, 88%; (i) **37** (3.0 equiv), Grubbs catalyst **36** (0.14 equiv), benzene, reflux, 30 min, 82% (*E:Z* ca. 5:1); (j) Me₃NO (4.0 equiv), THF, 65 °C, 1 h, 79%; (k) NaClO₂ (3.0 equiv), 2-methyl-2-butene (10 equiv), NaH₂PO₄ (5.0 equiv), *t*-BuOH:water (1:1), 25 °C, 30 min, 95%; (l) **70** (2.0 equiv), HATU (4.0 equiv), NEt₃ (6.0 equiv), DMF, 25 °C, 12 h, 60%; (m) Chiralpak AD column, 12% isopropyl alcohol/hexanes; (n) TASF (2.0 equiv), DMF, 40 °C, 92% (for **10**) or 91% (for **11**); (o) *m*-CPBA (1.1 equiv), CH₂Cl₂, 0 °C, 1 h, 88%. Abbreviations: *m*-CPBA = 3-chloroperoxybenzoic acid; DIBAL-H = diisobutylaluminum hydride; DMSO = dimethyl sulfoxide; HMPA = hexamethylphosphoramide; IBX = 2-iodoxybenzoic acid; MPO = 4-methoxypyridine-*N*-oxide; Tf = trifluoromethanesulfonyl; TMS = trimethylsilyl.

proceeding through aldehyde **68**, gave the desired carboxylic acid fragment **69**. HATU-mediated amide coupling of the latter intermediate with TMSE-protected aniline **70** (available from Giannis's nitro ester)[17,18] gave the corresponding racemic amide (±)-**71** (60% yield) that was separated into its two enantiomers,

71 and *ent*-**71**, using a chiral HPLC column. Due to **71** having the same sense of optical rotation as the analogous amides of the bioactive platensimycin congeners carba-[4] and adamanta-platensimycin,[5] only **71**, and not *ent*-**71**, was subsequently transformed into the final product. Thus, cleavage of the TMSE group from **71** under mild conditions (TASF, DMF, 40 °C, 92% yield) furnished the targeted analogue **10**. Alternatively, chemoselective epoxidation of the trisubstituted olefin in **71** with *m*-CPBA (CH$_2$Cl$_2$, 0 °C, 88% yield), followed by TMSE removal (TASF, DMF, 40 °C), gave epoxide surrogate **11** (91% yield).

3.5. Pyran Analogue 9. The synthesis of platensimycin analogue **9** began from previously prepared intermediate **64** (see Scheme 6). Thus, conjugate addition of cyanide to this compound using Nagata's reagent[19] (Et$_2$AlCN, toluene, 0 °C) gave an isomeric mixture of nitriles (dr 5:1) that was separated using standard column chromatography to give **73** as the major diastereomer in 74% yield (Scheme 7). Two-step reduction of the resulting nitrile (DIBAL-H) to the corresponding primary alcohol led to concomitant reduction of the enone to the corresponding allylic alcohol, which was immediately reoxidized with MnO$_2$ to give hydroxy enone **74** (66% overall yield). Triflic acid-initiated cyclization of the latter compound then gave key pyran derivative **75** (56% yield). Sequential methylation (KHMDS, MeI, 71% yield) and allylation (KHMDS, allyl iodide, 75% yield) yielded disubstituted system **77** as a single diastereomer. Once again, application of our standard three-step sequence[2a] consisting of cross-metathesis[8,9] (vinyl boronate **37**, Hoveyda−Grubbs II catalyst (**78**),[20] 76% yield, *E:Z* ca. 5:1), oxidative cleavage[9a,10] (Me$_3$NO), and Pinnick oxidation[11] (NaClO$_2$) furnished the desired racemic carboxylic acid fragment (±)-**79** in 71% yield over the last two steps. Formation of the corresponding methyl ester **80** (TMSCHN$_2$, 85% yield) allowed separation of the racemic esters by HPLC on a chiral stationary phase. At this stage, the absolute configuration of the separated enantiomers **80** remained unknown, and so both enantiopure esters were progressed to the target analogue. Thus, basic hydrolysis using aqueous LiOH revealed enantiopure acid **79** (and its enantiomer *ent*-**79**) in 82% yield and >99% ee. Finally, HATU-mediated coupling of this carboxylic acid (and its enantiomer) with fully protected aniline **41**[2a] gave amide **81** (and its enantiomer *ent*-**81**) in 66% yield, which led, after protecting group removal, to the desired pyran analogue **9** (and its enantiomer *ent*-**9**) (66% yield).

3.6. Adamantaplatensimycin 3. The total synthesis of adamantaplatensimycin (**3**) began from commercially available bromoadamantane (**82**) (Scheme 8). Thus, radical-mediated conjugate addition of **82** to methyl acrylate (*n*-Bu$_3$SnH, AIBN, toluene, reflux, 75% yield)[21] afforded the corresponding adamantyl methyl ester (75% yield), which was hydrolyzed (aqueous KOH, MeOH) to carboxylic acid **83** in 99% yield. Acid chloride formation [(COCl)$_2$] followed by treatment with freshly prepared diazomethane or diazoethane (THF, 0 °C)[22]

**Scheme 7.** Synthesis of Cage Analogue **9**[a]
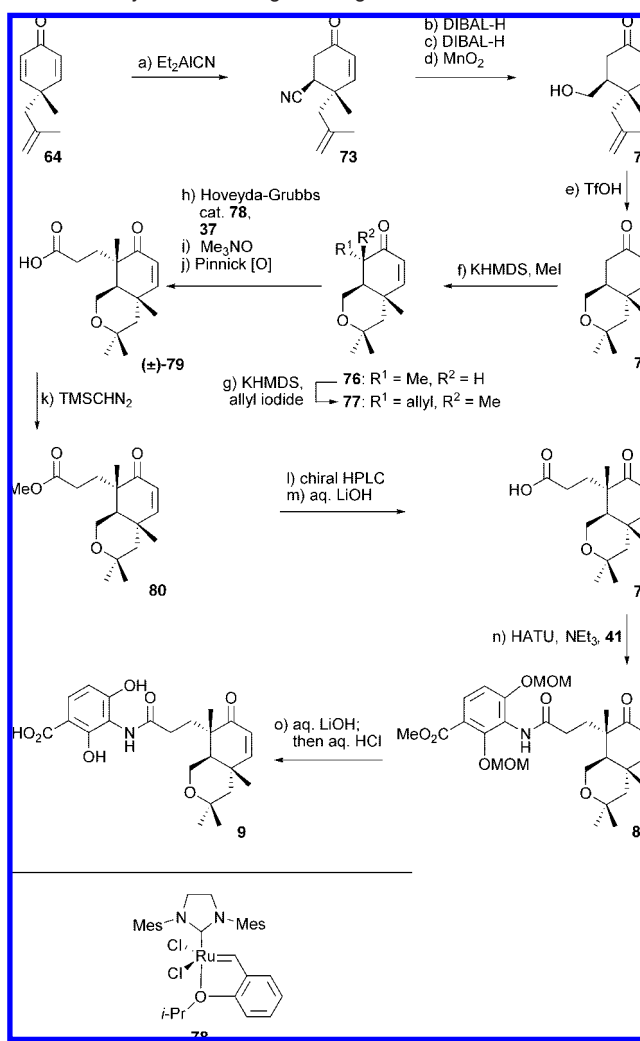
[a] Reagents and conditions: (a) Et$_2$AlCN (1.0 M in toluene, 1.1 equiv), toluene, 0 °C, 30 min, 74%; (b) DIBAL-H (1.0 M in CH$_2$Cl$_2$, 2.5 equiv), CH$_2$Cl$_2$, −78 °C, 30 min; (c) DIBAL-H (1.0 M in hexanes, 2.5 equiv), THF, −78 °C, 45 min; (d) MnO$_2$, EtOAc, 25 °C, 12 h, 66% over three steps; (e) TfOH (0.1 equiv), CH$_2$Cl$_2$, −20 °C, 1 h, 56%; (f) KHMDS (0.5 M in toluene, 1.5 equiv), THF:HMPA (5:1) −78 °C, 20 min; then MeI, −78 → −10 °C, 1 h, 71%; (g) KHMDS (0.5 M in toluene, 3.0 equiv), THF:HMPA (4:1), −78 °C, 20 min; then allyl iodide (3.0 equiv), −78 → −10 °C, 1 h, 75%; (h) **37** (5.0 equiv), Hoveyda−Grubbs catalyst **78** (0.2 equiv), CH$_2$Cl$_2$, 50 °C, 2 h, 76% (*E:Z* ca. 5:1); (i) Me$_3$NO (1.2 equiv), THF, 65 °C, 1 h; (j) NaClO$_2$ (2.3 equiv), 2-methyl-2-butene (7.6 equiv), NaH$_2$PO$_4$ (3.8 equiv), *t*-BuOH:water (1:1), 25 °C, 30 min, 71% over two steps; (k) TMSCHN$_2$ (2.0 M in Et$_2$O, 3.0 equiv), MeOH, 0 °C, 20 min, 85%; (l) Chiralcel OD column, 9% isopropyl alcohol/hexanes; (m) aq. LiOH (1.0 M, 20 equiv), THF, 25 °C, 82%; (n) **41** (3.0 equiv), HATU (3.5 equiv), NEt$_3$ (6.5 equiv), DMF, 25 °C, 18 h, 66%; (o) aq. LiOH (2.0 M, 25 equiv), THF, 45 °C, 3 h; then aq. HCl (2.0 M, 50 equiv), THF, 45 °C, 12 h, 66%. TMSCHN$_2$ = (trimethylsilyl)diazomethane.

gave diazoketones **84a** and **84b** in 75 and 70% overall yield, respectively. Exposure of **84a** to catalytic Rh$_2$(OAc)$_4$ (CH$_2$Cl$_2$, 25 °C) furnished cyclohexanone derivative **85a** (85% yield) through C−H insertion of the transient carbene species.[23] The potential to carry out an asymmetric variant of this

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(19) (a) Nagata, W.; Yoshioka, M.; Hirai, S. *Tetrahedron Lett.* **1962**, *3*, 461. (b) Nagata, W.; Yoshioka, M. *Tetrahedron Lett.* **1966**, *7*, 1913.
(20) Garber, S. B.; Kingsbury, J. S.; Gray, B. L.; Hoveyda, A. H. *J. Am. Chem. Soc.* **2000**, *122*, 8168.
(21) Ohno, M.; Ishizaki, K.; Egushi, S. *J. Org. Chem.* **1988**, *53*, 1285.
(22) A modification of the procedure reported in the following was used: Dyer, J. R.; Randall, R. B., Jr.; Deutsch, H. M. *J. Org. Chem.* **1964**, *29*, 3423.

(23) (a) Seminal publication on Rh$_2$(OAc)$_4$-mediated C−H insertion: Breslow, R.; Gellman, S. H. *J. Am. Chem. Soc.* **1983**, *105*, 6728. For selected reviews on metal−carbenoid C−H insertion, see: (b) Doyle, M. P.; Forbes, D. C. *Chem. Rev.* **1996**, *96*, 911. (c) Davies, H. M. L.; Beckwith, R. E. J. *Chem. Rev.* **2003**, *103*, 2861. (d) Davies, H. M. L.; Manning, J. R. *Nature* **2008**, *451*, 417.

Scheme 8. Synthesis of Adamantaplatensimycin 3[a]
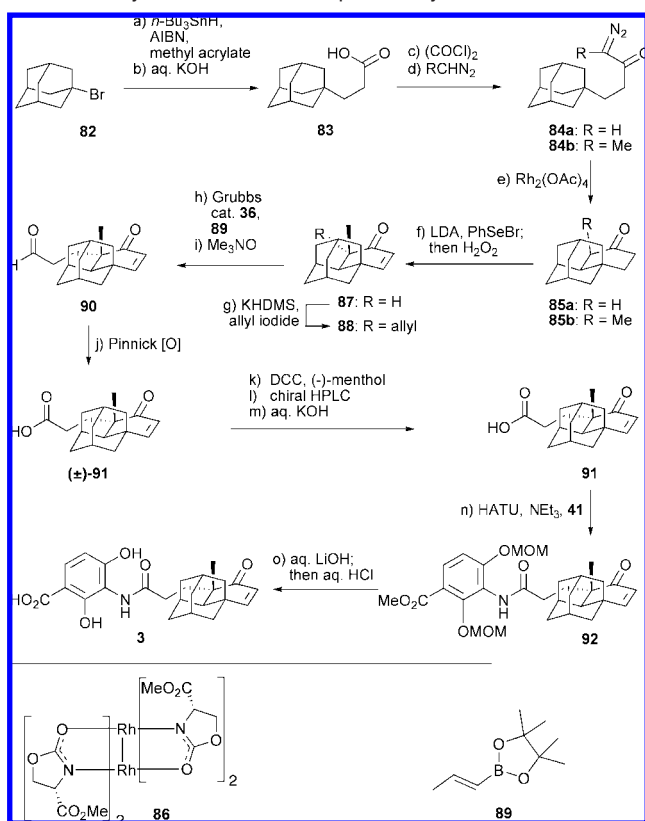
[a] Reagents and conditions: (a) n-Bu₃SnH (1.2 equiv), methyl acrylate (2.0 equiv), AIBN (0.05 equiv), toluene, reflux, 2 h, 75%; (b) KOH (6.0 equiv), MeOH:water (12:1), 25 °C, 12 h, 99%; (c) (COCl)₂ (1.2 equiv), CH₂Cl₂, DMF (drop), 0 °C, 2 h; (d) diazomethane (3.0 equiv), THF, 0 °C, 1 h, 75% over two steps (for 85a), diazoethane (2.9 equiv), THF, 0 °C, 1 h, 70% over two steps (for 85b); (e) Rh₂(OAc)₄ (0.03 equiv), CH₂Cl₂, 25 °C, 30 min, 65%; (f) i. LDA (1.0 M in THF, 1.2 equiv), −78 °C, 30 min; then PhSeBr, −78 → −50 °C, 30 min; ii. H₂O₂ (10 equiv), pyridine (2.0 equiv), CH₂Cl₂, 25 °C, 3 h, 88%; (g) KHMDS (0.5 M in toluene, 4.0 equiv), THF:HMPA (4:1), −78 °C, 15 min; then allyl iodide (8.0 equiv), −78 → −10 °C, 1 h, 86%; (h) 89 (3.0 equiv), Grubbs catalyst 36 (0.05 equiv), benzene, reflux, 1 h, 85% (*E:Z* ca. 5:1); (i) Me₃NO (2.7 equiv), THF, 65 °C, 1 h, 90%; (j) NaClO₂ (3.0 equiv), 2-methyl-2-butene (10 equiv), NaH₂PO₄ (5.0 equiv), *t*-BuOH:water (1:1), 25 °C, 30 min, 99%; (k) (−)-menthol (1.2 equiv), DCC (1.5 equiv), 4-DMAP (0.1 equiv), CH₂Cl₂, 25 °C, 1 h; (l) Chiralcel OD-H column, 0.5% isopropyl alcohol/hexanes; (m) aq. KOH (1.0 M, 6.0 equiv), MeOH:water (10:1), 25 °C, 85% over three steps; (n) 41 (2.0 equiv), HATU (4.0 equiv), NEt₃ (6.0 equiv), DMF, 25 °C, 12 h, 71%; (o) aq. LiOH (2.0 M, 30 equiv), THF, 45 °C, 3 h; then aq. HCl (2.0 M, 56 equiv), THF, 45 °C, 12 h, 92%. Abbreviations: AIBN = 2,2′-azobis(isobutyronitrile); DCC = *N,N*′-dicyclohexylcarbodiimide.

reaction was investigated using a variety of chiral rhodium catalysts, including Rh₂(MEOX)₄,[24a] Rh₂(MEAZ)₄,[24b] Rh₂(MPPIM)₄,[24c] and Rh₂(MEPY)₄.[24d−f] The desired product was formed only using the Rh₂(MEOX)₄ catalyst (86),[24a] which provided 85a in 70% ee and 64% yield. However, it proved impossible to effect regioselective enolization of the newly formed cyclohexanone ring of 85a, and an alternative means of differentiating the two similar methylene groups was sought. This obstacle was overcome by installing the methyl group prior to cyclization, which also served to reduce the length of the route by one step. Thus, decomposition of diazoketone 84b using catalytic Rh₂(OAc)₄ (CH₂Cl₂, 25 °C) led to the formation of α-methyl ketone 85b in 65% yield as a single (unassigned) diastereoisomer. Unfortunately, the more hindered diazoketone 84b would not undergo an

enantioselective C−H insertion reaction with any of the chiral catalysts investigated.[24]

Regioselective functionalization of ketone 85b through α-selenation (LDA, PhSeBr) and oxidation/*syn* elimination (H₂O₂, pyridine)[25] afforded 87 in 88% yield. Standard allylation (KHMDS, allyl iodide) gave diene 88 (86% yield), which entered into a cross-metathesis reaction with vinyl boronate 89 in the presence of Grubbs II catalyst[8,9] (benzene, reflux) to afford the corresponding vinyl boronates (85% yield, *E:Z* ca. 5:1). Oxidative cleavage of the boronate moiety from this mixture with Me₃NO[9a,10] (THF, 65 °C, 90% yield) gave aldehyde 90, which was converted to carboxylic acid (±)-91 through Pinnick oxidation[11] (NaClO₂, 99% yield). At this stage, transformation of racemic carboxylic acid (±)-91 into its diastereomeric (−)-menthol esters, under standard amide coupling conditions (DCC, 4-DMAP, CH₂Cl₂, 25 °C), enabled separation of the two isomers by chiral HPLC. Each diastereomer of the (−)-menthol esters so obtained was then saponified with aqueous KOH to afford the two enantiomerically pure carboxylic acids (91 and *ent*-91) in 85% combined yield from (±)-91. Each carboxylic acid (91 and *ent*-91) was coupled to aniline 41[2a] using HATU in the usual way to give amide 92 (and its enantiomer *ent*-92) in 71% yield. Global deprotection of the latter compounds with aqueous LiOH (THF, 45 °C) and then with aqueous HCl (THF, 45 °C) afforded the targeted adamantaplatensimycin 3 and its enantiomer *ent*-3 in 92% yield.

3.7. Carbaplatensimycin 2. The substitution of the ether oxygen atom within the platensimycin cage structure with a methylene moiety (CH₂) was deemed of interest since it was thought to provide a measure of the importance of the oxygen for the biological activity. Carbaplatensimycin (2), the all-carbon-ketolide isomer of platensimycin, was therefore defined as a target, and its synthesis is outlined in Scheme 9. Commencing with enantiopure aldehyde 93, an intermediate used in our previously reported asymmetric synthesis of platensimycin,[2b] this sequence relied on a base-induced anionic ring-closure and a radical-mediated cyclization to forge the tetracyclic core of the molecule. Thus, formation of cyanohydrin 94 from aldehyde 93 (TMSCN, Et₃N; then aqueous HCl) and protection (ethyl vinyl ether, PPTS, 99% yield over two steps) gave ethoxyethyl (EE)-protected cyanohydrin 95 (as an inconsequential 1:1:1:1 mixture of the four diastereomers). Exposure of this mixture to KHMDS in THF at −78 → 25 °C led to the formation of tricyclic compound 96 in 70% yield and as a single C-10 epimer (two diastereomers due to the EE group). Subsequent methylenation of the enone functionality within 96 (Ph₃P=CH₂, 92% yield) provided diene 97, which served as a protected surrogate for the required enone until a later stage.

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(25) A modification of the procedure reported in the following was used: Reich, H. J.; Reich, I. L.; Renga, J. M. *J. Am. Chem. Soc.* 1973, *95*, 5813.

Scheme 9. Synthesis of Carbaplatensimycin **2**[a]
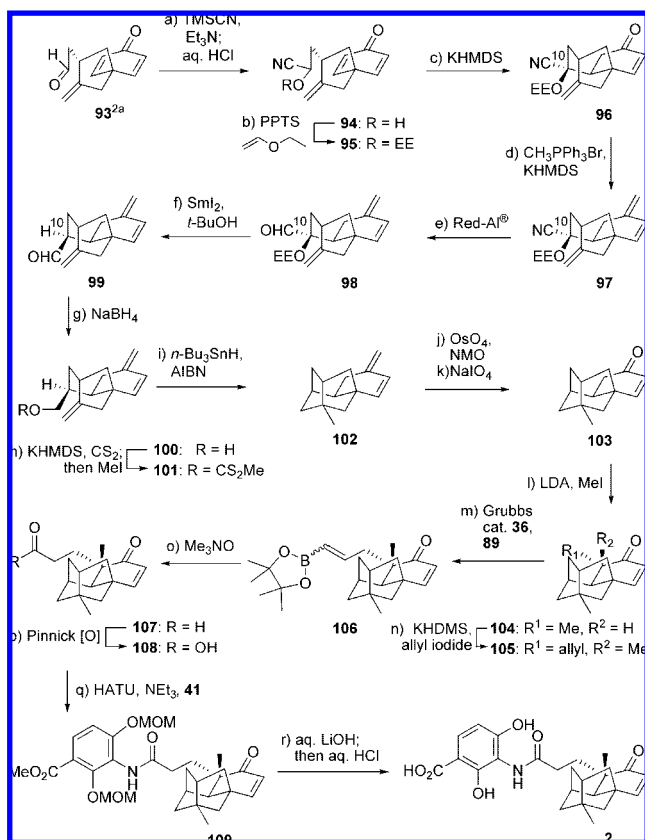
[a] Reagents and conditions: (a) TMSCN (1.5 equiv), Et$_3$N (0.5 equiv), CH$_2$Cl$_2$, 25 °C, 1 h; then aq. HCl (1.0 M, 8.1 equiv); (b) ethyl vinyl ether (excess), PPTS (0.5 equiv), CH$_2$Cl$_2$, 25 °C, 12 h, 99% over two steps; (c) KHMDS (0.5 M in toluene, 1.5 equiv), THF, −78 → 25 °C, 0.5 h, 70%; (d) CH$_3$PPh$_3$Br (2.5 equiv), KHMDS (0.5 M in toluene, 2.0 equiv), THF, −78 → 0 °C, 1 h, 92%; (e) Red-Al (3.5 M in toluene, 5.0 equiv), THF, −20 → 25 °C, 6 h, 90%; (f) SmI$_2$ (0.1 M in THF, 1.5 equiv), THF:t-BuOH (10:1), −10 → 25 °C, 2 h, 92%; (g) NaBH$_4$ (1.5 equiv), THF:CH$_3$OH (2:1), 0 → 25 °C, 30 min, 99%; (h) CS$_2$ (3.0 equiv), KHMDS (0.5 M in toluene, 3.0 equiv), THF, −78 → 25 °C, 1 h; then MeI (3.0 equiv), THF, 25 °C, 1 h, 99%; (i) n-Bu$_3$SnH (1.5 equiv), AIBN (0.75 equiv), benzene, 80 °C, 30 min, 65%; (j) OsO$_4$ (2.5 wt % in t-BuOH, 0.025 equiv), NMO (50 wt % in water, 2.0 equiv), acetone:water (8:1), 0 °C, 4 h, 80%; (k) NaIO$_4$ (2.0 equiv), THF:water (1:1), 25 °C, 1 h, 90%; (l) KHMDS (0.5 M in toluene, 2.0 equiv), THF:HMPA (5:1), −78 °C, 30 min; then MeI (8.0 equiv), −10 °C, 1 h, 92%; (m) KHMDS (0.5 M in toluene, 4.0 equiv), −78 °C, 30 min; then allyl iodide (8.0 equiv), −10 °C, 1 h, 87%; (n) Grubbs catalyst **36** (10 mol %), **89** (5.0 equiv), benzene, 80 °C, 1 h, 85% (E:Z ca. 4:1); (o) Me$_3$NO (6.0 equiv), THF, 65 °C, 2 h, 85%; (p) NaClO$_2$ (5.0 equiv), 2-methyl-2-butene (10 equiv), NaH$_2$PO$_4$ (7.0 equiv), t-BuOH:water (1:1), 25 °C, 30 min, 95%; (q) **41** (3.0 equiv), HATU (4.0 equiv), NEt$_3$ (6.0 equiv), DMF, 25 °C, 20 h, 80%; (r) aq. LiOH (2.0 M, 30 equiv), THF, 45 °C, 6 h; then aq. HCl (2.0 M, 60 equiv), THF, 45 °C, 24 h, 82%. Abbreviations: EE = 1-ethoxyethyl; NMO = 4-methylmorpholine N-oxide; PPTS = pyridinium 4-toluenesulfonate; Red-Al = sodium bis(2-methoxyethoxy)aluminum hydride; TBAF = tetra-n-butylammonium fluoride.

This tactic allowed the selective reduction of the nitrile moiety of the latter intermediate (Red-Al, 90% yield), leading to aldehyde **98**. Deoxygenation of **98** with SmI$_2$ [THF:t-BuOH (10:1), −10 → 25 °C] led to aldehyde **99** in 92% yield and as a single isomer with the configuration of the formyl-bearing carbon (C-10) inverted as shown. This aldehyde was then reduced to the primary alcohol (**100**) through the action of NaBH$_4$ (99% yield). Formation of xanthate **101** from alcohol **100** (KHMDS, CS$_2$; then MeI, 99% yield) and treatment with n-Bu$_3$SnH and AIBN resulted in a 5-*exo*-trig radical cyclization to cast the final ring, providing hydrocarbon **102** in 65% yield.

The now superfluous methylene group was removed through a two-step procedure involving dihydroxylation (NMO, OsO$_4$ catalyst, 80% yield) and NaIO$_4$ cleavage of the resulting 1,2-diol (90% yield) to afford the desired enone **103**. The latter compound (**103**) was then converted to carboxylic acid **108** through intermediates **104**−**107** and by the same sequence described previously.[2a] Thus, sequential alkylation of **103** (KHMDS, MeI, 92% yield; then KHMDS, allyl iodide, 87% yield) afforded diene **105** through **104**, which was transformed into the mixture of vinyl boronates (85% yield, E:Z ca. 4:1) through regioselective cross-metathesis with pinacol boronate **89** in the presence of Grubbs II catalyst.[8,9] The oxidation of boronates **106**, first with Me$_3$NO[9a,10] (85% yield) and then with NaClO$_2$[11] (95% yield), proceeded smoothly to give carboxylic acid **108** via aldehyde **107**. HATU-induced amide coupling of carboxylic acid **108** with aniline derivative **41**[2a] yielded amide **109** (80% yield), which upon global deprotection (aqueous LiOH, then aqueous HCl) furnished carbaplatensimycin (**2**) in 82% overall yield.
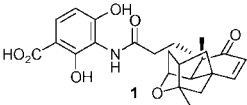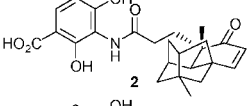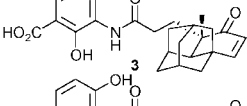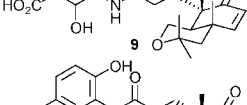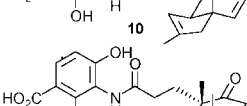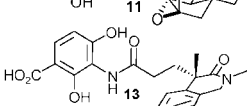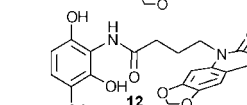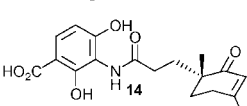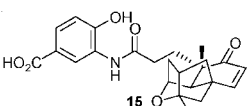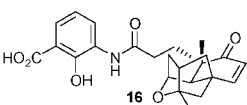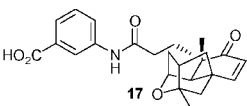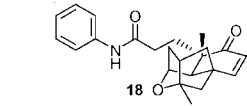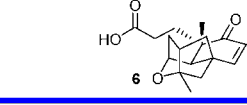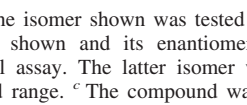
4. Biological Evaluation of Platensimycin Analogues. The antibacterial activities of the synthesized platensimycin analogues against MRSA and VREF were determined and compared to those of platensimycin. For these cell-based assays, inocula of MRSA (ATCC 33591), E. coli (ATCC 29425), and VREF (ATCC 51575) were prepared using overnight cultures of microorganisms grown in either Nutrient Broth (DIFCO, Detroit, MI) or Brain Heart Infusion (BHI, DIFCO, Detroit, MI) in the case of VREF. The inocula were then diluted to an approximate concentration of 5×10^5 cfu μL^{-1} in cation-adjusted Mueller−Hinton Broth II (Becton Dickinson, Sparks, MD), or BHI in the case of VREF, as determined by a colony count on a nutrient agar plate. Cells (80 μL) were dispensed into the 96-well microtiter plates that contained serial dilutions of the tested compounds (20 μL) or a known antibacterial agent (vancomycin for MRSA, streptomycin for E. coli, and dapto-mycin for VREF). Plates were incubated with gentle agitation at 37 °C for 24 h. The minimum inhibitory concentration (MIC) was determined as the lowest concentration at which no bacteria were observed to grow as determined by OD$_{595}$.

As seen in Table 1, the most complex members of analogue series I, carbaplatensimycin (**2**) and adamantaplatensimycin (**3**) (entries 2 and 3, respectively), retain nearly platensimycin-like antibacterial potency, exhibiting MIC values against MRSA and VREF of 1.1−2.2 and 1.3−1.8 μg mL^{-1}, respectively. These values are of similar magnitude to those found for platensimycin (**1**) (entry 1, MIC$_{MRSA}$ = 0.2−0.4 μg mL^{-1}, MIC$_{VREF}$ = 0.4−0.8 μg mL^{-1}).
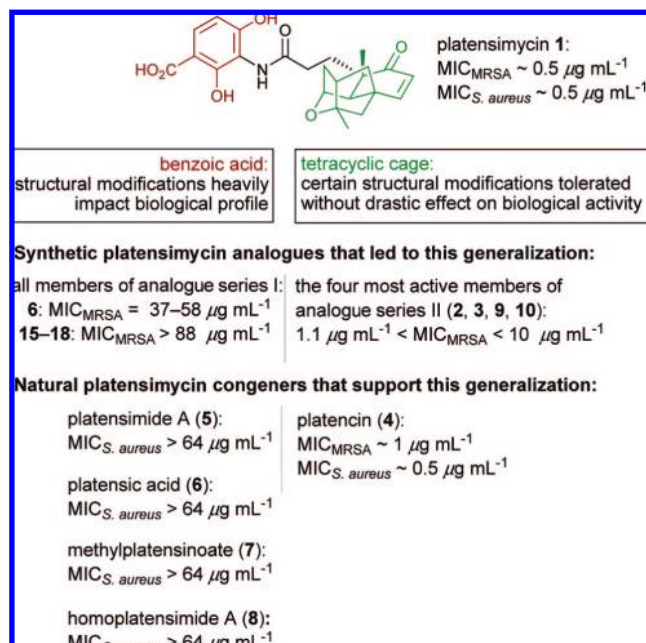
Strikingly, even derivatives **9** and **10**, which contain simple bicyclic structures as surrogates for the tetracyclic platensimycin cage, demonstrate some antibiotic activity (entries 4 and 5). Despite the lower level of architectural complexity, the measured MIC values for **9** (MIC$_{MRSA}$ = 3.5−4.3 μg mL^{-1}, MIC$_{VREF}$ = 6.5−8.6 μg mL^{-1}, entry 4) and **10** (MIC$_{MRSA}$ = 8−10 μg mL^{-1}, MIC$_{VREF}$ > 80 μg mL^{-1}, entry 5) were comparable to those of the parent natural product. Compounds **12**−**14** (entries 7−9), which contain highly altered cage motifs, show no measurable antibacterial effect (MIC$_{MRSA}$ > 69 μg mL^{-1}, MIC$_{VREF}$ > 69 μg mL^{-1}).

Interestingly, while certain modifications within analogue series II were permitted without having a significant adverse effect on the antibacterial potency, even small modifications within the benzoic acid region (analogue series I) proved detrimental to activity. As such, the platensimycin aniline

Table 1. Minimum Inhibitory Concentration Values (μg mL^{-1}) of Platensimycin and Analogues against Methicillin-Resistant *Staphylococcus aureus*, Vancomycin-Resistant *Enterococcus faecium*, and *E. coli*

Entry	Compound	MRSA	VREF	E. coli
1[a]	**1**	0.2–0.4	0.4–0.8	>88
2[a]	**2**	1.1–2.2	1.1–2.2	>88
3[b]	**3**	1.3–1.8	1.3–1.8	>88
4[b]	**9**	3.5–4.3	6.5–8.6	>86
5[b]	**10**	8.0–10	>80	>80
6[b]	**11**	17–20	>83	>83
7[c]	**13**	>88	>88	>88
8	**12**	>86	>86	>86
9[b]	**14**	>69	>69	>69
10[a]	**15**	>85	>85	>85
11[a]	**16**	>85	>85	>85
12[a]	**17**	>82	>82	>82
13[a]	**18**	>73	>73	>73
14[a]	**6**	37–58	>58	>58

[a] Only the isomer shown was tested in the antibacterial assay. [b] Both the isomer shown and its enantiomer were tested separately in the antibacterial assay. The latter isomer was found to be inactive in the investigated range. [c] The compound was tested as a racemic mixture in the antibacterial assay.

platensimycin **1**:
MIC$_{MRSA}$ ~ 0.5 μg mL^{-1}
MIC$_{S.\ aureus}$ ~ 0.5 μg mL^{-1}

benzoic acid: structural modifications heavily impact biological profile

tetracyclic cage: certain structural modifications tolerated without drastic effect on biological activity

Synthetic platensimycin analogues that led to this generalization:

all members of analogue series I:
6: MIC$_{MRSA}$ = 37–58 μg mL^{-1}
15–18: MIC$_{MRSA}$ > 88 μg mL^{-1}

the four most active members of analogue series II (**2, 3, 9, 10**):
1.1 μg mL^{-1} < MIC$_{MRSA}$ < 10 μg mL^{-1}

Natural platensimycin congeners that support this generalization:

platensimide A (**5**):
MIC$_{S.\ aureus}$ > 64 μg mL^{-1}

platensic acid (**6**):
MIC$_{S.\ aureus}$ > 64 μg mL^{-1}

methylplatensinoate (**7**):
MIC$_{S.\ aureus}$ > 64 μg mL^{-1}

homoplatensimide A (**8**):
MIC$_{S.\ aureus}$ > 64 μg mL^{-1}

platencin (**4**):
MIC$_{MRSA}$ ~ 1 μg mL^{-1}
MIC$_{S.\ aureus}$ ~ 0.5 μg mL^{-1}

Figure 4. Simple guide to the platensimycin pharmacophore with synthetic and natural analogues, illustrating the observed structure−activity relationships.

derivatives **15**−**18** all suffered drastically in terms of their potency, displaying no measurable antibacterial effect against both MRSA and VREF (MIC$_{MRSA}$ > 73 μg mL^{-1}, MIC$_{VREF}$ > 73 μg mL^{-1}, entries 10−13).

Our results suggest that the structural requirements to retain antibacterial activity are far more stringent with respect to the benzoic acid than with respect to the tetracyclic cage domain (Figure 4). While only minor modifications in the benzoic acid region led to a loss of the antibacterial activity, the tetracyclic cage is far more tolerant to modification and can be substituted for simpler bicyclic structures that still exhibit platensimycin-like antibacterial activity.

Within this context, the recent isolation of the platensimycin congeners platensimide A (**5**),[7a] platensic acid (**6**),[7a] methyl-platensinoate (**7**),[7a] homoplatensimide A (**8**),[7a] and platencin (**4**)[6] should also be considered (Figure 4). Compounds **5**−**8** were found to be inactive against *S. aureus* at concentrations of 64 μg mL^{-1} (i.e., MIC$_{S.\ aureus}$ > 64 μg mL^{-1}) and showed considerably lower activity than platensimycin in fatty acid biosynthesis assays.[7] In contrast, platencin (**4**), which retains the benzoic acid moiety but has an altered ketolide ring system lacking the characteristic ether linkage, showed similar antibiotic activity to platensimycin across a range of bacterial strains, including MRSA and *S. aureus*. It should be noted, however, that platencin is a significantly weaker inhibitor of FabF compared with platensimycin and rather acts as a dual inhibitor of FabF and the initiation-condensing enzyme FabH (against which platensimycin is only a weak inhibitor). As our MIC assay alone cannot distinguish these mechanisms of action, we cannot discount the possibility that some or all of our synthetic analogues in series II may act in a similar way to platencin.

Conclusion

In conclusion, we have synthesized two series of designed platensimycin surrogates, and our initial antibacterial assays indicate that certain modifications of the intricate cage region can be made without detrimental effects on potency, whereas

even small modifications of the benzoic acid region result in a drastic loss of activity. These results should serve as guidelines for the design and synthesis of antibacterial agents inspired by the platensimycin lead structure and, thus, ultimately facilitate the fight against multiple-drug-resistant bacteria.

Supporting Information Available: Complete refs 1a and 6a; experimental procedures and compound characterization. This material is available free of charge via the Internet at http://pubs.acs.org.

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