Natural Products Synthesis

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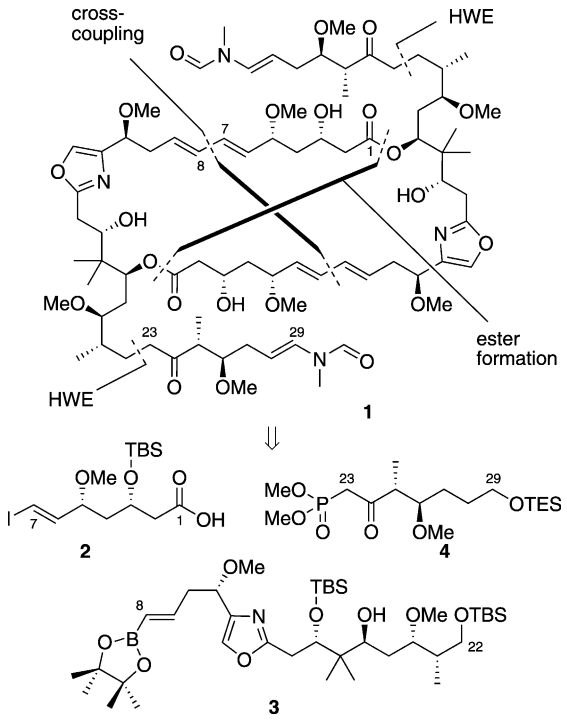
Total Synthesis of Rhizopodin**

*Michael Dieckmann, Manuel Kretschmer, Pengfei Li, Sven Rudolph, Daniel Herkommer, and Dirk Menche**

Dedicated to Professor Gerhard Bringmann on the occasion of his 60th birthday

Rhizopodin (**1**, Scheme 1) is an architecturally unique polyketide macrolide that was originally isolated from the myxobacterium *Myxococcus stipitatus* by the groups of Höfle and Reichenbach.[1,2] It shows potent antifungal and antiproliferative cytotoxicity against a range of tumor cell lines with IC_{50} values in the low nanomolar range.[1] On a molecular level, **1** disrupts the cytoskeleton of actin by binding specifically to a few critical sites of G-actin.[3] Actin presents one of the two major components of the cytoskeleton in eukaryotic cells, in which it plays a critical role in the determination of cell shape and a variety of cellular processes, including cell motility, division, adhesion, and intracellular transportation.[4] Actin-binding agents are becoming increasingly important as molecular probes to understand the organization and unveil the cellular functions of actin, and as potential lead structures for the development of novel chemotherapeutic anticancer agents.[4] Rhizopodin has also been shown to reduce phagocytosis efficiency of yeast cells.[5] While rhizopodin was initially reported to be a planar monomeric structure,[1,3] its revised structure was reported to be a C_2-symmetric dimer, which is distinguished by a 38-membered macrolide ring, two conjugated diene systems in combination with two disubstituted oxazole systems, and two side chains, which are terminated by *N*-vinylformamide moieties.[6] In total, it contains 18 stereogenic centers in the carbon backbone. The full stereochemistry was independently assigned by our group through high-field NMR experiments in combination with molecular modeling and chemical derivatization,[7] and by Schubert and co-workers with an X-ray structure of an actin–rhizopodin complex.[8] The important biological properties of rhizopodin and its rare occurrence in nature, together with its unique and intriguing molecular architecture, renders this natural product an attractive synthetic target, and several syntheses of fragments have been reported,[9] including the preparation of the originally proposed monomeric structure.[9d] Herein, we present the first total synthesis of rhizopodin and unequivocally confirm its relative and absolute configuration.



Scheme 1. Retrosynthetic analysis of rhizopodin. HWE = Horner–Wadsworth–Emmons reaction, TBS = *tert*-butyldimethylsilyl, TES = triethylsilyl.

Scheme 1 outlines our retrosynthetic analysis. The symmetry of the molecule allows the indicated double disconnections and the adoption of a highly convergent synthetic plan by using three building blocks of similar complexity, that is, the macrocyclic fragments **2** and **3**, and the side chain **4**. In detail, sequential disconnection at the C6–C8 diene linkages implies a cross-coupling toward a macrocycle and a conven-

[*] Dipl.-Chem. M. Dieckmann,[+] Dipl.-Chem. M. Kretschmer,[+] Dr. P. Li,[$] Dr. S. Rudolph[#]
Universität Heidelberg, Organisch-Chemisches Institut
INF 270, 69120 Heidelberg (Germany)
Dipl.-Chem. D. Herkommer, Prof. Dr. D. Menche
Kekulé-Institut für Organische Chemie und
Biochemie der Universität Bonn
Gerhard-Domagk-Str. 1, 53121 Bonn (Germany)
E-mail: dirk.menche@uni-bonn.de
[$] Present address: Xi'an Jiaotong University, Xi'an, Shaanxi Province (PR China)
[#] Present address: ASM Research Chemicals, Hannover (Germany)
[+] These authors contributed equally to this work.

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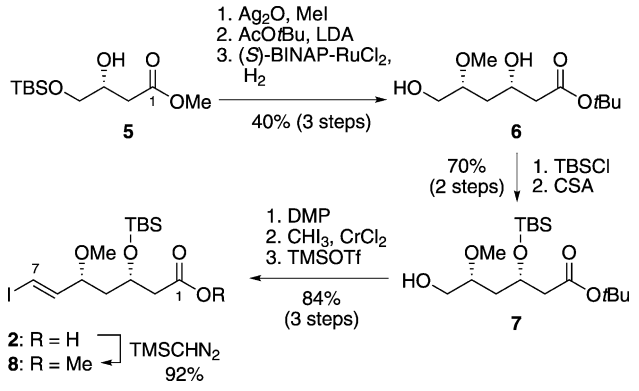
tional cross-coupling as late steps in the synthesis. Two esterification reactions, in turn, point to two common precursors, a C1–C7 fragment **2** and a C8–C22 segment **3**, for both halves of the macrocycle of the target molecule. Finally, introduction of the C23–C30 side-chain segment **4** was planned through a suitable HWE-coupling/hydrogenation sequence. In principle, several methodologies may be employed for ring closure, thus offering considerable flexibility in the synthesis. Importantly, the modular synthetic approach employed is highly convergent and concise, and thus offers the potential to provide a range of structural derivatives for structure–activity relationship studies to further explore the biological potential of this promising macrolide antibiotic.

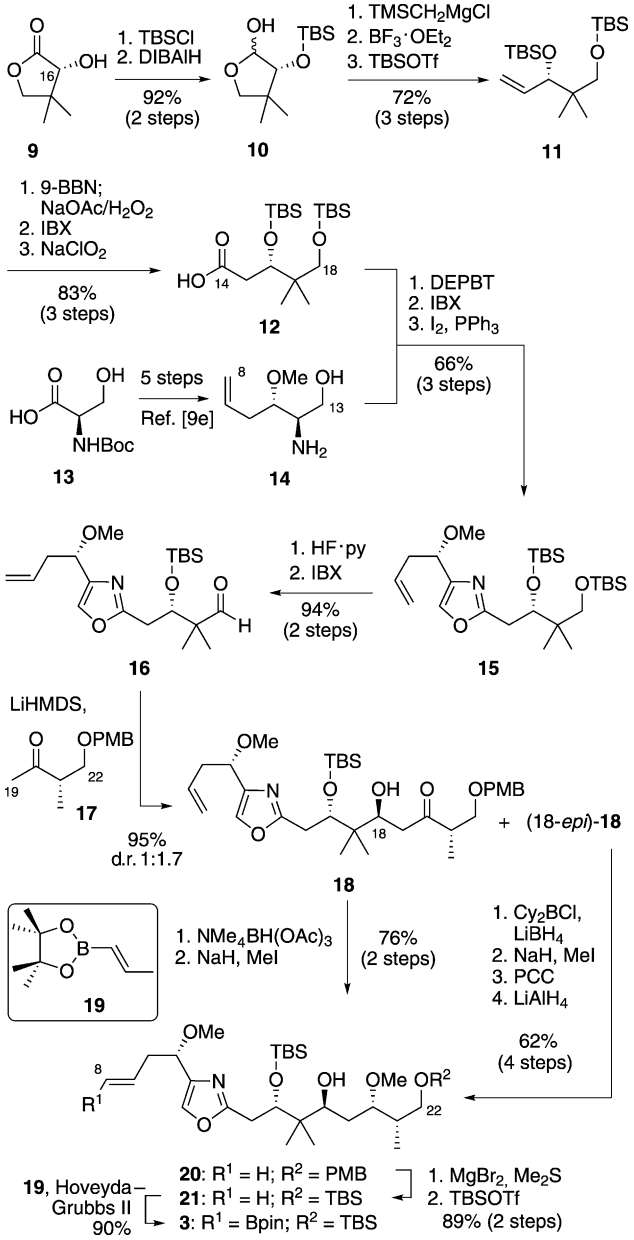
Construction of vinyl iodide **2** started from D-malic acid derived ester **5**,[10] which was methylated and homologated by a Claisen condensation with lithio *tert*-butyl acetate[11] to give the corresponding β-keto ester together with minor amounts of the tertiary alcohol arising from double addition (Scheme 2). Asymmetric hydrogenation in the presence of



Scheme 2. Synthesis of the C1–C7 subunit **2**. BINAP = 2,2′-bis(diphenylphosphino)-1,1′-binaphthyl, CSA = campher sulfonic acid, DMP = Dess–Martin periodinane, LDA = lithium diisopropylamide, Tf = trifluoromethanesulfonyl, TMS = trimethylsilyl.

Noyori's catalyst[12] gave the β-hydroxy ketone **6**, in which the TBS protecting groups was also removed, with excellent diastereoselectivity (>20:1). Selective protection of the secondary alcohol was effected in two steps, which involved double protection and subsequent liberation of the primary alcohol **7**. Homologation to building block **2** was achieved by oxidation to the respective aldehyde, introduction of the vinyl iodide by a Takai reaction[13] (89%, *E*/*Z* 7:1), and TMSOTf-mediated cleavage of the ester (3 steps, 84%). The corresponding methyl ester **8**, which was required for selective coupling, was readily available upon exposure to $TMSCHN_2$.

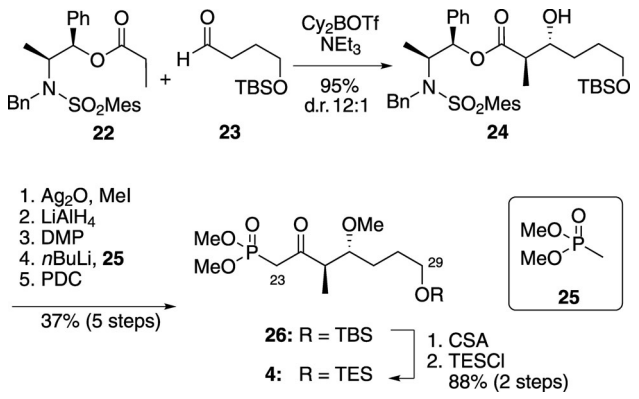
For the construction of the C8–C22 subunit **3**, we had previously reported a route,[9e] which however proved to be problematic during scale-up. Consequently, we developed an alternative and more convergent strategy which relied on the coupling of two segments of similar complexity, that is, our previously described C8–C13 fragment **14** (derived from *N*-Boc-D-serine (**13**) in 5 steps)[9e] and the C14–C18 fragment **12**, which arises from (−)-pantolactone (**9**, Scheme 3). In detail, commercially available lactone **9** already features the required hydroxy-bearing stereogenic center at C16 and was thus transformed into the TBS-protected lactol **10**, which was functionalized to alkene **11** (3 steps, 72%) by a Peterson-olefination sequence.[14] Oxidation of alkene **11** to acid **12** proceeded smoothly in three steps. This route proved superior to a shorter sequence that involved an asymmetric allylation of an aldehyde derived from 2,2-dimethyl-1,3-propandiol, both in terms of price and scalability.[15] With multigram quantities of **12** and **14** in hand, we proceeded with their transformation to **15** under slightly modified Wipf condi-



Scheme 3. Preparation of the C8–C22 subunit **3**. 9-BBN = 9-borabicyclo[3.3.1]nonane, Boc = *tert*-butoxycarbonyl, DEPBT = 3-(diethoxyphosphoryloxy)-1,2,3-benzotria-zin-4(3*H*)-one, DIBAlH = diisobutylaluminium hydride, Cy = cyclohexyl, IBX = *ortho*-iodoxybenzoic acid, LiHMDS = lithium hexamethyldisilazanide, PCC = pyridinium chlorochromate, pin = pinacolato, PMB = *para*-methoxybenzyl.

tions.[16] This conversion involved amide formation with 3-(diethoxyphosphoryloxy)-1,2,3-benzotriazin-4(3*H*)-one (DEPBT) and subsequent oxidation/cyclodehydration with IBX and I_2/PPh_3, respectively, to afford **15** in good yield. Next, the primary alcohol was liberated selectively and oxidized to aldehyde **16**, thus setting the stage for an aldol coupling with methyl ketone **17**. All attempts to install the stereocenter at C18 selectively were however unsuccessful (Mukaiyama and various boron-mediated aldol reactions, among others). After extensive optimization, we found that use of LiHMDS as base resulted in a mixture of the chromatographically separable diastereomeric alcohols **18** and (18-*epi*)-**18** (d.r. 1:1.7) in excellent yield. Both products could be transformed into building block **20** in a stereoconvergent manner, by a 1,3-*anti* reduction[17] and methylation (**18**), and by a 1,3-*syn*-reduction,[18] methylation, and inversion of the configuration at C-18 [(18-*epi*)-**18**], respectively. After introduction of the terminal TBS ether (2 steps, 89%),[19] the required boronate **3** was obtained from **21** by cross-metathesis reaction with vinyl boronate **19** in very good yield in the presence of the Hoveyda-Grubbs II catalyst.[20]

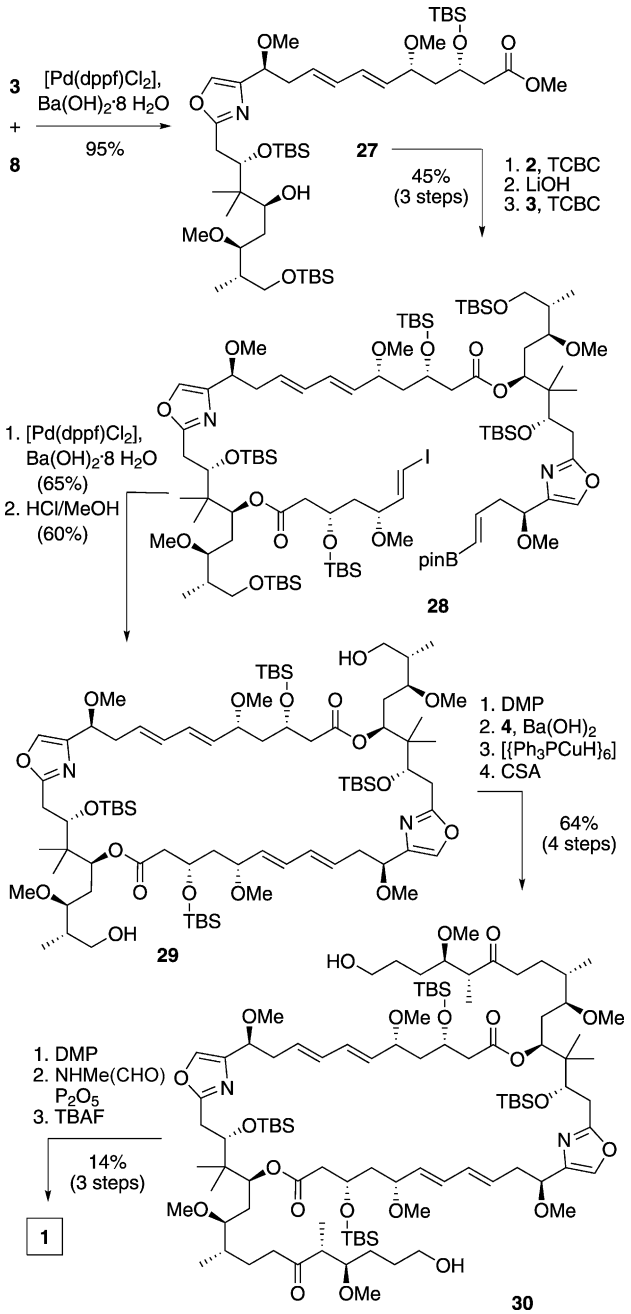
Our synthesis of the side-chain fragment **4** was based on an *anti*-aldol reaction of Masamune's chiral ephedrine-derived ethyl ketone **22**[21] with aldehyde **23**, giving alcohol **24** in high yield and good selectivity (Scheme 4). After methylation, the transformation to phosphonate **26** proved



Scheme 4. Assembly of the C23–C29 subunit **4**. Bn = benzyl, Mes = 2,4,6-trimethylphenyl, PDC = pyridinium dichromate.

most reliable by using a four-step sequence, which relied on the initial reductive removal of the auxiliary with $LiAlH_4$ and oxidation of the resulting primary alcohol with DMP.[22] After addition of lithiated phosphonate **25**, the intermediary β-hydroxyphosphonate was oxidized with PDC to furnish **26** in acceptable yield (37%) over five steps. Finally, liberation of the primary alcohol and re-protection as the TES ether gave the desired side-chain building block **4** in good yield.[23]

In a rationale to install the labile side chain, which contains the sensitive *N*-vinylformamide functionality, in a late stage of the synthesis, our strategy to unite the fragments relied on closing the macrocyclic core first (Scheme 5). After coupling of **3** and **8** by Suzuki reaction[24] under the conditions described by Gopalarathnam and Nelson,[25] the derived diene **27** was transformed to **28**. To



Scheme 5. Completion of the total synthesis. dppf = diphenylphosphinoferrocene, TCBC = 2,4,6-trichlorobenzoyl chloride, TBAF = tetra-*n*-butyl ammonium fluoride.

this end, it was esterified with **2** using a Yamaguchi procedure,[26] before the resulting terminal methyl ester was cleaved selectively and another Yamaguchi esterification with **8** gave **28** in good yields. Finally, Suzuki macrocyclization proceeded in 65% yield, thus giving the desired C_2-symmetric macrocyclic core. Concomitant cleavage of the two terminal TBS ethers was best effected with HCl/MeOH and gave diol **29**. Efforts were then directed to the challenging introduction of the two full side chains of rhizopodin in a bidirectional manner. Ultimately, the successful strategy involved a $Ba(OH)_2$-mediated coupling of the phosphonate **4** with

the bis-aldehyde derived from **29**,[27,28] followed by 1,4-reduction of the intermediate enone with Stryker's reagent[29,30] and selective cleavage of the primary TES ethers to furnish **30**. For the introduction of the sensitive *N*-vinylformamide units, we eventually found that **30** was best oxidized to the respective aldehyde with DMP before reaction with HNMe(CHO) and P_2O_5.[31] Since purification of the resulting fully protected natural product was tedious, the crude product was directly exposed to TBAF, thus giving rhizopodin (**1**) in 14% yield over three steps after workup and purification by reverse-phase HPLC. The spectroscopic data (^{1}H NMR, ^{13}C NMR) and specific rotation of our synthetic material were in agreement with those published for an isolated sample of rhizopodin, thus confirming the relative and absolute configuration of rhizopodin.

In conclusion, this first total synthesis of rhizopodin proceeds in 31 steps (longest linear sequence) and confirms unambiguously the relative and absolute configuration that was determined through our earlier stereochemical analysis[7] and with the X-ray structure of the actin complex reported by Schubert and co-workers.[8] Notable features include a convergent preparation of the oxazole rings from advanced fragments, a concise assembly of the macrocyclic core, involving a Suzuki macrocyclization of a highly elaborated substrate and a late stage introduction of the labile side chains in a bidirectional manner based on an HWE-coupling/hydrogenation sequence. In combination with the available structural data on actin-bound rhizopodin[8] and related macrolides,[4] the design of novel analogues with improved functional properties and/or simplified core structures can now be envisaged.

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[1] F. Sasse, H. Steinmetz, G. Höfle, H. Reichenbach, *J. Antibiot.* **1993**, *46*, 741.
[2] For recent reviews on the chemistry and biology of myxobacterial polyketides, see: a) D. Menche, *Nat. Prod. Rep.* **2008**, *25*, 905; b) K. J. Weissman, R. Müller, *Nat. Prod. Rep.* **2010**, *27*, 1276; c) M. Kretschmer, D. Menche, *Synlett* **2010**, 2989.
[3] T. M. A. Gronewold, F. Sasse, H. Lünsdorf, H. Reichenbach, *Cell Tissue Res.* **1999**, *295*, 121.
[4] For reviews on actin-binding macrolides, see: a) K.-S. Yeung, I. Paterson, *Angew. Chem.* **2002**, *114*, 4826; *Angew. Chem. Int. Ed.* **2002**, *41*, 4632; b) J. S. Allingham, V. A. Klenchin, I. Rayment, *Cell. Mol. Life Sci.* **2006**, *63*, 2119.
[5] N. Klippel, U. Bilitewski, *Anal. Lett.* **2007**, *40*, 1400.
[6] R. Jansen, H. Steinmetz, F. Sasse, W.-D. Schubert, G. Hagelüken, S. C. Albrecht, R. Müller, *Tetrahedron Lett.* **2008**, *49*, 5796.
[7] N. Horstmann, D. Menche, *Chem. Commun.* **2008**, *5173*.
[8] G. Hagelüken, G. Albrecht, R. Jansen, H. Steinmetz, D. Heinz, M. Kalesse, F. Sasse, W.-D. Schubert, *Angew. Chem.* **2009**, *121*, 603; *Angew. Chem. Int. Ed.* **2009**, *48*, 595.
[9] a) Z. Cheng, L. Song, Z. Xu, T. Ye, *Org. Lett.* **2010**, *12*, 2036; b) T. K. Chakraborty, K. K. Pulukuri, M. Sreekanth, *Tetrahedron Lett.* **2010**, *51*, 6444; c) T. K. Chakraborty, M. Sreekanth, K. K. Pulukuri, *Tetrahedron Lett.* **2011**, *52*, 59; d) K. C. Nicolaou, X. Jiang, P. J. Lindsay-Scott, A. Corbu, S. Yamashiro, A. Bacconi, V. M. Fowler, *Angew. Chem.* **2011**, *123*, 1171; *Angew. Chem. Int. Ed.* **2011**, *50*, 1139; e) M. Kretschmer, D. Menche, *Org. Lett.* **2012**, *14*, 382.
[10] a) S. Saito, T. Ishikawa, A. Kuroda, K. Koga, T. Moriwake, *Tetrahedron* **1992**, *48*, 4067; b) I. Paterson, C. De Savi, M. Tudge, *Org. Lett.* **2001**, *3*, 3149.
[11] a) I. V. Tararov, G. König, A. Börner, *Adv. Synth. Catal.* **2006**, *348*, 2633; b) A. Fürstner, M. Albert, J. Mlynarski, M. Matheu, E. DeClercq, *J. Am. Chem. Soc.* **2003**, *125*, 13132.
[12] a) M. Kitamura, M. Tokunaga, T. Ohkuma, R. Noyori, *Org. Synth.* **1993**, *71*, 1; b) M. Kitamura, M. Tokunaga, T. Ohkuma, R. Noyori, *Tetrahedron Lett.* **1991**, *32*, 4163.
[13] T. Okazoe, K. Takai, K. Utimoto, *J. Am. Chem. Soc.* **1987**, *109*, 951.
[14] R. I. Storer, T. Takemoto, P. S. Jackson, D. S. Brown, I. R. Baxendale, S. V. Ley, *Chem. Eur. J.* **2004**, *10*, 2529.
[15] Asymmetric Brown allylation of an aldehyde derived from mono-PMB-protected 2,2-dimethyl-1,3-propandiol proceeded in small scale in good selectivity (92% *ee*) and yield (71%). On large scale, removal of the chiral *iso*-pinocamphenyl ligand proved difficult.
[16] P. Wipf, C. P. Miller, *J. Org. Chem.* **1993**, *58*, 3604.
[17] D. A. Evans, K. T. Chapman, E. M. Carreira, *J. Am. Chem. Soc.* **1988**, *110*, 3560.
[18] I. Paterson, M. Donghi, K. Gerlach, *Angew. Chem.* **2000**, *112*, 3453; *Angew. Chem. Int. Ed.* **2000**, *39*, 3315.
[19] This sequence was necessary, because attempts to disconnect the PMB group from the fully formed macrocycle only led to complete decomposition. We tracked this behavior back to a comparable reactivity of the diene units. However, when the TBS-protected congener of **17** was used in the respective aldol reaction, separation of the TBS-protected analogues of **18** proved impossible.
[20] C. Morrill, R. H. Grubbs, *J. Org. Chem.* **2003**, *68*, 6031.
[21] A. Abiko, J.-F. Liu, S. Masamune, *J. Am. Chem. Soc.* **1997**, *119*, 2586.
[22] Attempts to remove the auxiliary from **24** directly with lithiated **25** were unsuccessful in this case, see: J. Li, D. Menche, *Synlett* **2009**, 2417.
[23] The TES-protected analogue of **23** was also employed as starting material to avoid the transformation of **26** to **4**. It turned out, however, that the yield in the first aldol reaction was significantly lower.
[24] N. Miyaura, A. Suzuki, *Chem. Rev.* **1995**, *95*, 2457.
[25] A. Gopalarathnam, S. G. Nelson, *Org. Lett.* **2006**, *8*, 7.
[26] J. Inanaga, K. Hirata, H. Saeki, T. Katsuki, M. Yamaguchi, *Bull. Chem. Soc. Jpn.* **1979**, *52*, 1989.
[27] I. Paterson, K.-S. Yeung, J. B. Smaill, *Synlett* **1993**, 774.
[28] Alternatively, the fully formed side chain incorporating the sensitive N-vinylformamide may be introduced by a boron-mediated aldol reaction of a methyl ketone equivalent to **4**. This sequence, however, proved less reliable, see: I. Paterson, K. Ashton, R. Britton, G. Cecere, G. Chouraqui, G. J. Florence, J. Stafford, *Angew. Chem.* **2007**, *119*, 6279; *Angew. Chem. Int. Ed.* **2007**, *46*, 6167.
[29] W. S. Mahoney, D. M. Brestensky, J. M. Stryker, *J. Am. Chem. Soc.* **1988**, *110*, 291.
[30] For a related strategy, see: Ref. [9d].
[31] a) I. Paterson, C. Cowden, C. Watson, *Synlett* **1996**, 209; b) I. Paterson, C. Watson, K.-S. Yeung, P. A. Wallace, R. A. Ward, *J. Org. Chem.* **1997**, *62*, 452.

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M. Dieckmann, M. Kretschmer, P. Li, S. Rudolph, D. Herkommer, D. Menche* _____ ■■■■–■■■■

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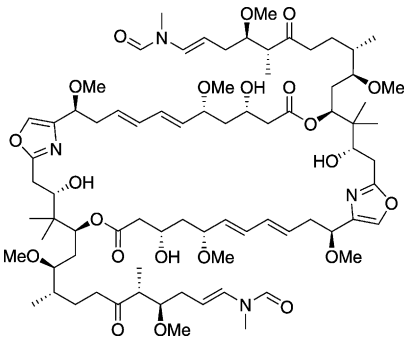

Symmetry helps: The total synthesis of the potent actin-targeting C_2-symmetric myxobacterial macrolide rhizopodin (see scheme) is accomplished by the convergent assembly of three building blocks of similar complexity, a concise macrocyclization strategy, and a late-stage introduction of the labile side chains.