Synthesis of the Apoptosis Inducing Agent Apoptolidin. Assembly of the C(16)–C(28) Fragment
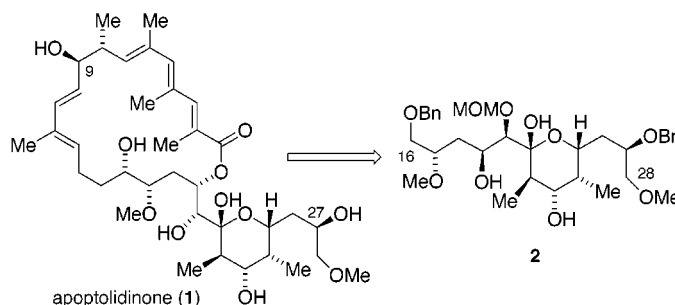
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ABSTRACT

A stereoselective synthesis of the C(16)–C(28) fragment of the apoptosis inducing agent apoptolidin is described. Key steps include two propionate aldol reactions and a stereoselective Mukaiyama aldol addition of enolsilane 19 to β-methoxy aldehyde 4.

In an effort to identify natural products with selective cytotoxicity against tumor cells, Hayakawa and co-workers established several immortalized cell lines in order to screen soil samples for specific apoptosis inducing agents.[1] Utilizing this screening method, the Hayakawa group reported in 1997 the identification of apoptolidin produced by an actinomycete identified as *Nocardiopsis* sp.[2] Significantly, apoptolidin was found to induce cell death in E1A transformed cells but not in normal cells. The two-dimensional structure of apoptolidin was reported in 1997 and the stereochemical assignment followed in 1998 based on extensive NMR analysis and degradation studies. The structure of apoptolidin consists of an aglycon (1) and two sugar units located at C(9) and C(27). The unique pattern of cytotoxicity and molecular architecture of apoptolidin stimulated our interest in this natural product

as a synthetic target.[3] In this Letter we describe the stereocontrolled assembly of the C(16)–C(28) fragment (2) of apoptolidin.

Apoptolidinone (1) consists of a 20-membered macrolactone incorporating a unique triene unit and 12 stereogenic centers, primarily located within the C(16)–C(28) fragment (Scheme 1). For the purpose of retrosynthetic analysis, the C(16)–C(28) substructure (2) is represented as the open-chain tautomer 3. Disconnection at the C(19)–C(20) carbon–carbon bond of 3 leads to aldehyde 4 and ketone 5. In the synthetic direction, a Mukaiyama aldol addition of an enolsilane derivative of 5 to β-alkoxy aldehyde 4 would proceed with the desired stereoselectivity due to the propensity of enolsilane additions to β-alkoxy aldehydes to occur with high *anti* 1,3-asymmetric induction.[4] Turning our attention to ketone 5, we required two consecutive *syn* propionate aldol reactions with opposite absolute stereo-

(1) (a) Hayakawa, Y.; Sohda, K. Y.; Furihata, K.; Kuzuyama, T.; ShinYa, K.; Seto, H. *J. Antibiot.* **1996**, *49*, 974–979. (b) Hayakawa, Y.; Sohda, K.; Shinya, K.; Hidaka, T.; Seto, H. *J. Antibiot.* **1995**, *48*, 954–961.

(2) (a) Hayakawa, Y.; Kim, J. W.; Adachi, H.; Shin-ya, K.; Fujita, K.-i.; Seto, H. *J. Am. Chem. Soc.* **1998**, *120*, 3524–3525. (b) Kim, J. W.; Adachi, H.; ShinYa, K.; Hayakawa, Y.; Seto, H. *J. Antibiot.* **1997**, *50*, 628–630.

(3) For recent synthetic studies, see: (a) Schuppan, J.; Ziemer, B.; Koert, U. *Tetrahedron Lett*. **2000**, *41*, 621–624. (b) Nicolaou, K. C.; Li, Y.; Weyershausen, B.; Weit, H.-x. *J. Chem. Soc., Chem. Commun.* **2000**, 307–308.
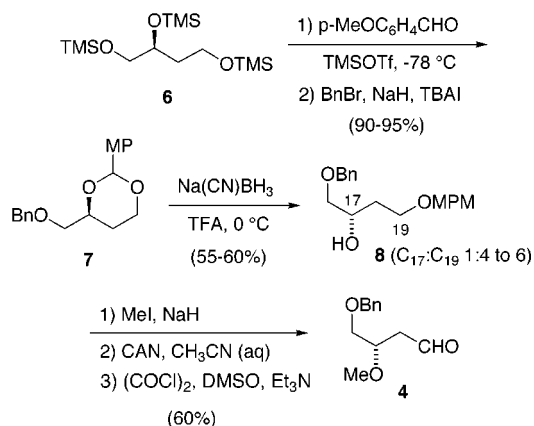
Scheme 1

apoptolidinone (**1**)

2

3 R=TBS

4

5 R' = TES; R = TBS
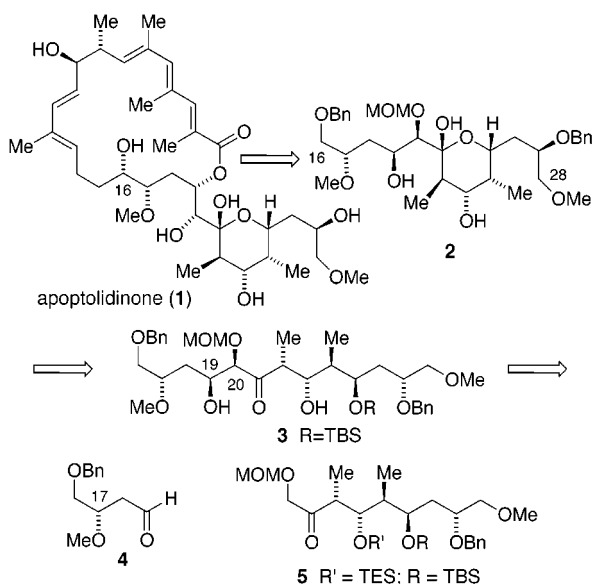
chemical control. In this case Crimmins' aldol methodology presented an attractive solution since this would require the preparation of a single acyloxazolidinethione in order to access both *syn* aldol isomers.[5]

Our first objective was the preparation of aldehyde **4** starting from known tris-trimethylsilyl ether **6**, readily derived from L-malic acid (Scheme 2).[6,7] Anisylidene formation under

Scheme 2

6

1) p-MeOC$_6$H$_4$CHO
TMSOTf, -78 °C
2) BnBr, NaH, TBAI
(90-95%)

7

Na(CN)BH$_3$
TFA, 0 °C
(55-60%)

8 (C$_{17}$:C$_{19}$ 1:4 to 6)

1) MeI, NaH
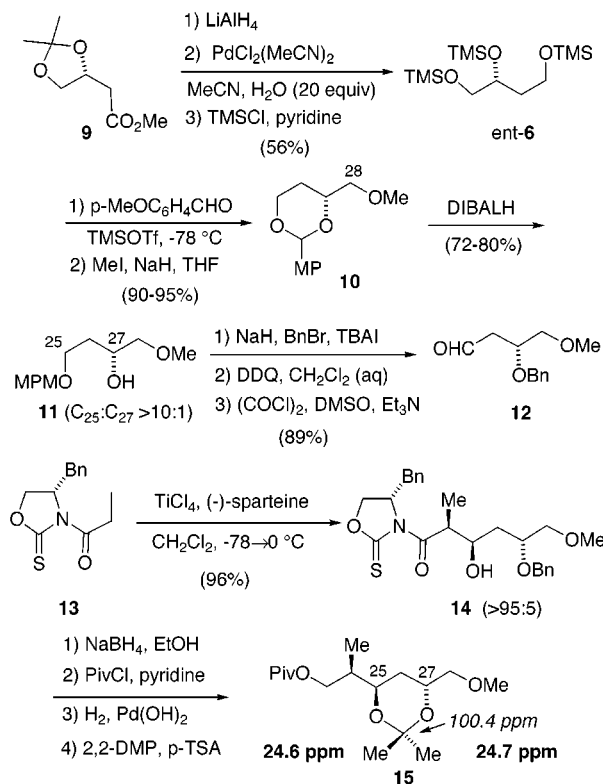2) CAN, CH$_3$CN (aq)
3) (COCl)$_2$, DMSO, Et$_3$N
(60%)

4

Noyori conditions followed by benzylation of the resulting primary alcohol gave 1,3-anisylidene **7** as the only isomer.[8,9]

(4) (a) Evans, D. A.; Yang, M. G.; Dart, M. J.; L.; Duffy, J. L.; Kim, A. S. *J. Am. Chem. Soc.* **1995**, *117*, 9598−9599. (b) Evans, D. A.; Dart, M. J.; Duffy, J. L.; Yang, M. G. *J. Am. Chem. Soc.* **1996**, *118*, 4322−4343. (c) Reetz, M. T.; Jung, A. *J. Am. Chem. Soc.* **1983**, *105*, 4833−4845.

(5) (a) Crimmins, M. T.; King, B. W.; Tabet, E. A. *J. Am. Chem. Soc.* **1997**, *119*, 7883−7884. (b) Crimmins, M. T.; Chaudhary, K. *Org. Lett.* **2000**, *2*, 775−777. (c) Yan, T.-H.; Tan, C.-W.; Lee, H.-C.; Lo, H.-C.; Huang, T.-Y. *J. Am. Chem. Soc.* **1993**, *115*, 2613−2621. (d) Nerz-Stormes, M.; Thorton, E. R. *J. Org. Chem.* **1992**, *56*, 2489−2498.

Reductive cleavage of **7** using Na(CN)BH$_3$ in trifluoroacetic acid afforded **8** as a ca. 1:5 mixture of C(17) and C(19) *p*-methoxyphenyl ethers which were readily separated by flash chromatography.[10] The major isomer was methylated, the *p*-methoxyphenyl group was removed, and the resulting alcohol was oxidized to complete the synthesis of aldehyde **4**.

The synthesis of aldehyde **12**, which can be viewed as a pseudo enantiomer of aldehyde **4**, started from methyl ester **9**, readily derived from L-ascorbic acid in six easily scalable steps (Scheme 3).[11] Reduction of **9** using LiAlH$_4$ in THF

Scheme 3

9

1) LiAlH$_4$
2) PdCl$_2$(MeCN)$_2$
MeCN, H$_2$O (20 equiv)
3) TMSCl, pyridine
(56%)

ent-**6**

1) p-MeOC$_6$H$_4$CHO
TMSOTf, -78 °C
2) MeI, NaH, THF
(90-95%)

10

DIBALH
(72-80%)

11 (C$_{25}$:C$_{27}$ >10:1)

1) NaH, BnBr, TBAI
2) DDQ, CH$_2$Cl$_2$ (aq)
3) (COCl)$_2$, DMSO, Et$_3$N
(89%)

12

13

TiCl$_4$, (-)-sparteine
CH$_2$Cl$_2$, -78→0 °C
(96%)

14 (>95:5)

1) NaBH$_4$, EtOH
2) PivCl, pyridine
3) H$_2$, Pd(OH)$_2$
4) 2,2-DMP, p-TSA

15

followed by acetonide removal employing palladium(II) catalysis and per-silylation provided ent-**6**.[12] Next, conversion to the 1,3-anisylidene followed by methylation of the remaining primary alcohol gave **10**. We found that reductive cleavage of **10** with DIBAL in dichloromethane resulted in selective protection of the C(25) alcohol as its *p*-methoxy-

(6) Kocienski, P. J.; Yeates, C.; Street, S. D. A.; Campbell, S. F. *J. Chem. Soc., Perkin Trans. 1* **1987**, 2183−2187.

(7) Sprung, M. M.; Nelson, L. S. *J. Org. Chem.* **1955**, *20*, 1750−1756.

(8) (a) Breuilles, P.; Oddon, G.; Uguen, D. *Tetrahedron Lett.* **1997**, *38*, 6607−6610. (b) Tsunoda, T.; Suzuki, M.; Noyori, R. *Tetrahedron Lett.* **1980**, *21*, 1357−1358.

(9) All new compounds gave characterization data that are fully consistent with the assigned structures.

(10) Johansson, R.; Samuelsson, B. *J. Chem. Soc., Perkin Trans. 1* **1984**, 2371−2374.

(11) (a) Wei, C. C.; Debernardo, S.; Tengi, J. P.; Borgese, J.; Weigele, M. *J. Org. Chem.* **1985**, *50*, 3462−3467. (b) Tanaka, A.; Yamashita, K. *Synthesis* **1987**, 570−573.

(12) Lipshutz, B. H.; Pollart, D.; Monforte, J.; Kotsuki, H. *Tetrahedron Lett.* **1985**, *26*, 705−708.

benzyl ether. A related contrasteric regioselective reduction of an anisylidene has been reported and attributed to a chelated intermediate.[13] In the current example, we attribute this regioselectivity to a chelating effect imposed by the neighboring C(28) methyl ether.[14] Finally, benzylation of the C(27) alcohol, removal of the *p*-methoxybenzyl group, and Swern oxidation provided aldehyde **12**. Aldol reaction between **12** and acyloxazolidinethione **13** using 1 equiv of TiCl₄ with (−)-sparteine furnished *syn* aldol **14** in 96% yield (>95:5 diastereoselectivity). The assigned *syn* aldol stereochemistry of the major isomer was consistent with the observed coupling constant between H(24) and H(25) which was 3.9 Hz.[15] Turning our attention to establishing the C(25)−C(27) relative stereochemistry, **14** was converted to acetonide **15** by the reaction sequence shown in Scheme 3 and the relative stereochemistry assigned on the basis of the Rychnovsky acetonide method.[16] Accordingly, the assigned C(25)−C(27) *anti* relative stereochemistry was consistent with the observed chemical shifts for the acetonide methyl groups and central carbon (cf. **15**).

In preparation for a second aldol reaction, silyl protection (TBSOTf, 2,6-lutidine) of **14** followed by NaBH₄ reduction and Swern oxidation gave aldehyde **16** in 69−80% overall yield (Scheme 4). Optimal conditions for effecting aldol reaction between **13** and **16**, in the desired non-Evans sense,
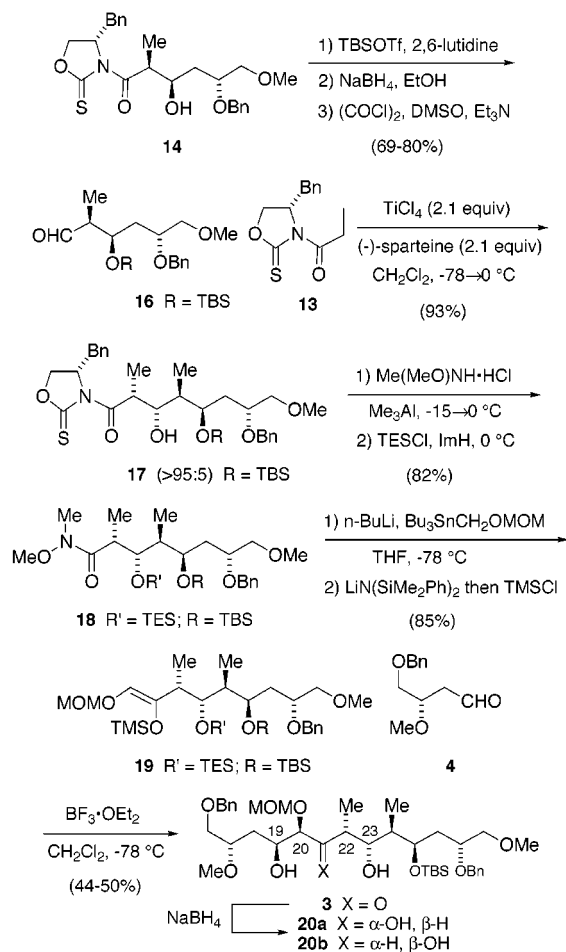
employed 2.1 equiv of TiCl₄ in combination with 2.1 equiv of (−)-sparteine to afford *syn* aldol adduct **17** in 93% yield and excellent diastereoselectivity.[17] The relative stereochemistry assignment of **17** was based on ¹³C NMR analysis of the appropriate acetonide derivative.[18] Exchange of the oxazolidinethione auxiliary with *N,O*-dimethylhydroxylamine gave the corresponding Weinreb amide which following silyl protection (TESCl, imidazole) of the remaining hydroxyl group produced **18**.[19] Addition of (methoxymethoxy)methyllithium, generated by the in situ transmetalation of (methyloxymethyoxy)methyltributylstannane, to **18** afforded the corresponding ketone.[20] During the course of investigations into double diastereoselective Mukaiyama aldol addition reactions, Evans observed that (*Z*)-enolsilanes favor 1,3-*anti*-dimethyl relationships across the emerging keto group as well as high levels of *anti* 1,3-asymmetric induction in Lewis acid promoted enolsilane additions to *β*-alkoxy aldehydes.[4] With these reports in mind, we prepared (*Z*)-enolsilane **19** using Masamune's base[21] and were delighted to observe a single aldol adduct upon Lewis acid promoted coupling of **4** and **19**. Simultaneously, with the formation of a newly formed carbon−carbon bond, we observed loss of the C(23) silyl protecting group. The aldol adduct (**3**) was assigned *syn* relative stereochemistry on the basis of an observed 3.3 Hz coupling constant between H(19) and H(20). Unambiguous assignment of the C(19)/C(20) and C(22)/C(23) relative stereochemistry was achieved using the [¹³C]acetonide analysis method.

Reduction of ketone **3** with NaBH₄ provided triols **20a** and **20b**; assignment of the relative configuration of C(19)−C(23) followed formation and ¹³C NMR analysis of the C(19)−C(21) and C(21)−C(23) acetonides. Treatment of **20a** with dimethoxypropane and PPTS afforded the C(19)−C(21) acetonide, assigned the *anti* configuration on the basis of ¹³C NMR acetonide analysis (Scheme 5). The corresponding

Scheme 4

Scheme 5
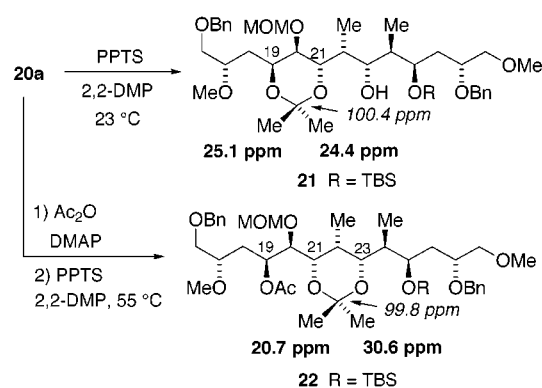
C(21)−C(23) acetonide **22** was derived from **20a** following acetylation of the C(19) hydroxyl group and assigned *syn* relative stereochemistry. Taken together, acetonides **21** and **22** confirm the assignment of the C(19)/C(20) and C(22)/C(23) relative stereochemistry as *anti*.

(13) Evans, D. A.; Kaldor, S. W.; Jones, T. K.; Clardy, J.; Stout, T. J. *J. Am. Chem. Soc.* **1990**, *112*, 7001−7031.

Having defined the full stereochemical assignment of **3**, we removed the C(25) TBS protecting group (TBAF, −10 °C) to give pyran **2** as a ca. 5:1 mixture of C(21) anomers (Scheme 6). The major anomer is assumed to possess the natural apoptolidin configuration.

Scheme 6
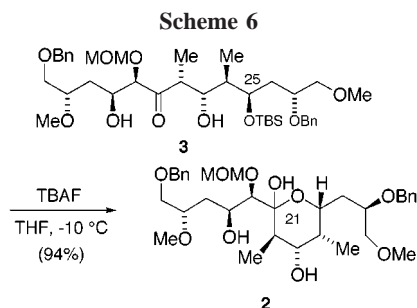
In summary, we have completed a stereocontrolled synthesis of the C(16)−C(28) fragment of apoptolidin as well as full stereochemical assignment based on the Rychnovsky acetonide method. Progress on the total synthesis of apoptolidin will be reported in due course.

(14) For a discussion of aluminum as a chelating Lewis acid, see: (a) Ooi, T.; Kagoshima, N.; Maruoka, K. *J. Am. Chem. Soc.* **1997**, *119*, 5754−5744. (b) Ooi, T.; Uraguchi, D.; N.; K.; Maruoka, K. *J. Am. Chem. Soc.* **1998**, *120*, 5327−5328.

Supporting Information Available: Spectroscopic and analytical data for compounds **2**−**4**, **6**−**8**, **10**−**12**, **14**−**19**, and **21**−**22** and proof of stereochemistry of **17**. This material is available free of charge via the Internet at http://pubs.acs.org.

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(15) Heathcock, C. H.; Pirrung, M. C.; Sohn, J. E. *J. Org. Chem.* **1979**, *44*, 4294−4299.

(16) (a) Rychnovsky, S. D.; Rogers, B. N.; Richardson, T. I. *Acc. Chem. Res.* **1998**, *31*, 9−17. (b) Evans, D. A.; Rieger, D. L.; Gage, J. R. *Tetrahedron Lett.* **1990**, *31*, 7099−7100.

(17) In Crimmins' original report (ref 5), Hunig's base provided superior selectivity in the non-Evans aldol reaction manifold; in the case of **16** we found that Hunig's base led to diminished chemical yield. In contrast (−)-sparteine provided both high stereoselectivity and chemical yield.

(18) See Supporting Information.

(19) (a) Basha, A.; Lipton, M.; Weibreb, S. M. *Tetrahedron Lett.* **1977**, 4171−4174. As in the case of oxazolidinones, the order of transamination and beta hydroxy protection is important, see: (b) Evans, D. A.; Gage, J. R.; Leighton, J. L. *J. Am. Chem. Soc.* **1992**, *114*, 9434−9453.

(20) Johnson, C. R.; Medich, J. R. *J. Org. Chem.* **1988**, *53*, 4131−4133.

(21) Masamune, S.; Ellingboe, J. W.; Choy, W. *J. Am. Chem. Soc.* **1982**, *104*, 5526−5528.