

Synthesis and Stereochemical Assignment of (+)-Chamuvarinin

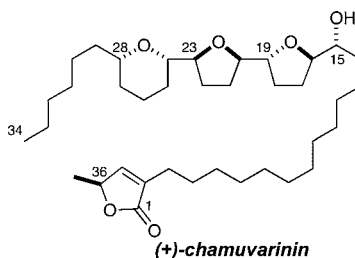
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ABSTRACT



A stereocontrolled total synthesis of (+)-chamuvarinin, isolated from the root extract of *Uvaria Chamae*, utilizes a convergent modular strategy to construct the adjacently linked C15–C28 ether array, followed by a late-stage Julia–Kocienski olefination to append the butenolide motif. This constitutes the first total synthesis of (+)-chamuvarinin, defining the relative and absolute configuration of this unique annonaceous acetogenin.

Isolated in 2004 by Laurens and co-workers from root extracts of the West African plant, *Uvaria chamae*, chamuvarinin (**1**, Scheme 1)^{1,2} is a unique member of the annonaceous acetogenin family of natural products.³ The crude extracts of *Uvaria chamae* are widely used in traditional medicinal practices for the treatment of a range of ailments including parasitic-borne West African sleeping sickness and has proven to be a rich source of novel acetogenins.⁴ An initial biological screening of chamuvarinin showed significant cytotoxicity toward KB 3-1 cervix cancer

cell lines ($IC_{50} = 0.8$ nM). Structurally, chamuvarinin is the first acetogenin to contain a tetrahydropyran (THP) ring linked adjacently to a bis-tetrahydrofuran (THF) ring system, spanning the C15–28 region of the carbon backbone. In common with the majority of acetogenins,³ **1** bears the 36S-configuration, but the assignment of the relative and absolute configuration within the C15–C28 region proved to be a more significant challenge. This quandary was partially resolved by Poupon and co-workers in 2007,² who showed that **1** is not derived directly from squamocin leading to the proposed relative configuration as shown,^{3,5} with two possible diastereomeric structures for the structure of **1**. Herein, we report the total synthesis of chamuvarinin and stereochemical assignment of **1**, utilizing a highly convergent strategy for the construction of the central C15–C28 region. We chose to pursue the 15R-diastereomeric series based on

(1) Fall, D.; Duval, R. A.; Gleye, C.; Laurens, A.; Hocquemiller, R. *J. Nat. Prod.* **2004**, *67*, 1041.

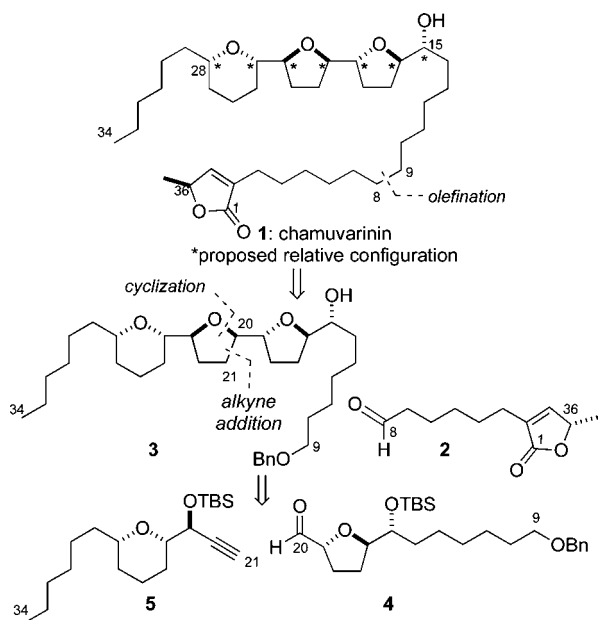
(2) Derbré, S.; Poupon, E.; Gleye, C.; Hocquemiller, R. *J. Nat. Prod.* **2004**, *70*, 300.

(3) (a) Bermejo, A.; Figadère, B.; Zafra-Polo, M.-C.; Barrachina, I.; Estornell, E.; Cortes, D. *Nat. Prod. Rep.* **2005**, *22*, 269. (b) Spurr, I. B.; Brown, R. C. D. *Molecules* **2010**, *15*, 460. (c) Alali, F. Q.; Liu, X.-X.; McLaughlin, J. L. *J. Nat. Prod.* **1999**, *62*, 504. (d) Zafra-Polo, M. C.; Figadère, B.; Gallardo, T.; Tormo, J. R.; Cortes, D. *Phytochemistry* **1998**, *48*, 1087. (e) Zafra-Polo, M. C.; González, M. C.; Estornell, E.; Sahpaz, S.; Cortes, D. *Phytochemistry* **1996**, *42*, 253. (f) Zeng, L.; Ye, Q.; Oberlies, N. H.; Shi, G.; Gu, Z.-M.; He, K.; McLaughlin, J. L. *Nat. Prod. Rep.* **1996**, *13*, 275. (g) Rupprecht, J. K.; Hui, Y. H.; McLaughlin, J. L. *J. Nat. Prod.* **1990**, *53*, 237.

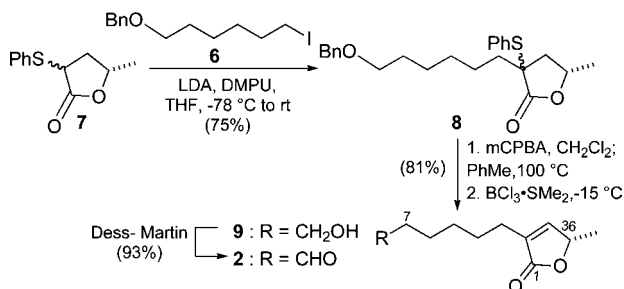
(4) (a) Fall, D.; Gleye, C.; Franck, X.; Laurens, A.; Hocquemiller, R. *Nat. Prod. Lett.* **2002**, *16*, 315. (b) Fall, D.; Gleye, C.; Laurens, A.; Hocquemiller, R. *Planta Med.* **2006**, *72*, 938.

(5) Sahai, M.; Singh, S.; Singh, M.; Gupta, Y. K.; Akashi, S.; Yuji, R.; Hirayama, K.; Asaki, H.; Araya, H.; Hara, N.; Eguchi, T.; Kakinuma, K.; Fujimoto, Y. *Chem. Pharm. Bull.* **1994**, *42*, 1163.

Scheme 1. Synthetic Strategy for Chamuvarinin 1



Scheme 2. Synthesis of C1–C8 Aldehyde 2



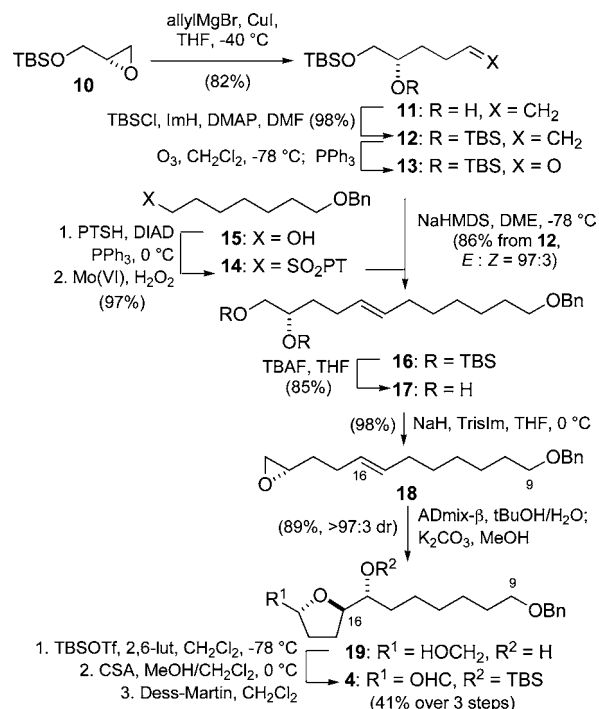
a comparison of acetogenins bearing a C15-carbinol linked to a bis-THF motif and the biosynthetic likelihood that **1** would share the common 15*R*-configuration.^{5,6} Our synthetic strategy relied on a late-stage olefination of the C1–C8 aldehyde **2** and a suitable coupling partner derived from the C9–C34 intermediate **3**, as outlined in Scheme 1. In turn, assembly of the central C15–C28 polyether array found in **3** would arise from the coupling of aldehyde (**4**, C9–C20) and alkyne (**5**, C21–C34), followed by reduction and cyclization to install the C20–C23 THF motif.

As outlined in Scheme 2, the synthesis of the C1–C8 aldehyde **2** started with the alkylation of iodide **6**⁷ with the lithium enolate of lactone **7** to provide **8** in 75% yield.^{8,9} Oxidation to the sulfoxide (mCPBA) and 1,2-syn elimination,

(6) (a) Hoye, T. R.; Hanson, P. R.; Kovelsky, A. C.; Ocain, T. D.; Zhuang, Z. *J. Am. Chem. Soc.* **1991**, *113*, 9369. (b) Hoye, T. R.; Tan, L. *Tetrahedron Lett.* **1995**, *36*, 1981. (c) Yang, H.; Zhang, N.; Li, X.; Chen, J.; Cai, B. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 2199.

(7) Iodide **6** was prepared from 1,6-hexanediol ((i) BnBr, NaH, DMF, 44%; (ii) I₂, PPh₃, ImH, Et₂O/MeCN (2:1), 88%); see: (a) Narayan, R. S.; Borhan, B. *J. Org. Chem.* **2006**, *71*, 1416. (b) Shimojo, M.; Matsumoto, K.; Hatanaka, M. *Tetrahedron* **2000**, *56*, 9281.

Scheme 3. Synthesis of C9–C20 Subunit 4^a



^a PTSH = 1H-mercaptophenyltetrazole; Mo(VI) = (NH₄)₆Mo₇O₂₄·4H₂O.

followed by cleavage of the C8-benzyl ether with BCl₃·SMe₂, provided alcohol **9** (81% over 2 steps).¹⁰ Finally, Dess–Martin oxidation of **9** provided the C1–C8 aldehyde **2** in excellent yield, as required for the final C8–C9 bond coupling.

As shown in Scheme 3, the synthesis of the C9–C20 subunit **4** began with the Cu(I)-promoted opening of (*S*)-TBS-glycidol ether **10** with allylmagnesium bromide to provide **11** (82%). TBS ether formation (TBSCl, ImH) gave **12** (98%). Ozonolysis and reductive PPh₃ workup of **12** provided aldehyde **13** which was used directly in the Julia–Kocienski olefination with sulfone **14**^{11,12} (readily prepared from alcohol **15**¹³ via Mitsunobu with DIAD, PPh₃, 1H-mercaptophenyltetrazole, and Mo(VI)/H₂O₂ oxidation). Treatment of sulfone **14** with NaHMDS in DME at -78 °C, followed by addition of aldehyde **13**, provided olefin **16** in 86% yield from **12** (*E/Z* = 97:3). The C16–C19 THF ring was conveniently installed by sequential application of

(8) Marshall, J. A.; Piettre, A.; Paige, M. A.; Valeriote, F. *J. Org. Chem.* **2003**, *68*, 1771.

(9) Lactone **7** was prepared from (*S*)-propylene oxide and (phenylthio) acetic acid ((i) *n*-BuLi, Pr₂NET, THF, -78 → rt; (ii) *p*-TsOH, PhMe, rt, 78%); see: White, J. D.; Somers, T. C.; Reddy, G. N. *J. Org. Chem.* **1992**, *57*, 4991.

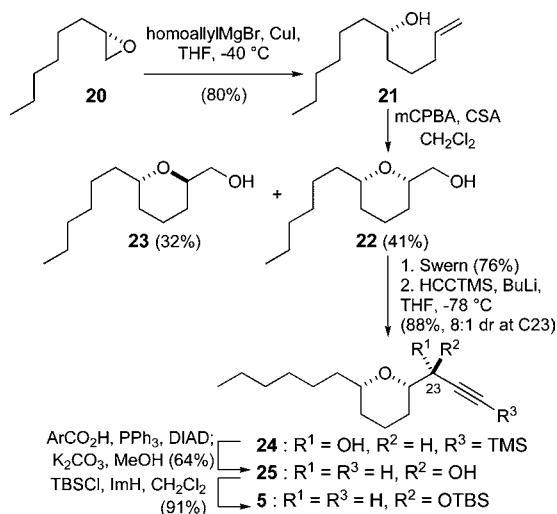
(10) Congreve, M. S.; Davidson, E. C.; Fuhry, M. A. M.; Holmes, A. B.; Payne, A. N.; Robinson, A.; Ward, S. E. *Synlett* **1993**, 663.

(11) Blakemore, P. R.; Cole, W. J.; Kocienski, P. J.; Morley, A. A. *Synlett* **1998**, 26.

(12) (a) Blakemore, P. R. *J. Chem. Soc., Perkin Trans. 1* **2002**, 2563. (b) Dumeunier, R. I.; Markó, E. *Modern Carbonyl Olefination*; Takeda, T., Ed.; WILEY-VCH: Weinheim, Germany, 2004; Chapter 3, p 104.

(13) Alcohol **15** was prepared from 1,7-heptanediol (BnBr, NaH, TBAF, THF, 69%); see: (a) Morimoto, Y.; Yokoe, C. *Tetrahedron Lett.* **1997**, *38*, 8981. (b) Morimoto, Y.; Yokoe, C.; Kurihara, H.; Kinoshita, T. *Tetrahedron* **1998**, *54*, 12197.

Scheme 4. Synthesis of C21–C34 Subunit 5^a



^a Ar = 4-BrC₆H₄.

protocols developed by Forsyth and co-workers.^{14,15} Cleavage of the TBS ethers in **16** with TBAF gave diol **17**, which was cleanly converted to epoxide **18** by treatment with NaH and TrisIm (98%).¹⁴ Subsequent asymmetric dihydroxylation of **18** (AD-mix- β),¹⁶ followed by base-mediated 5-exo cyclization (K₂CO₃, MeOH), provided the 2,5-*anti*-THF diol **19** in 89% yield with >97:3 dr.¹⁵ A three-step sequence was then required to complete the C9–C20 aldehyde **4**, involving TBS ether formation (TBSOTf, 2,6-lutidine), selective primary silyl cleavage, and Dess–Martin oxidation (41% over three steps). Using this approach the C9–C20 subunit was completed in ten steps and 21% overall yield from **10**.

As outlined in Scheme 4, the synthesis of the C21–C34 subunit **5** began with the opening of (*S*)-1-epoxyoctane **20**¹⁷ with homoallylmagnesium bromide¹⁸ and catalytic CuI to afford **21**. In light of the initial uncertainty around the substitution of the C24–C28 THP ring system in **1**,¹ we adopted a divergent approach to access both syn-**22** and anti-**23** from **21**. Thus, epoxidation of **21** with mCPBA, followed by addition of catalytic CSA (20 mol %) cleanly promoted the *in situ* 6-exo cyclization to provide readily separable THP alcohols **22** (41%) and **23** (32%). Oxidation of **22** and addition of the lithium anion of trimethylsilylacetylene to the intermediate aldehyde provided **24** with the undesired

23*R* configuration in 88% yield (8:1 dr).¹⁹ The minor 23*S* diastereomer proved readily separable by column chromatography, and Mitsunobu inversion of the C23-hydroxyl in **24**,²⁰ followed by base-mediated methanolysis/desilylation (K₂CO₃, MeOH), gave **25** (64%) with the requisite 23*S* configuration. TBS protection (TBSCl/ImH, 91%) then completed the C21–C34 subunit **5** in six steps and 11% overall yield.

With our key subunits in hand our attention turned to the C20–C21 bond construction and the installation of the central C20–C23 syn-substituted THF motif. In practice, treatment of **5** with *n*-BuLi, followed by addition of the resulting lithium anion to **4**, gave the expected Felkin–Anh adduct **26** in 71% yield with good levels of stereocontrol at C20 (3:1 dr). Diimide reduction of **26** with TsNHNH₂ and NaOAc in DME at reflux proved the most efficient reduction protocol to afford **27** in 79% yield.²¹ At this point the C20 diastereomers were readily separated by column chromatography.¹⁹ The stage was now set for the construction of the central C20–C23 THF ring. In the event the C20-hydroxyl in **27** was readily activated as its mesylate (MsCl, Et₃N), which upon treatment with TBAF promoted silyl ether cleavage and concomitant 5-exo ring closure to provide **3** in excellent yield. At this point, comparison of the ¹H and ¹³C NMR spectra of the advanced C9–C34 intermediate **3** with the reported data for the C15–C28 region of chamuvarinin showed them to be essentially in complete agreement,^{1,2} providing us with the first clear indication of the correct stereochemical assignment of the relative configuration of the C15–C28 region of chamuvarinin.

With the advanced intermediate **3** in hand, attachment of a suitable C1–C8 subunit at C9 was required to facilitate the completion of chamuvarinin. In considering a suitable C8–C9 coupling reaction, we initially focused on using a C9–C34 aldehyde for olefination with either a C1–C8 Wittig salt or sulfone derived from **9**; however this approach proved unsuccessful. As a result we employed the reversed coupling strategy detailed in Scheme 5. Thus, the C15-OH in **3** was readily protected as its TBS ether (TBSOTf, 2,6-lutidine, 88%) and subsequent hydrogenolysis of **28** provided alcohol **29** in excellent yield. Treatment of **29** with 1*H*-mercaptophenyltetrazole under Mitsunobu conditions (99%) and subsequent oxidation of the intermediate sulfide (H₂O₂, cat Mo(VI), 77%) provided the corresponding sulfone **30**, in readiness for the final C8–C9 Julia–Kocienski olefination.^{11,12,22} In the event, treatment of sulfone **30** with

(14) (a) Cink, R. D.; Forsyth, C. J. *J. Org. Chem.* **1995**, *60*, 8122. (b) Corey, E. J.; Weigel, L. O.; Chamberlin, A. R.; Lipshutz, B. *J. Am. Chem. Soc.* **1980**, *102*, 1439.

(15) (a) Dounay, A. B.; Florence, G. J.; Saito, A.; Forsyth, C. J. *Tetrahedron* **2002**, *58*, 1865. (b) Dounay, A. B.; Forsyth, C. J. *Org. Lett.* **2001**, *3*, 975.

(16) Kolb, H. C.; VanNieuwenhze, M. S.; Sharpless, K. B. *Chem. Rev.* **1994**, *94*, 2483.

(17) Enantioenriched (*S*)-1-epoxyoctane **20** was prepared by hydrolytic kinetic resolution of (\pm)-epoxyoctane in 41% yield with \geq 95% ee; see: (a) Paddon-Jones, G. C.; McErlean, C. S. P.; Hayes, P.; Moore, C. J.; Konig, W. A.; Kitching, W. J. *Org. Chem.* **2001**, *66*, 7487. (b) Schaus, S. E.; Brandes, A. D.; Larrow, J. F.; Tokunga, M.; Hansen, K. B.; Gould, A. E.; Furrow, M. E.; Jacobsen, E. N. *J. Am. Chem. Soc.* **2002**, *124*, 1307.

(18) Français, A.; Bedel, O.; Picoul, W.; Meddour, A.; Courtien, J.; Haudrechy, A. *Tetrahedron: Asymmetry* **2005**, *16*, 1141.

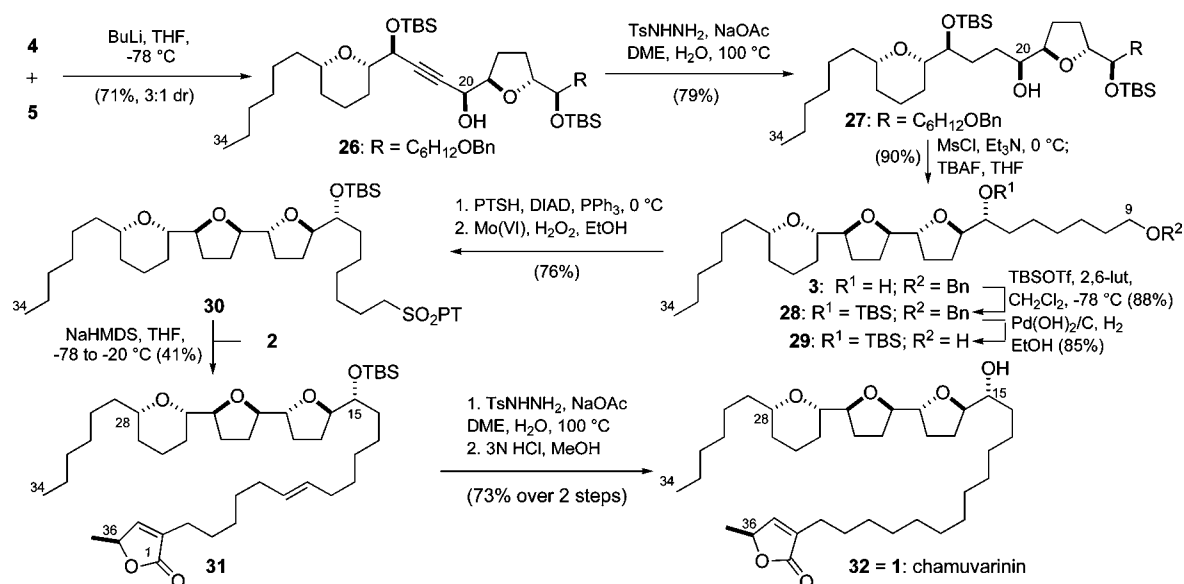
(19) The configurations of the carbinol stereocenters at C23 in **24** and at C20 in **27** were assigned by advanced Mosher ester analysis. (a) Ohtani, I.; Kusumi, T.; Kashman, Y.; Kakisawa, H. *J. Am. Chem. Soc.* **1991**, *113*, 4092. (b) Kusumi, T.; Hamada, T.; Ishitsuka, M. O.; Ohtani, I.; Kakisawa, H. *J. Org. Chem.* **1992**, *57*, 1033. (c) Sullivan, J. R.; Dale, J. A.; Mosher, H. S. *J. Org. Chem.* **1973**, *38*, 2143. (d) Hoye, T. R.; Jeffrey, C. S.; Shao, F. *Nat. Protoc.* **2007**, *2*, 2451.

(20) For a review of the Mitsunobu reaction, see: Swamy, K. C. K.; Kumar, N. N. B.; Balaraman, E.; Kumar, K. V. P. *Chem. Rev.* **2009**, *109*, 2551.

(21) (a) Marshall, J. A.; Chen, M. *J. Org. Chem.* **1997**, *62*, 5996. (b) Crimmins, M. T.; Zhang, Y.; Diaz, F. A. *Org. Lett.* **2006**, *8*, 2369.

(22) (a) Wilcox, C. S.; Gudipati, V.; Lu, H.; Turkyilmaz, S.; Curran, D. P. *Angew. Chem., Int. Ed.* **2005**, *44*, 6938. (b) Curran, D. P.; Zhang, Q.; Richard, C.; Lu, H.; Gudipati, V.; Wilcox, C. S. *J. Am. Chem. Soc.* **2006**, *128*, 9561.

Scheme 5. Completion of Chamuvarinin^a



^a PTSH = 1H-mercaptophenyltetrazole; Mo(VI) = (NH₄)₆Mo₇O₂₄·4H₂O.

NaHMDS in THF at $-78\text{ }^{\circ}\text{C}$ followed by the addition of aldehyde **2** and warming to $-20\text{ }^{\circ}\text{C}$ over 4 h gave the advanced intermediate **31** in 41% yield. Diimide reduction of the C8–C9 alkene (TsNHNH₂/NaOAc in DME/H₂O),²¹ followed by deprotection of the C15-TBS ether (3 N HCl, MeOH), provided the 15*R*, 16*R*, 19*R*, 20*R*, 23*S*, 24*S*, 28*S*, 36*S*-diastereomer **32** in excellent yield over two steps. Gratifyingly, the spectroscopic data obtained for **32** (¹H and ¹³C NMR, IR, and MS) correlated fully with that of natural chamuvarinin.¹ In the absence of an authentic sample for direct specific rotation and HPLC comparison, the measured specific rotation $[\alpha]_{\text{D}}^{20} +9.9$ (*c* 0.1, CHCl₃) [lit.¹ +25 (*c* 0.026, CHCl₃)] was consistent with that of the natural material. Further convincing evidence for **32** being the correct stereostructure of **1** is provided by the comparable levels of biological activity displayed by the synthetic material. In screening assays against the HeLa cervix cancer cell line and the bloodstream form of the parasite *Trypanosoma brucei*, the synthetic material displayed ED₅₀ values of 2.88 ± 0.66 and $1.37 \pm 0.08\ \mu\text{M}$, respectively,²³ providing unambiguous proof of the relative and absolute configuration of (+)-chamuvarinin (**1**), as that being indicated in structure **32**.

(23) The trypanocidal activity of **32** was tested against cultured bloodstream *T. brucei* (strain 427), and their cytotoxicity against HeLa cells was determined using the Alamar Blue viability test, as described in: Mikus, J.; Steverding, D. *Parasitol. Int.* **2000**, *48*, 265.

In conclusion, we have resolved the stereochemical ambiguity surrounding the structure of chamuvarinin by completing the first total synthesis (20 longest linear steps, 1.4% overall yield) and enabling further biological studies. The present work demonstrates the versatility of our modular alkyne coupling/cyclization strategy to construct adjacently linked acetogenin frameworks in an efficient manner to provide synthetic access to these rare bioactive metabolites and for establishing their full stereochemistry.

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Supporting Information Available: Experimental procedures and spectroscopic data for new compounds and copies of ¹H and ¹³C NMR spectra for synthetic and natural chamuvarinin. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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