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Synthesis and Structure Revision of Nakiterpiosin

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Abstract

This manuscript describes a convergent synthesis and the revision of the relative stereochemistry of nakiterpiosin, a marine C-nor-D-homosteroid. Our synthesis features a late-stage carbonylative Stille cross-coupling reaction and a photo-Nazarov cyclization reaction that deliver the complete nakiterpiosin skeleton efficiently.

Nakiterpiosin (**1**) is a marine sponge metabolite that exhibits potent cytotoxicity against the P388 murine leukemia cell line (GI₅₀ 10 ng/mL) (Figure 1).¹ It was the first C-nor-D-homosteroid isolated from a marine source. Its unique chemical structure and strong P388 growth inhibition property prompted us to initiate a research program to explore its laboratory synthesis² and biological function. We report herein the synthesis and structure revision³ of nakiterpiosin.

The C-nor-D-homosteroids are skeletally rearranged steroids with their C-ring contracted and D-ring expanded by one carbon. The veratrum alkaloid cyclopamine (**3**) and veratramine (**4**) are arguably the best known members.^{4,5} The teratogenic alkaloid **3** inhibits the Hedgehog (Hh) signaling by binding to Smoothened (Smo),^{6,7} and the chronotropic alkaloid **4** induces serotonin (5-HT) syndrome.⁸ While **1** possesses potent cytotoxicity against P388, its molecular target is not known. Furthermore, the complete biological profile of **1** could not be obtained due to the scarcity of the material. From 30 kg of sponge *T. hoshinota*, only 0.4 mg of nakiterpiosin was obtained. Its chemical structure was assigned as **2** in the original reports.¹

We were puzzled by the inconsistency between the C-20 stereochemistry reported for **2** and **3/4** and therefore set out to probe the relative stereochemistry of nakiterpiosin. Our model studies indicated potential misassignment of the C-6, C-20 and C-25 stereogenic centers.⁹ We next considered the biogenesis of the halogen atoms¹⁰ of nakiterpiosin to rationalize the C-6 and C-20 stereochemistry. We envisioned that the C-21 chlorine atoms of nakiterpiosin might be introduced by radical chlorination and the C-6 bromine atom by bromoetherification (as shown in **5**) to result in retention of the C-20 configuration and the *anti* C-5,6 bromohydrin stereochemistry. Taken together, these considerations led us to propose **1** to be the correct structure of nakiterpiosin. Indeed, we found that the ¹H and ¹³C NMR spectra of our synthetic sample of **1** agreed with those of the natural product.¹¹ In contrast, those of synthetic **2**,¹¹ and the natural product are significantly different. We thus revised the relative stereochemistry

of nakiterpiosin to be that indicated in **1**, which shares the same configuration at the C-20 and C-25 positions with **3** and **4**.

Our synthetic strategy is outlined in Figure 2. We dissected **1** into fragments **6** and **7** and constructed the central cyclopentanone ring with a carbonylative cross-coupling reaction¹² and a photo-Nazarov cyclization reaction.¹³ The electrophilic coupling component **6** was synthesized by an intramolecular Diels–Alder reaction,¹⁴ and the nucleophilic coupling component **7** by vinylogous Mukaiyama aldol reaction.¹⁵

To synthesize **6**, we first converted acid **8**¹⁶ to the corresponding Weinreb amide and then set the C-6 stereocenter by Noyori reduction¹⁷ (Scheme 1). The in-water hydrogenation protocol¹⁸ provided significant enhancement of the reaction rate, allowed low catalyst loading and suppressed the formation of the undesired lactone. Subsequently, isopropenyl Grignard addition to **9** followed by Me₂AlCl-promoted intramolecular Diels–Alder reaction gave the exo cycloaddition product. The C-6 hydroxyl group controlled the stereoselectivity¹⁹ of this Diels–Alder reaction to afford single diastereomer. The C-6 hydroxyl group was then activated with an electron-deficient aryl sulfonate group for the introduction of the C-6 bromine atom. We found that **10** underwent retro-Diels–Alder reaction readily at elevated temperature, particularly in the presence of Lewis acids. This characteristic imposed significant challenges for the introduction of the C-6 bromine atom. We therefore dihydroxylated the olefin group to prevent the retro-Diels–Alder reaction even through this functionalization created serious steric congestion around the C-6 position. The C-6 bromine atom could then be introduced with inversion of configuration and concomitant acetonide protection. The absolute and relative configurations of **11** were confirmed by X-ray analysis.¹¹ Finally, enol triflate installation completed the synthesis of **6**.

For the synthesis of **7**, we utilized the Sharpless asymmetric epoxidation reaction²⁰ of **12**¹¹ to set the C-20 stereochemistry (Scheme 2). The subsequent TBS protection and Yamamoto epoxide rearrangement reaction²¹ gave aldehyde **13**. We note that aldehyde **13** was sensitive to both acidic and basic conditions and underwent elimination and racemization readily. The enantiomeric purity was eroded after the rearrangement reaction. We also found that Bi(OTf)₃ promoted the vinylogous Mukaiyama aldol reaction of **13** with good levels of diastereoselectivity.²² However, further loss of enantiomeric purity could not be avoided. With the complete C-20–C-26 carbon framework of the side-chain in place, we then sought to set its *anti–anti–trans* configuration. The C-25 stereochemistry was set by a chelation-controlled hydrogenation of **14** using Crabtree's catalyst.^{23,24} We then inverted the C-22 stereocenter of **15** and protected the hydroxyl group as TBS ether to give **16** with the desired *anti–anti–trans* configuration. Selective deprotection of the primary TBS ether followed by Dess–Martin oxidation yielded aldehyde **17**. The *gem*-dichloromethyl group was introduced by Cl₂/P(OPh)₃ to give **18**.²⁵ No epimerization of the C-20 stereocenter was observed. The X-ray analysis on des-TBS-**18** confirmed the absolute and relative configurations of **18**.¹¹ Finally, the palladium-catalyzed stannylation afforded the nucleophilic coupling component **7**.

The carbonylative coupling of **6** and **7** was achieved with a modified Stille's protocol²⁶ using Pd(PPh₃)₄/CuCl in DMSO under 1 atm CO. The CuCl additive and DMSO solvent provided dramatic rate enhancement²⁷ and were crucial to the success of this reaction. It is worth noting that **6** and **19** were highly sensitive to both acidic and basic reaction conditions. Addition of LiCl and prolonged heating led to the elimination of bromide. Enone **19** was obtained as a 4:1 mixture of inseparable diastereomers due to the diminished enantiomeric purity of **7**. Nonetheless, the minor diastereomer could be removed at a later stage (**20**).

With **19** in hand, we next explored the key Nazarov cyclization reaction to complete the construction of the C-nor-D-homosteroid skeleton of nakiterpiosin. Remarkably, irradiation of

a solution of **19** in acetonitrile at 350 nm smoothly delivered the desired annulation product as a 1:1 mixture of C-9 diastereomers, which converged to **20** upon treating with diisopropylamine in methanol. The photo-Nazarov cyclization reaction of aryl vinyl ketones was first reported by Smith and Agosta.²⁸ Subsequent mechanistic studies by Leitich and Schaffner revealed the reaction mechanism to be thermal electrocyclozation induced by photolytic enone isomerization.²⁹ The mildness of the reaction conditions and the selective activation of the enone functional group allowed the facile transformation of the densely functionalized **19** to **20**. It should be noted that the Lewis acid-promoted Nazarov cyclization of aryl vinyl ketones normally requires much harsher reaction conditions or activated substrates.³⁰ Indeed, exposure of the model systems of **19** with simplified side-chains to various Lewis acids only resulted in substrate decomposition.

To complete the synthesis of **1**, we first removed the acetonide protecting group of **20** and cleaved the diol to afford the corresponding bis-hemiacetal. Selective reduction of the less hindered hemiacetal followed by TBS deprotection³¹ furnished **1**. The spectroscopic data of **1** is fully consistent with that of the natural sample.¹¹ In contrast, synthetic **2**, which was obtained by a similar approach,^{9,11} exhibits significantly different ¹H and ¹³C NMR spectra. Notably, synthetic **2** existed as an equilibrium mixture of the C-4 hemiacetal and aldehyde forms.¹¹ We have also compared the chemical shifts of the Mosher esters of synthetic **1** with those reported for the natural product and found that they were in agreement.^{11,32}

In conclusion, the structure of nakiterpiosin is revised as **1**. The newly assigned structure is supported by total synthesis and biogenesis rationale. Our synthetic approach is highly convergent and allows for the synthesis of the analogs and derivatives of **1** for further biological studies. We are investigating the biological function and molecular target of **1**. These results will be reported in due course.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. (a) Teruya T, Nakagawa S, Koyama T, Arimoto H, Kita M, Uemura D. *Tetrahedron* 2004;60:6989–6993. (b) Teruya T, Nakagawa S, Koyama T, Suenaga K, Kita M, Uemura D. *Tetrahedron Lett* 2003;44:5171–5173.
2. For a report of the synthetic studies toward nakiterpiosin, see: Ito T, Ito M, Arimoto H, Takamura H, Uemura D. *Tetrahedron* 2007;48:5465–5469.
3. For a review on structure revisions of natural products through synthetic studies, see: Nicolaou KC, Snyder SA. *Angew Chem Int Ed* 2005;44:1012–1044.
4. Cycloamine is also known as 11-deoxojervine. For the seminal semi-synthesis of jervine and veratramine, see: (a) Masamune T, Takasugi M, Murai A, Kobayashi K. *J Am Chem Soc* 1967;89:4521–4523. (b) Johnson WS, deJongh HAP, Coverdale CE, Scott JW, Burckhardt U. *J Am Chem Soc* 1967;89:4523–4524. (c) Masamune T, Takasugi M, Murai A. *Tetrahedron* 1971;27:3369–3386. (d) Kutney JP, Cable J, Gladstone WAF, Hanssen HW, Nair GV, Torupka EJ, Warnock WDC. *Can J Chem* 1976;53:1796–1817.

5. For reviews, see: (a) McFerren MA. *J Am Acad Dermatol* 2006;54:718–720. [PubMed: 16546599] (b) Keeler RF. *Lipids* 1978;13:708–715. [PubMed: 723484] (c) James LF. *J Nat Toxins* 1999;8:63–80. [PubMed: 10091129] (d) Binns W, James LF, Keeler RF, Balls LD. *Cancer Res* 1968;28:2323–2326. [PubMed: 5749423]
6. (a) Cooper MK, Porter JA, Young KE, Beachy PA. *Science* 1998;280:1603–1607. [PubMed: 9616123] (b) Taipale J, Chen JK, Cooper MK, Wang B, Mann RK, Milenkovic L, Scott MP, Beachy PA. *Nature* 2000;406:1005–1009. [PubMed: 10984056] (c) Chen JK, Taipale J, Cooper MK, Beachy PA. *Gen Dev* 2002;16:2743–2748.
7. For clinical potentials of the Hh antagonists, see: (a) Yauch RL, Gould SE, Scales SJ, Tang T, Tian H, Ahn CP, Marshall D, Fu L, Januario T, Kallop D, Nannini-Pepe M, Kotkow K, Marsters JC, Rubin LL, de Sauvage FJ. *Nature* 2008;455:406–410. [PubMed: 18754008] (b) van den Brink GR. *Physiol Rev* 2007;87:1343–1375. [PubMed: 17928586] (c) Borzillo GV, Lippa B. *Curr Top Med Chem* 2005;147–157. [PubMed: 15853643] (d) Lum L, Beachy PA. *Science* 2004;304:1755–1759. [PubMed: 15205520]
8. (a) Thron CD, McCann FV. *Gen Pharmac* 1999;32:81–89. (b) Nagata R, Izumi K. *Jpn J Pharmacol* 1991;55:129–137. [PubMed: 1710297] (c) Nagata R, Izumi K, Iwata Si, Shimizu T, Fukuda T. *Jpn J Pharmacol* 1991;55:139–146. [PubMed: 1710298]
9. Details will be reported in a full account.
10. For reviews, see: (a) Neumann CS, Fujimori DG, Walsh CT. *Chem Biol* 2008;15:99–109. [PubMed: 18291314] (b) Vaillancourt FH, Yeh E, Vosburg DA, Garneau-Tsodikova S, Walsh CT. *Chem Rev* 2006;106:3364–3378. [PubMed: 16895332]
11. See Supporting Information for further information.
12. For reviews on palladium-catalyzed cross-coupling reactions in natural product synthesis, see: (a) Nicolaou KC, Bulger PG, Sarlah D. *Angew Chem Int Ed* 2005;44:4442–4489. For review on diaryl ketone synthesis through carbonylative coupling, see: (b) Brunet JJ, Chauvin R. *Chem Soc Rev* 1995;24:89–95. For a leading reference on the carbonylative cross-coupling reaction, see: (c) Wang Q, Chen C. *Tetrahedron Lett* 2008;49:2916–2921.
13. For reviews, see: (a) Frontier AJ, Collison C. *Tetrahedron* 2005;61:7577–7606. (b) Pellissier H. *Tetrahedron* 2005;61:6479–6517. (c) Tius MA. *Eur J Org Chem* 2005:2193–2206. (d) Habermas KL, Denmark SE, Jones TK. *Org React* 1994;45:1–158.
14. For reviews, see: (a) Takao, Ki; Munakata, R.; Tadano, Ki. *Chem Rev* 2005;105:4779–4807. [PubMed: 16351062] (b) Roush WR. *Advances in Cycloaddition* Curran DP. Jai Press Greenwich 1990;29:1146 (c) Craig D. *Chem Soc Rev* 1987;16:187–238.
15. For reviews, see: (a) Denmark SE, Heemstra J, John R, Beutner GL. *Angew Chem Int Ed* 2005;44:4682–4698. (b) Casiraghi G, Zanardi F, Appendino G, Rassu G. *Chem Rev* 2000;100:1929–1972. [PubMed: 11749280] (c) Rassu G, Zanardi F, Battistini L, Casiraghi G. *Chem Soc Rev* 2000;29:109–118.
16. Obtained in one step by treating furan and succinic anhydride with AlCl_3 in CH_2Cl_2 according to the literature: Chen L, Yin Bl, Xu Hh, Chiu Mh, Wu Yl. *Chin J Chem* 2004;22:92–99.
17. (a) Fujii A, Hashiguchi S, Uematsu N, Ikariya T, Noyori R. *J Am Chem Soc* 1996;118:2521–2522. (b) Hashiguchi S, Fujii A, Takehara J, Ikariya T, Noyori R. *J Am Chem Soc* 1995;117:7562–7563. (c) Ohkuma T, Utsumi N, Tsutsumi K, Murata K, Sandoval C, Noyori R. *J Am Chem Soc* 2006;128:8724–8725. [PubMed: 16819854]
18. (a) Wu X, Li X, Hems W, King F, Xiao J. *Org Biomol Chem* 2004;2:1818–1821. [PubMed: 15227530] (b) Wu X, Li X, King F, Xiao J. *Angew Chem Int Ed* 2005;44:3407–3411. (c) Wu X, Li X, Zanotti-Gerosa A, Pettman A, Liu J, Mills AJ, Xiao J. *Chem Eur J* 2008;14:2209–2222.
19. For the pioneering work on controlling the stereoselectivity of the intramolecular Diels-Alder reaction using a stereogenic center on the tether, see: (a) Boeckman RK, Barta TE. *J Org Chem* 1985;50:3421–3423. (b) Roush WR, Kageyama M, Riva R, Brown BB, Warmus JS, Moriarty KJ. *J Org Chem* 1991;56:1191–1220.
20. (a) Katsuki T, Martin VS. *Org React* 1996;48:1–300. (b) Katsuki T, Sharpless KB. *J Am Chem Soc* 1980;102:5974–5976. (c) Gao Y, Klunder JM, Hanson RM, Masamune H, Ko SY, Sharpless KB. *J Am Chem Soc* 1987;109:5765–5780.

21. (a) Maruoka K, Ooi T, Yamamoto H. *J Am Chem Soc* 1989;111:6431–6432. (b) Maruoka K, Ooi T, Nagahara S, Yamamoto H. *Tetrahedron* 1991;47:6983–6998. (c) Suda K, Kikkawa T, Nakajima Si, Takanami T. *J Am Chem Soc* 2004;126:9554–9555. [PubMed: 15291555]
22. Ollevier T, Bouchard JE, Desyroy V. *J Org Chem* 2008;73:331–334. [PubMed: 18069852]
23. (a) Crabtree R. *Acc Chem Res* 1979;12:331–337. (b) Crabtree RH, Davis MW. *Organometallics* 1983;2:681–682.
24. Evans DA, Dunn TB, Kværnø L, Beauchemin A, Raymer B, Olhava EJ, Mulder JA, Juhl M, Kagechika K, Favor DA. *Angew Chem Int Ed* 2007;46:4698–4703.
25. (a) Spaggiari A, Vaccari D, Davoli P, Torre G, Prati F. *J Org Chem* 2007;72:2216–2219. [PubMed: 17295542] (b) Hoffmann RW, Bovicelli P. *Synthesis* 1990:657–659.
26. (a) Baillargeon VP, Stille JK. *J Am Chem Soc* 1983;105:7175–7176. (b) Goure WF, Wright ME, Davis PD, Labadie SS, Stille JK. *J Am Chem Soc* 1984;106:6417–6422. (c) Crisp GT, Scott WJ, Stille JK. *J Am Chem Soc* 1984;106:7500–7506. (d) Echavarrent AM, Stille JK. *J Am Chem Soc* 1988;110:1557–1565.
27. Han X, Stoltz BM, Corey EJ. *J Am Chem Soc* 1999;121:7600–7605.
28. Smith AB III, Agosta WC. *J Am Chem Soc* 1973;95:1961–1968.
29. (a) Leitich J, Heise I, Werner S, Krüger C, Schaffner K. *J Photochem Photobiol A: Chem* 1991;57:127–151. (b) Leitich J, Heise I, Jürgen R, Schaffner K. *Eur J Org Chem* 2001:2719–2726.
30. (a) Liang G, Xu Y, Seiple IB, Trauner D. *J Am Chem Soc* 2006;128:11022–11023. [PubMed: 16925409] (b) He W, Herrick IR, Atesin TA, Caruana PA, Kellenberger CA, Frontier AJ. *J Am Chem Soc* 2008;130:1003–1011. [PubMed: 18161974] (c) Marcus AP, Lee AS, Davis RL, Tantillo DJ, Sarpong R. *Angew Chem Int Ed* 2008;47:6379–6383.
31. For a convenient workup method, see: Kaburagi Y, Kishi Y. *Org Lett* 2007;9:723–726. [PubMed: 17286380]
32. The optical rotation of the natural product was not reported. Our synthetic **1** is levorotatory with $[\alpha]_D^{20} -33^\circ$ (*c* 0.067, MeOH). Since **6** was obtained in 91% ee and **7** in 60% ee, synthetic **1** was estimated to be 98% ee: $(0.955 \times 0.80 - 0.045 \times 0.20) / (0.955 \times 0.80 + 0.045 \times 0.20) = 0.98$.

Figure 1.
The C-nor-D-homosteroids.

Figure 2.
Our synthetic approach to **1**.

Scheme 1.

The synthesis of **6**^a

^aReaction conditions: (a) CDI, NH(OMe)Me•HCl, CH₂Cl₂, 23 °C, 74%. (b) 2 mol % RuCl [(*S,S*)-TsDPEN](*p*-cymene), HCOONa, H₂O, 40 °C, 87%, 91% ee. (c) H₂C=C(Me)MgBr, THF, 0 to 23 °C, 84%. (d) Me₂AlCl, CH₂Cl₂, -78 to -30 °C, 71%. (e) DMAP, ArSO₂Cl, CH₂Cl₂, 0 to 23 °C, 100%; Ar = 2-(MeOOC)Ph. (f) 20 mol % OsO₄, NMO, acetone, H₂O, 23 °C, 89%. (g) LiBr, acetone, 70 °C, 62%. (h) KHMDS, PhNTf₂, THF, -78 °C, 96%.

Scheme 2.

Synthesis of **7** and completion of the synthesis of **1^a**

^aReaction conditions: (a) 15 mol % $\text{Ti}(\text{O}^i\text{Pr})_4$, 18 mol % (-)-diethyl tartrate, $t\text{BuOOH}$, 4Å MS, CH_2Cl_2 , -20°C , 98%, 92% ee. (b) TBSCl, imidazole, DMF, 23°C , 97%. (c) $(\text{Ar}'\text{O})_2\text{AlMe}$, CH_2Cl_2 , -78°C , 79%, 71% ee; $\text{Ar}' = 4\text{-Br-2,6-}t\text{Bu}_2\text{-Ph}$. (d) 10 mol % $\text{Bi}(\text{OTf})_3$, 3-Me-2-TIPSO-furan, CH_2Cl_2 , -40°C , 76% (56% conversion), dr 11:1, 60% ee. (e) H_2 , 10 mol % $[(\text{cod})\text{Ir}(\text{PCy}_3)(\text{Py})]\text{PF}_6$, CH_2Cl_2 , 23°C , 97%, dr 6:1. (f) Dess-Martin periodinane, H_2O , CH_2Cl_2 , 23°C , 98%. (g) NaBH_4 , THF, EtOH, -78°C , 93%. (h) TBSOTf, 2,6-lutidine, CH_2Cl_2 , 40°C , 93%. (i) 1) HOAc, THF, H_2O , 60°C , 76%. (j) Dess-Martin periodinane, H_2O , CH_2Cl_2 , 23°C , 91%. (k) Cl_2 , $\text{P}(\text{OPh})_3$, NEt_3 , CH_2Cl_2 , -78°C , 83%. (l) 45 mol % $\text{Pd}(\text{PPh}_3)_4$, $\text{Me}_3\text{Sn-SnMe}_3$, dioxane, 100°C , 50%. (m) 1 atm CO, $\text{Pd}(\text{PPh}_3)_4$, CuCl, DMSO, 55°C , 62%. (n) $h\nu$ (350 nm), CH_3CN , 23°C . (o) $i\text{Pr}_2\text{NH}$, MeOH, 50°C , 60% for two steps. (p) TFA, CH_2Cl_2 , H_2O , 23°C . (q) NaIO_4 , acetone, pH 7.4 buffer, 23°C . (r) $\text{BF}_3 \cdot \text{OEt}_2$, Et_3SiH , CH_2Cl_2 , 0 to 23°C , 44% for three steps. (s) TBAF, THF, 23°C , 79%.