

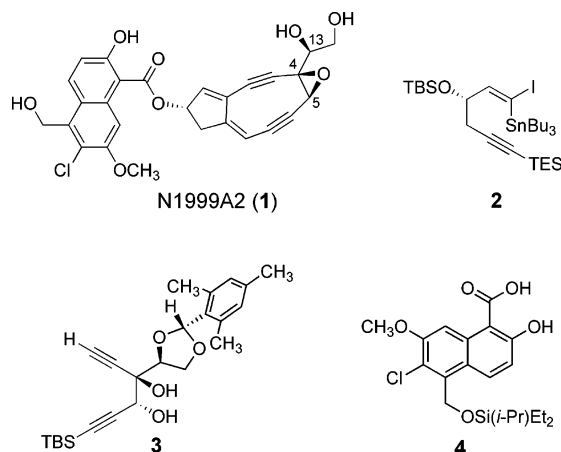
Enantioselective Synthesis of N1999A2

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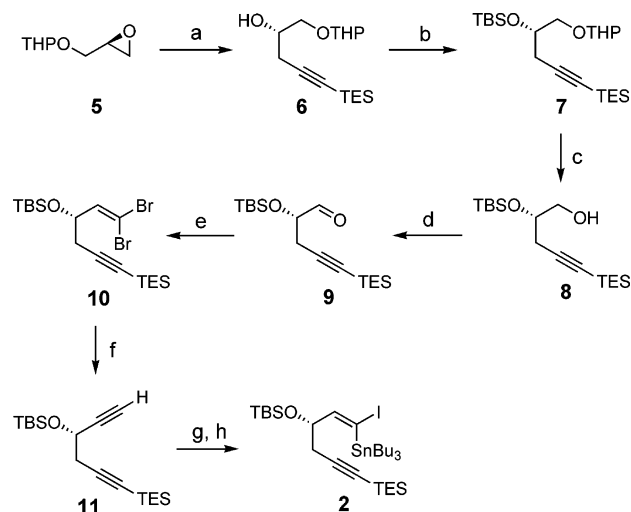
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N1999A2 (**1**) is a nine-membered “enediynes” natural product with extraordinary antiproliferative effects in human cancer cells.¹ Although it is structurally similar to neocarzinostatin chromophore,² N1999A2 (**1**) lacks an aminoglycoside residue and was isolated without an associated carrier protein.^{1a} Also, as Hirama and co-workers established in their prior synthetic studies,^{1b,c} N1999A2 (**1**) is epimeric with the neocarzinostatin chromophore at positions 4, 5, and 13. Here, we describe a second, enantioselective synthetic route to N1999A2 (**1**) that proceeds by the convergent assembly of the (1-iodovinyl) stannane **2**, the 1,5-hexadiyne-3,4-diol derivative **3**, and the substituted naphthoic acid **4**. Each of these starting materials resulted from an empirical selection process, not detailed here,³ that was necessary to achieve the appropriate balance of stability and lability of each protective group. In this way, for example, it was possible to remove three protective groups in one operation in the final step of the synthetic route.



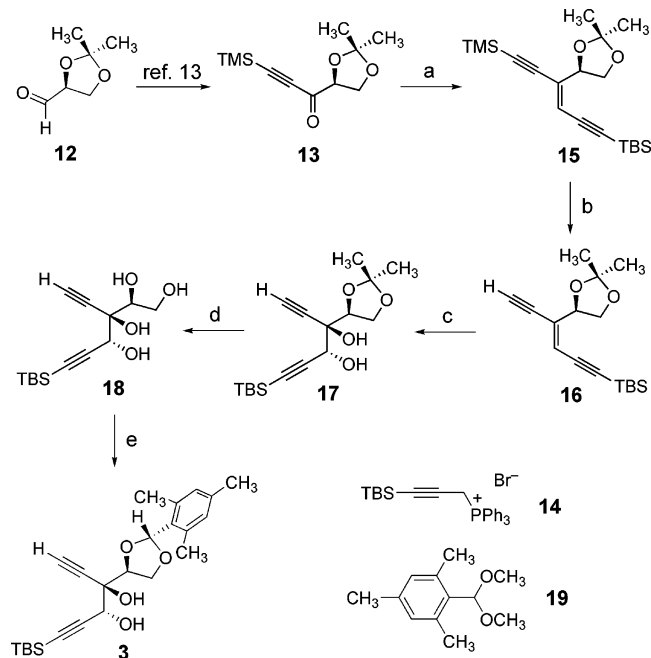
Synthesis of the (1-iodovinyl) stannane **2** began by epoxide opening of the known (*R*)-(+)-glycidol derivative **5** with lithium (triethylsilyl)acetylide to furnish the secondary alcohol **6** (Scheme 1).^{4,5} Silyl ether formation (TBSCl, imidazole, giving **7**)⁶ followed by tetrahydropyranyl protective group removal ((CH₃)₂AlCl, THF, -78→23 °C)⁷ afforded the primary alcohol **8**. Oxidation of **8** under the Parikh–Doering conditions produced the aldehyde **9**.⁸ Treatment of **9** with carbon tetrabromide and triphenylphosphine in dichloromethane then provided the dibromoolefin **10** (95% ee, determined by Mosher ester analysis of a derivative; see Supporting Information),⁹ which was transformed into the terminal alkyne **11** in one operation (NaHMDS, THF, -78 °C, then *n*-BuLi).¹⁰ The latter product (**11**) was stannylated (*n*-BuLi; Bu₃SnCl, THF, -78→23 °C), and the resulting alkynyl tributylstannane was treated with Cp₂Zr(H)Cl (Schwartz's reagent) in THF; iodination of the vinyl zirconium intermediate that was formed then produced the (1-iodovinyl) stannane **2**.¹¹

Synthesis of the 1,5-hexadiyne-3,4-diol derivative **3** began with the known propargylic ketone **13**, available in two steps from (*S*)-

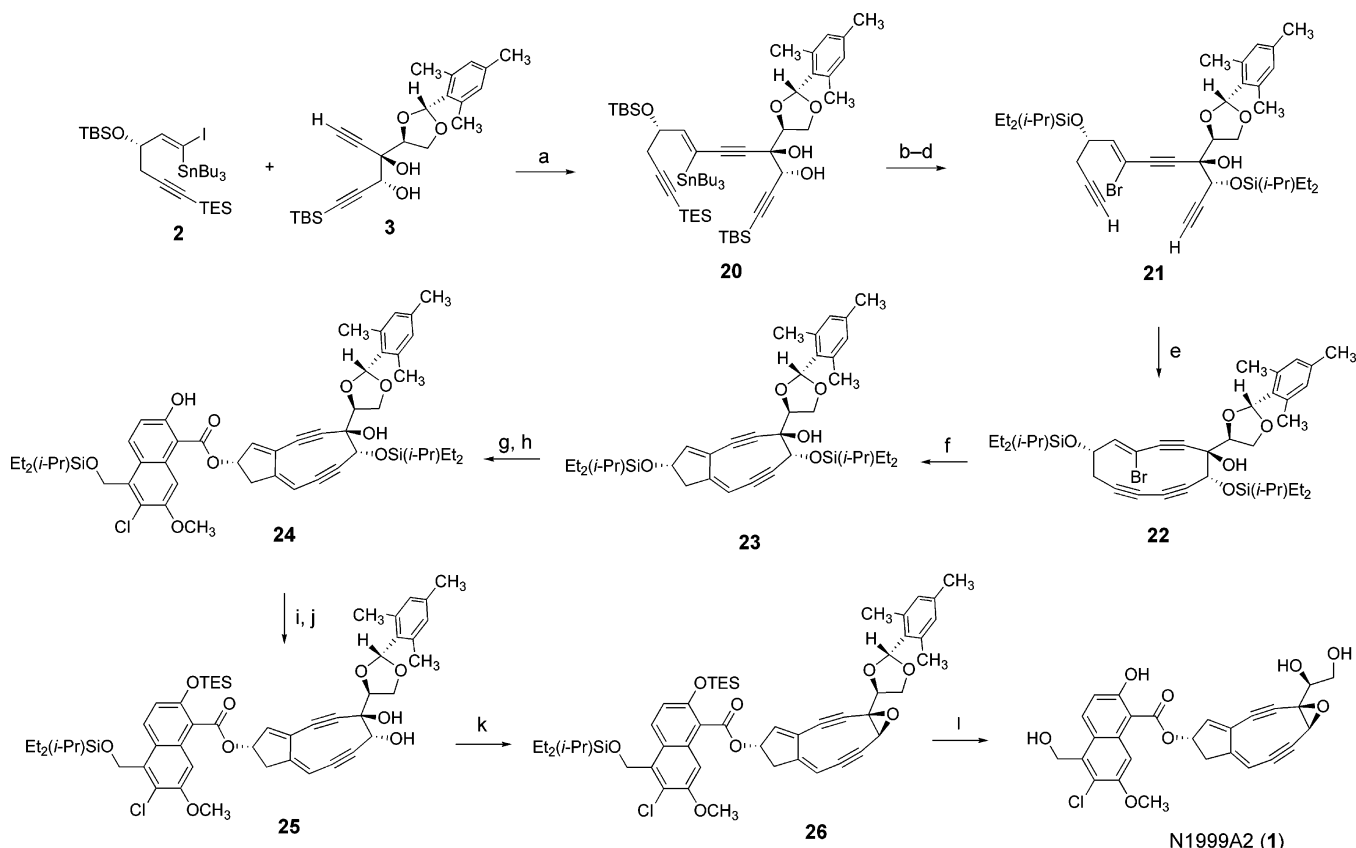
Scheme 1^a

^a Conditions: (a) (Triethylsilyl)acetylene, LHMDs, -78 °C; BF₃·OEt₂, then **5**; (b) TBSCl, imidazole, THF, 23 °C, 95% (two steps); (c) (CH₃)₂AlCl, THF, -78→23 °C, 93%; (d) pyridine sulfur trioxide complex, Et₃N, CH₂Cl₂, DMSO, 0 °C; (e) CBr₄, PPh₃, CH₂Cl₂, 77% (two steps); (f) NaHMDS, THF, -78 °C; *n*-BuLi, 99%; (g) *n*-BuLi, Bu₃SnCl, THF, -78→23 °C; (h) Cp₂Zr(H)Cl, THF, 23 °C; I₂, THF, 69% (two steps).

glyceraldehyde acetonide (**12**) (Scheme 2).^{12,13} In the course of synthetic studies of the neocarzinostatin chromophore, conditions had been developed for an *E*-selective Wittig reaction between **13** and the phosphonium salt **14** (KHMDs, THF, -78 °C, *E*:*Z* = 3:1).¹³ In this work, we report that *Z*-selective coupling of **13** and **14** occurs in the presence of lithium halide additives (5 equiv, deprotonation of **14** with *n*-butyllithium). Lithium iodide was superior to other lithium halide additives (*Z*:*E* ratios: I, 5.54; Br, 3.72; Cl, 2.20; F, 1.98).¹⁴ Wittig reaction of **13** and **14** modified by the inclusion of lithium iodide as an additive typically provided the pure *Z*-olefin **15** in 74% yield after flash-column chromatography. Treatment of the latter product (**15**) with potassium carbonate in methanol at 0 °C selectively removed the trimethylsilyl protective group, providing the enediynes **16**. We conducted a screen of several different osmylation reagents to achieve a diastereoselective dihydroxylation of the enediynes **16** and thus identified as optimal Sharpless' catalytic system involving 1,4-bis(9-*O*-dihydroquinidine)-diphenylpyrimidine ((DHQD)₂PYR) as ligand (20 mol %), K₂OsO₄·2H₂O as osmium source (4 mol %), and potassium ferricyanide as stoichiometric oxidant (5 equiv).^{15,16} The diol diastereomer **17** was produced with >95:5 selectivity; pure **17** was obtained in 53% yield after flash-column chromatography. The acetonide protective group of the dihydroxylation product **17** was cleaved quantitatively (FeCl₃·6H₂O, CH₃CN, 23 °C)¹⁷ to provide the corresponding tetraol (**18**).¹⁸ Selective protection of the latter substance was achieved in the presence of mesitaldehyde dimethyl acetal (**19**)¹⁹ and camphor-sulfonic acid. The diastereomeric mixture of acetals that was formed was separated by flash-column chromatography (3.9:1 dr, 55%

Scheme 2^a

^a Conditions: (a) **14**, *n*-BuLi, LiI, THF, -78°C ; then **13**, 76% (Z-olefin); (b) K_2CO_3 , MeOH, 0°C , 95%; (c) $(\text{DHQD})_2\text{PYR}$ (20 mol %), $\text{K}_2\text{OsO}_4 \cdot 2\text{H}_2\text{O}$ (4 mol %), $\text{K}_3\text{Fe}(\text{CN})_6$ (5.0 equiv), K_2CO_3 (5.0 equiv), $\text{CH}_3\text{SO}_2\text{NH}_2$ (3.0 equiv), *t*-BuOH, H_2O , 0°C , 53%; (d) $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$, CH_3CN , 23°C , 100%; (e) **19**, CSA, THF, 23°C , 3.9:1 dr, 55% (major diastereomer).

Scheme 3^a

^a Conditions: (a) $\text{Pd}(\text{PPh}_3)_4$, CuI, Et_3N , benzene, 23°C , 64%; (b) TBAF, THF, 0°C , 91%; (c) DEIPSCI, imidazole, DMF, 0°C , 89%; (d) NBS, CH_2Cl_2 , 23°C , 89%; (e) $\text{Cu}(\text{OAc})_2$, THF–pyridine (1:1), 60°C , 75%; (f) LHMDS, THF–toluene (1:1), -78°C ; *t*-BuLi (4.0 equiv), then HOAc, 30–40%; (g) $\text{Et}_3\text{N} \cdot 3\text{HF}$, CH_3CN , -25°C ; (h) **4**, DCC, THF, 0°C , 44% (two steps); (i) TBAF (6.0 equiv), *o*-nitrophenol (6.0 equiv), THF, 0°C ; (j) TESCl, Et_3N , CH_2Cl_2 , -78°C ; (k) TsCl, DABCO, CH_2Cl_2 , 23°C , 33% (three steps); (l) TFA, THF, H_2O , 0°C , 5 h, 76%.

isolated yield of the major diastereomer **3**, stereochemistry determined by nOe analysis). Only the major diastereomer (**3**) was used in the subsequent coupling reaction.

The (1-iodovinyl) stannane **2** and the 1,5-hexadiyne-3,4-diol **3** were coupled in the presence of a palladium catalyst, forming the enetriyne diol **20** (Scheme 3). One *O*-silyl and two *C*-silyl protective groups of the coupling product **20** were cleaved in the presence of tetrabutylammonium fluoride in THF. Two new *O*-diethylisopropylsilyl protective groups were then introduced selectively (the tertiary alcohol remained free); bromodestannylation with *N*-bromosuccinimide then afforded the substrate for intramolecular oxidative acetylene coupling (**21**). Attempts to cyclize **21** under modified Eglinton conditions ($\text{Cu}(\text{OAc})_2$, CuI, pyridine, THF), proven to be optimal in a closely analogous system, met with varied success.^{20,21} Further experiments revealed that by simply omitting CuI from the cyclization reaction the cyclic bromoenetriyne **22** could be produced in high yield and, importantly, in a reproducible manner. Solutions of this and all subsequent intermediates could not be concentrated without extensive decomposition. As a general practice, these unstable intermediates were purified by flash-column chromatography, fractions containing pure intermediates were pooled, and the combined fractions were diluted with a deuterated solvent (C_6D_6 or CD_3CN). The resulting solution was then partially concentrated to allow for ^1H NMR analysis; yields were determined using an internal standard (see Supporting Information). In the key step of the sequence, a solution of the substrate **22** in THF–toluene (1:1, stirred with 4 Å molecular sieves, 23°C , 15 min) was treated at -78°C with a solution of lithium hexamethyldisilazide (LHMDS) in THF (1.0 M, 1.1 equiv) to deprotonate the tertiary

hydroxyl group. Subsequent addition of a solution of *tert*-butyllithium in pentane (1.7 M, 4.0 equiv) at -78°C , followed immediately (<3 s) by quenching with a solution of acetic acid (30 equiv) in THF, afforded the transannular cyclization product **23** in 30–40% yield.²¹ Typically, the latter reaction was performed on scales of 20–25 mg; larger-scale reactions were less efficient. Selective removal of the allylic diethylisopropylsilyl ether group within the transannular product **23** was achieved by treatment of **23** with an excess of triethylamine trihydrofluoride in acetonitrile at -25°C . The desilylated product was then coupled with the naphthoic acid **4**²² in the presence of *N,N'*-dicyclohexylcarbodiimide (DCC) in THF, providing the ester **24** (44%, two steps). Addition of tetrabutylammonium fluoride (6.0 equiv) to a solution of the ester **24** and the buffering agent *o*-nitrophenol (6.0 equiv) in THF at 0°C cleanly removed the propargylic silyl ether within **24**; subsequent protection of the phenolic hydroxyl group as the corresponding triethylsilyl ether (TESCl, Et_3N , CH_2Cl_2 , -78°C) then afforded the highly unstable diol **25**. The *trans*-diol function within the latter product was transformed to the corresponding epoxide by tosylation under basic conditions (TsCl , DABCO, CH_2Cl_2 , 23°C), providing N1999A2 in fully protected form (**26**), a more stable intermediate relative to others (**22** and beyond) in the sequence. During the latter transformation (**25**→**26**) the tosylate intermediate was observable by TLC analysis, but it did not accumulate, being transformed to **26** as it was formed. Global deprotection of the epoxide **26** was accomplished with trifluoroacetic acid in a mixture of THF and water at 0°C .^{1c} In this reaction, both silyl groups were cleaved within 2 h, and the mesitylene protective group was removed within 5–8 h. To isolate synthetic N1999A2, particular workup conditions were necessary. For example, addition of saturated sodium bicarbonate aqueous solution led to extensive decomposition of the product. By using pH 7 aqueous phosphate buffer as a quenching solution, decomposition was minimized. The product was extracted into ether, and the ethereal solution was dried (Na_2SO_4) and then concentrated to a volume of 4–8 mL. Since N1999A2 is an appreciably polar compound, a rather polar eluent (5% CD_3CN –ether) was necessary for its purification by flash-column chromatography. Although some decomposition did occur during chromatography, the product obtained was estimated to be $>90\%$ pure by ^1H NMR analysis. A much simpler and more convenient purification procedure was found serendipitously. When pentane (5 mL) was added to a solution of synthetic N1999A2 (~ 5 mg) in ether (8 mL), a light-yellow solid precipitated from the solution; solvents were removed, and the resulting solid was triturated with a 5:1 mixture of ether and pentane. N1999A2 had not previously been reported to be a solid. ^1H NMR and UV spectra of synthetic (solid) N1999A2 were identical to published spectra.^{1a,c} Thus, for the second time, the stereostructure of N1999A2 proposed by Hiram and co-workers has been confirmed by an unambiguous synthetic route.^{1b,c} A high-resolution “exact” mass spectrum of synthetic N1999A2 was also obtained using a Micromass LCT/TOF spectrometer equipped with an electrospray ionization source (calcd for $\text{C}_{27}\text{H}_{25}\text{ClNO}_8$ $[\text{M}+\text{NH}_4]^+$ 526.1269, found 526.1286). Synthetic N1999A2 was surprisingly stable. In the solid state, N1999A2 could be left in the air for 10 min at ambient temperature without evident decomposition by subsequent ^1H NMR analysis, and solutions of N1999A2 in ether or DMSO have been stored at -25°C for 2 months without detectable decomposition by HPLC analysis.

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Supporting Information Available: Detailed experimental procedures and tabulated spectroscopic data (^1H and ^{13}C NMR, FT-IR, and HRMS) for all new compounds. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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