N1999A2 (1) is a nine-membered “enediyne” natural product with extraordinary antiproliferative effects in human cancer cells. Although it is structurally similar to neocarzinostatin chromophore, N1999A2 (1) lacks an aminoglycoside residue and was isolated without an associated carrier protein. Also, as Hirama and co-workers established in their prior synthetic studies, 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, 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.

**Scheme 1**

**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). Silyl ether formation (TBSCl, imidazole, giving (triethylsilyl)acetylide to furnish the secondary alcohol

![Diagram](Image 71x271 to 277x437)

The latter product (11) was stannylated (Bu3SnLi; Bu3SnCl, THF, −78 to 23 °C) to provide the corresponding tetraol 12. Each of these starting materials resulted from an empirical selection process, not detailed here, 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.

![Diagram](Image 78x107 to 107x230)

glyceraldehyde acetonide (12) (Scheme 2). 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 (KHMS, THF, −78 °C; E:Z = 3:1). 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). 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 enediyne 16. We conducted a screen of several different osmylation reagents to achieve a diastereoselective dihydroxylation of the enediyne 16 and thus identified as optimal Sharpless’ catalytic system involving 1,4-bis(9-O-dihydroquinidine)-diphenylpyrrole ((DHQD)2PYR) as ligand (20 mol %), K2OsO4·2H2O as osmium source (4 mol %), and potassium ferricyanide as stoichiometric oxidant (5 equiv). The diol diastereomer 17 was produced with >95:5 selectivity; pure 17 was obtained in 53% yield after flash-column chromatography. The acetone protective group of the dihydroxylation product 17 was cleaved quantitatively (FeCl3·6H2O, CH3CN, 23 °C) to provide the corresponding tetracol (18). Selective protection of the latter substance was achieved in the presence of mesitaldehyde dimethyl acetal (19) and camphorsulfonic acid. The diastereomeric mixture of acetics that was formed was separated by flash-column chromatography (3.9:1 dr, 55%...
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); bromodesstannylation with N-bromosuccinimide then afforded the substrate for intramolecular oxidative acetylene coupling (21). Attempts to cyclize 21 under modified Eglinton conditions (Cu(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 (C₆D₆ or CD₃CN). 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

Scheme 2

Scheme 3

a Conditions: (a) 14, n-BuLi, Lii, THF, −78 °C; then 13, 76% (Z-olefin); (b) K₂CO₃, MeOH, 0 °C, 95%; (c) (DHQD)PYR (20 mol %), K₂O₂C₂H₅O (4 mol %), K₂Fe(CN)₆ (5.0 equiv), K₂CO₃ (5.0 equiv), CH₃SO₂NH₂ (3.0 equiv), r-BuOH, H₂O, 0 °C, 53%; (d) FeCl₃, 6H₂O, CH₃CN, 23 °C, 100%; (e) 19, CSA, THF, 23 °C, 3.9:1 dr, 55% (major diastereomer).

a Conditions: (a) Pd(PPh₃)₄, CuI, Et₃N, benzene, 23 °C, 64%; (b) TBAF, THF, 0 °C, 91%; (c) DEIPSCl, imidazole, DMF, 0 °C, 89%; (d) NBS, CH₃Cl, 23 °C, 89%; (e) Cu(OAc)₂, THF—pyridine (1:1), 60 °C, 75%; (f) LHMDS, THF—toluene (1:1), −78 °C; r-BuLi (4.0 equiv), then H₂O, 30–40%; (g) Et₃N, 3HF, CH₂Cl₂, −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, Et₃N, CH₂Cl₂, −78 °C; (k) TsCl, DABCO, CH₂Cl₂, 23 °C, 33% (three steps); (l) TFA, THF, H₂O, 0 °C, 5 h, 76%.
hydroxyl group. Subsequent addition of a solution of tert-butyl-lithium 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.\textsuperscript{21} Typically, the latter reaction was performed on scales of 20-25 mg; larger-scale reactions were less efficient. Selective removal of the allylic diethyisopropylsilyl 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\textsuperscript{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 triethylosilyl ether (TESCl, Et3N, CH2Cl2, -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, CH2Cl2, 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.\textsuperscript{14} 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 (Na2SO4) and then concentrated to a small ethereal solution was obtained. Subsequently, the mixture was treated with tetrafluoroboric acid (TFA, THF, H2O, 0 °C),\textsuperscript{41} followed by stirring at 23 °C for 2 hours. The resulting solution was filtered, and the filtrate was concentrated to a solid residue. The residue was triturated with a 5:1 mixture of ether and pentane. N1999A2 had not previously been reported to be a solid.\textsuperscript{41} \textsuperscript{14} With the use of alkynyl trifluoroborates in epoxide-opening reactions, see: Parikh, J. R.; Doering, W. v. E. J. Am. Chem. Soc. 1967, 89, 5505.

Supporting Information Available: Detailed experimental procedures and tabulated spectroscopic data (\textsuperscript{1}H and \textsuperscript{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.

Acknowledgment. Generous financial support from the National Institutes of Health and Pfizer, Inc. is gratefully acknowledged.

References


(12) (S)-Glyceraldehyde acetate (12) was synthesized in one step from (R)-(-)-2,2-dimethyl-1,3-dioxolane-4-methanol (Alfa Aesar) by the method of Janssens et al. See: Janssens, M.; Vanbrakel, I.; Svensson, S. C. T.; Samuelson, B. C. B. Synthesis 1993, 129.


(14) In the absence of any lithium halide additive, Wittig coupling of 13 and 14 using n-butyl lithium as base afforded a 1.5:1 mixture of E- and Z- e-olefins, respectively. The undesired E-isomer could be transformed into a 55:45 mixture of E- and Z-isomers, respectively, by UV irradiation. See Supporting Information for details.


(16) Brummond and co-workers have reported that the (DHQD)PFYR ligand was optimal for the enantioselective dihydroxylation of an achiral enyne substrate. See: Brummond, K. M.; Lu, J.; Petersen, J. J. Am. Chem. Soc. 2000, 122, 4915.

(17) In this application, acetonitrile was found to be a more effective solvent than dichloromethane. This solvent was used by Sen et al. See: Sen, S. E.; Roach, S. L.; Bogg, J. K.; Ewing, G. H.; Magrath, J. J. Org. Chem. 1997, 62, 6684.

(18) The acetonide protective group proved to be too robust to remove without decomposition in later-stage synthetic intermediates and was therefore modified. Model studies established that the mesitylene protective group was an ideal replacement, being readily removed under acidic conditions (TFA, THF, H2O, 0 °C).\textsuperscript{41}

(19) Mesityldehyde dimethyl acetal (19) was synthesized in one step from mesitaldehyde as follows: camphorsulfonic acid (10 mg) was added to a solution of mesitaldehyde (3.00 g, 20.2 mmol, 1 equiv) and trimethyl orthoformate (3.22 g, 30.4 mmol, 1.5 equiv) in methanol (30 mL) at 23 °C. After stirring at 23 °C for 12 h, the reaction mixture was partitioned between ether (30 mL) and saturated aqueous sodium bicarbonate solution (30 mL). The aqueous layer was separated and further extracted with ether (30 mL). The organic extracts were combined, and the combined solution was dried over anhydrous sodium sulfate. The dried solution was filtered, and the filtrate was concentrated to provide mesitaldehyde dimethyl acetal (19) as a colorless oil (3.90 g, 99%).


(22) The diethylisopropylsilyl-protected naphthoic acid 4 was synthesized in two steps from the corresponding triisopropylsilyl-protected naphthoic acid; see Supporting Information. For synthesis of the triisopropylsilyl-protected naphthoic acid, see: Ji, N.; Rosen, B. M.; Myers, A. G. Org. Lett. 2004, 6, 4551.

JA0662467