

Total Synthesis of FR901464, an Antitumor Agent that Regulates the Transcription of Oncogenes and Tumor Suppressor Genes

Brian J. Albert, Ananthapadmanabhan Sivaramakrishnan, Tadaatsu Naka, and Kazunori Koide*

Department of Chemistry, University of Pittsburgh, 219 Parkman Avenue, Pittsburgh, Pennsylvania 15260

Received December 2, 2005; E-mail: koide@pitt.edu

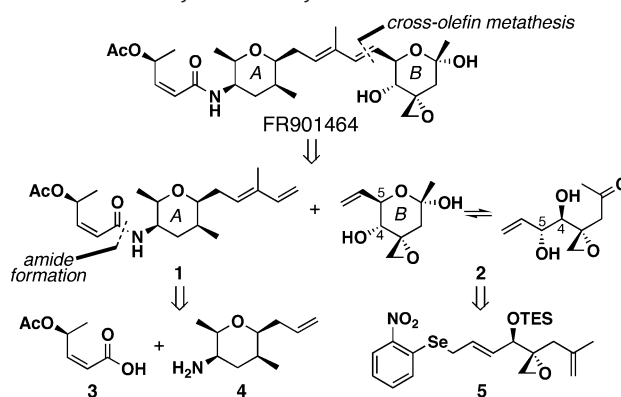
In search for anticancer natural products with new modes of action, the Fujisawa group isolated FR901464 (Scheme 1) from the culture broth of a bacterium of *Pseudomonas* sp. No.2663 as a novel transcriptional activator.¹ This natural product lowers the mRNA levels of *p53*, *p21*, *c-myc*, and *E2F-1* in MCF-7 cells at 20 nM^{1b} and induces apparent apoptosis in MCF-7 cells with the impressive LC₅₀ of 0.5 nM. It also exhibits an antitumor activity in a mouse model at remarkably low concentrations (0.056–0.18 mg/kg).^{1b} This unprecedented pharmacological profile of FR901464 has drawn considerable interest² and prompted us to further investigate the biology of FR901464.

Despite the two previous syntheses of FR901464,³ a more concise synthetic approach was highly desirable to take full advantage of such biological activities. Scheme 1 illustrates our retrosynthetic analysis of FR901464, in which the priority was to accomplish a coupling between A- and B-ring fragments with complete functionality for ultimate convergency. While several intramolecular diene-ene olefin metathesis reactions have been reported,⁴ the corresponding intermolecular version was unprecedented in natural product synthesis at the outset of this research.⁵ Nonetheless, we reasoned that the ruthenium–alkylidene complex with **2** would be more reactive than that of **1** (if the terminal olefin reacts), the trisubstituted and electron-deficient olefin would not react with the ruthenium catalyst due to steric and electronic reasons, and thermodynamics would favor FR901464 over the homodimer of **2** under reversible conditions. Further retrosynthetic analysis of the A-ring fragment **1** and B-ring fragment **2** revealed acid **3**, amine **4**, and selenide **5**.

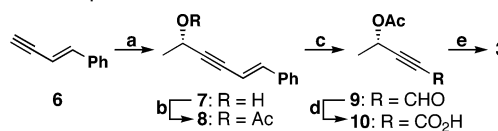
With this strategy in mind, carboxylic acid **3** was prepared as shown in Scheme 2. We chose to use the styrene unit as a masked aldehyde because the styryl group significantly suppressed the volatility of otherwise low molecular weight intermediates. Known enyne **6** was prepared from cinnamaldehyde according to the literature (TMSCHN₂, LDA, 84%).⁶ The next step employed a Carreira asymmetric alkynylation between **6** and acetaldehyde to generate alcohol **7**.⁷ This alcohol was then converted to acetate **8**, and subsequent ozonolysis afforded aldehyde **9**. Further oxidation of this aldehyde gave **10**, which was then partially hydrogenated with Lindlar's catalyst to afford **3**.

Scheme 3 outlines the preparation of **1**. The L-threonine derivative **11**, prepared in one step (2-methoxypropene, CSA; quant.) from commercially available *N*-Boc-L-threonine methyl ester, was transformed to **12** using a one-pot procedure (DIBAL-H; Ph₃P=CH₂).⁸ Removal of the oxazolidine ring of **12** using CSA in MeOH generated alcohol **13**, and subsequent O-methallylation afforded diene **14**. The ring-closing metathesis of **14** was quantitative using 1 mol % of Grubbs' 2nd generation catalyst⁹ to provide **15**. To prepare lactone **16**, we found that allylic oxidation of **15** with PDC was most regioselective and efficient. Subsequent stereoselective hydrogenation of **16** gave desired lactone **17** and its C12-epimer in a 10:1 ratio. The allylation of **17** gave hemiketal

Scheme 1. Retrosynthetic Analysis of FR901464



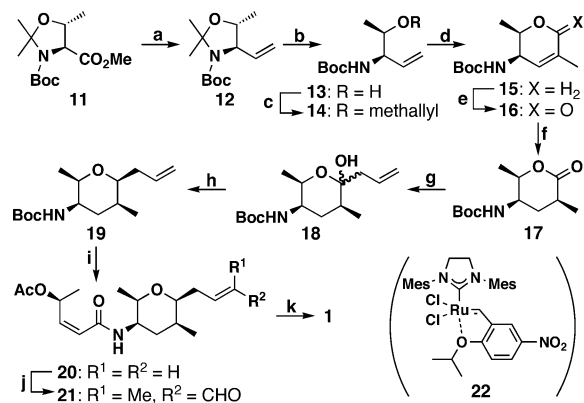
Scheme 2. Preparation of **3**^a



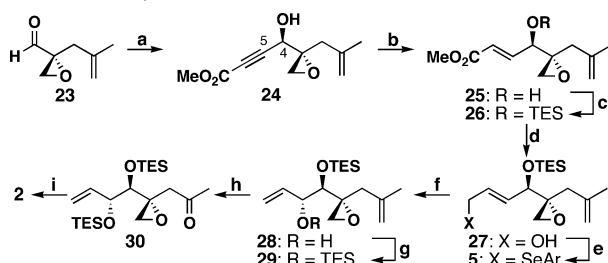
^a Conditions: (a) CH₃CHO (2.3 equiv), Zn(OTf)₂ (1.0 equiv), Et₃N (1.0 equiv), (–)-*N*-methylephedrine (1.0 equiv), toluene, 23 °C, 41% (72% ee); (b) Ac₂O (5.0 equiv), pyridine, 23 °C, quant.; (c) O₃, CH₂Cl₂, –78 °C; Me₂S (10 equiv), –78→23 °C, 89%; (d) NaClO₂ (3.0 equiv), NaH₂PO₄ (3.0 equiv), 2-methyl-2-butene (15 equiv), H₂O/BuOH (1:1), 23 °C; (e) H₂ (1 atm), Lindlar's catalyst (1 mol %), quinoline (10 mol %), EtOH, 23 °C, 75% (2 steps).

18, which is in equilibrium with an aminal. Due to the presence of two anomers for both **18** and the aminal, we were not able to determine the relative ratio among these four compounds. In the next step, this mixture was subjected to reduction conditions (BF₃·OEt₂, Et₃SiH, CF₃CH₂OH), providing the desired compound **19** along with a pyrrolidine derivative (see Supporting Information). Subsequent coupling of **3** and **19** (via amine **4**) gave amide **20**. Methacrolein and **20** were then subjected to the cross-olefin metathesis conditions using 5 mol % of catalyst **22**¹⁰ to form the desired aldehyde **21**, which was then converted to diene **1** upon addition of Ph₃P=CH₂.

B-ring fragment **2** was prepared according to Scheme 4. Through the three-step sequence that we previously reported, aldehyde **23** was prepared from methallyl bromide and propargyl alcohol.¹¹ The subsequent C4–C5 bond formation was most stereoselective and efficient using the Zr/Ag-promoted alkynylation method developed in our laboratory to afford **24** and C4-epimer in a ratio of 6:1 in favor of **24**.¹² While the partial hydrogenation of **24** or its TES ether failed, the Red-Al reduction protocol from our laboratory successfully afforded allylic alcohol **25**.¹³ This alcohol was protected as the TES ether **26**, which was then reduced by DIBAL-H to furnish the primary alcohol **27**. Transformation of the hydroxy group of **27** to the *o*-nitrophenylselenide gave **5**. Despite the lack of closely related Mislow–Evans-type [2,3]-sigmatropic rearrangements of

Scheme 3. Preparation of **1**^a

^a Conditions: (a) DIBAL-H (2.0 equiv), CH₂Cl₂, -78 °C; Ph₃PCH₃Br (2.1 equiv), 'BuOK (2.0 equiv), THF, -78→48 °C, 77%; (b) CSA (10 mol %), MeOH, 23 °C, 95%; (c) methallyl bromide (4.0 equiv), Ag₂O (1.5 equiv), DMF, 23 °C, 86%; (d) Grubbs' 2nd cat. (1 mol %), PhH, reflux, quant.; (e) PDC (6.0 equiv), (ClCH₂)₂, reflux, 72%; (f) H₂ (1 atm), PtO₂ (1 mol %), EtOH, 23 °C, quant.; (g) allyl-MgBr (2.0 equiv), THF, -78 °C, 96%; (h) Et₃SiH (10 equiv), BF₃·OEt₂ (4.0 equiv), CF₃CH₂OH (8.0 equiv), -78 °C, 38%; (i) TFA/CH₂Cl₂ (1:9), 23 °C, 3 (1.2 equiv), HATU (1.2 equiv), 'Pr₂NEt (4.0 equiv), 23 °C, 86%; (j) **22** (5 mol %), methacrolein (20 equiv), CH₂Cl₂, 23 °C, 57% (67% based on recovered **20**); (k) Ph₃PCH₃Br (1.4 equiv), 'BuOK (1.2 equiv), THF, 0 °C, 86%.

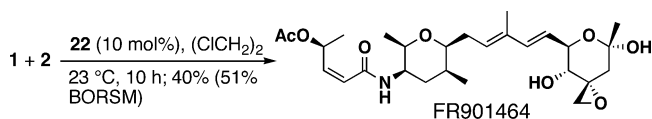
Scheme 4. Preparation of **2**^a

^a Conditions: (a) Ag-C≡C-CO₂Me (1.7 equiv), Cp₂ZrCl₂ (1.3 equiv), AgOTf (0.2 equiv), CH₂Cl₂, 23 °C, 84%; (b) Red-Al (2.0 equiv), -72 °C, 81%; (c) TESCl (1.4 equiv), imidazole (1.5 equiv), THF, 0 °C, quant.; (d) DIBAL-H (3.0 equiv), THF, -78 °C, 95%; (e) *o*-O₂N-PhSeCN (1.2 equiv), 'Bu₃P (1.4 equiv), THF, 0 °C, quant.; (f) H₂O₂ (30% v/v in H₂O, excess), DMAP (5.0 equiv), THF, -44→23 °C, 96%; (g) TESCl (1.4 equiv), imidazole (1.6 equiv), THF, 0 °C, 95%; (h) OsO₄ (1 mol %), NMO (0.96 equiv), THF/H₂O (10:1), 0→23 °C; Pb(OAc)₄ (1.2 equiv), PhH, 0→23 °C, 71% (86% based on recovered **29**); (i) AcOH/THF/H₂O (3:3:1), 0→23 °C, 91%.

chiral *E*-allylselenides, we proceeded to treat substrate **5** with H₂O₂ and DMAP, which promoted a rearrangement via the putative selenoxide to provide the desired allylic alcohol **28** and its diastereomer with a pleasantly surprising diastereomeric ratio of 7.5:1.¹⁴ Alcohol **28** was protected as the TES ether **29**, which dramatically improved the regioselectivity of the oxidative cleavage sequence (OsO₄-NMO; Pb(OAc)₄), giving ketone **30**. Finally, both TES groups were hydrolyzed under carefully optimized conditions to form the fully functionalized B-ring fragment **2**.

The stage was set to test the cross diene-ene metathesis between **1** and **2** (Scheme 5). Gratifyingly, despite the absence of protecting groups, the coupling of these two fragments in the presence of catalyst **22** furnished FR901464 in 40% yield after subjecting the unreacted **1** and **2** to the same conditions without a detectable *cis* isomer. The decomposition of FR901464 during column chromatog-

Scheme 5. Final Stage



raphy^{3a} partly accounts for the loss of the material. Only 5% of homodimers of **2** were detected, and diene **1** did not form its homodimer under the reaction conditions. The fragile nature of **2** (thermal decomposition at ≥47 °C) precluded more forcing reaction conditions.

In summary, we completed the total synthesis of FR901464 in the 13 longest linear steps with 31 total steps, which features Zr/Ag-promoted alkynylation using electron-deficient methyl propiolate, mild Red-Al reduction, stereoselective [2,3]-sigmatropic rearrangement via a selenoxide, and diene-ene cross olefin metathesis without protecting groups. Biological studies of FR901464 and its analogs are underway in our laboratory.

Acknowledgment. We wish to dedicate this paper to the 60th birthday of Professor K. C. Nicolaou. This work was supported by the University of Pittsburgh, the American Chemical Society (PRF No. 38542-G1), The American Cancer Society George Heckman Institutional Research Grant, and The Competitive Medical Research Fund. B.J.A. is thankful for a Graduate Excellence Fellowship.

Supporting Information Available: Experimental procedures and spectroscopic data for all the new compounds and FR901464. This material is available free of charge via the Internet at <http://pubs.acs.org>.

References

- (1) (a) Nakajima, H.; Sato, B.; Fujita, T.; Takase, S.; Terano, H.; Okuhara, M. *J. Antibiot.* **1996**, *49*, 1196–1203. (b) Nakajima, H.; Hori, Y.; Terano, H.; Okuhara, M.; Manda, T.; Matsumoto, S.; Shimomura, K. *J. Antibiot.* **1996**, *49*, 1204–1211. (c) Nakajima, H.; Takase, S.; Terano, H.; Tanaka, H. *J. Antibiot.* **1997**, *50*, 96–99.
- (2) A few examples: (a) Imamura, Y.; Ohtsu, Y.; Tanaka, H.; Hatakeyama, M.; Manabe, T.; Kawaguchi, H.; Handa, H.; Takahashi, T. *Heterocycles* **2004**, *64*, 51–56. (b) Tashiro, E.; Imoto, M.; Yoshida, M.; Kitahara, T.; Watanabe, H.; Nakajima, H. *Jpn. Kokai Tokkyo Koho JP 2005323514*, Nov. 24, 2005.
- (3) (a) Thompson, C. F.; Jamison, T. F.; Jacobsen, E. N. *J. Am. Chem. Soc.* **2001**, *123*, 9974–9983; 40 total, 20 longest linear steps. (b) Horigome, M.; Motoyoshi, H.; Watanabe, H.; Kitahara, T. *Tetrahedron Lett.* **2001**, *42*, 8207–8210; 42 total, 24 longest linear steps.
- (4) For examples, see: (a) Yang, Z. Q.; Geng, X. D.; Solit, D.; Pratilas, C. A.; Rosen, N.; Danishefsky, S. J. *J. Am. Chem. Soc.* **2004**, *126*, 7881–7889. (b) Snider, B. B.; Hawryluk, N. A. *Org. Lett.* **2001**, *3*, 569–572. (c) Clive, D. L. J.; Liu, D. Z. *Tetrahedron Lett.* **2005**, *46*, 5305–5307.
- (5) Presentation of preliminary results: (a) Albert, B. J.; Sivaramakrishnan, A.; Koide, K. 228th ACS National Meeting, Philadelphia, PA, Aug 22–26; American Chemical Society: Washington, DC, 2004. First appearance in the literature: (b) Crimmins, M. T.; Christie, H. S.; Chaudary, K.; Long, A. *J. Am. Chem. Soc.* **2005**, *127*, 13810–13812.
- (6) Miwa, K.; Aoyama, T.; Shioiri, T. *Synlett* **1994**, 107–108.
- (7) (a) Frantz, D. E.; Fassler, R.; Carreira, E. M. *J. Am. Chem. Soc.* **2000**, *122*, 1806–1807. (b) The catalytic version was unsuccessful. (c) Alcohol **7** was recrystallized to 95% ee and used in subsequent steps.
- (8) Wei, Z.-Y.; Knaus, E. E. *Synthesis* **1994**, 1463–1466.
- (9) Scholl, M.; Ding, S.; Lee, C. W.; Grubbs, R. H. *Org. Lett.* **1999**, *1*, 953–956.
- (10) Grela, K.; Harutyunyan, S.; Michrowska, A. *Angew. Chem., Int. Ed.* **2002**, *41*, 4038–4040.
- (11) Albert, B. J.; Koide, K. *Org. Lett.* **2004**, *6*, 3655–3658.
- (12) Shahi, S. P.; Koide, K. *Angew. Chem., Int. Ed.* **2004**, *43*, 2525–2527.
- (13) Meta, C. T.; Koide, K. *Org. Lett.* **2004**, *6*, 1785–1787.
- (14) (a) Sharpless, K. B.; Lauer, R. F. *J. Am. Chem. Soc.* **1972**, *94*, 7154–7155. (b) Reich, H. J. *J. Org. Chem.* **1975**, *40*, 2570–2572.

JA058216U