

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

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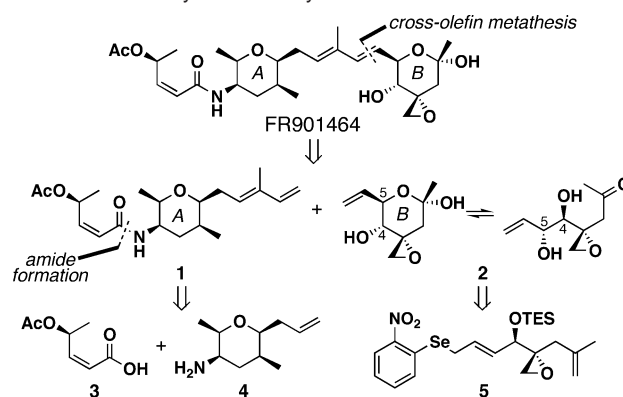
In search for anticancer natural products with new modes of action, the Fujisawa group isolated FR901464 (Scheme 1) from the culture broth of a bacterium of *Pseudomonas* sp. No.2663 as a novel transcriptional activator.¹ This natural product lowers the mRNA levels of *p53*, *p21*, *c-myc*, and *E2F-1* 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**.

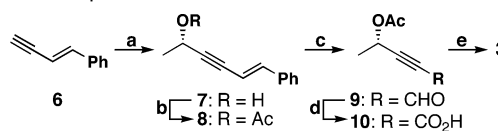
With this strategy in mind, carboxylic acid **3** was prepared as shown in Scheme 2. We chose to use the styrene unit as a masked aldehyde because the styryl group significantly suppressed the volatility of otherwise low molecular weight intermediates. Known enyne **6** was prepared from cinnamaldehyde 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



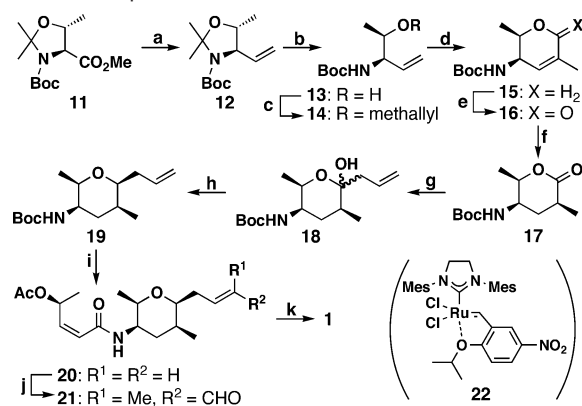
Scheme 2. Preparation of **3**^a



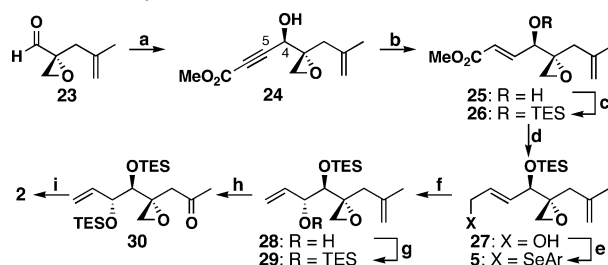
^a Conditions: (a) CH₃CHO (2.3 equiv), Zn(OTf)₂ (1.0 equiv), Et₃N (1.0 equiv), (–)-*N*-methylphenylhydrazine (1.0 equiv), toluene, 23 °C, 41% (72% ee); (b) Ac₂O (5.0 equiv), pyridine, 23 °C, quant.; (c) O₃, CH₂Cl₂, –78 °C; Me₂S (10 equiv), –78→23 °C, 89%; (d) NaClO₂ (3.0 equiv), NaH₂PO₄ (3.0 equiv), 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 aminor. Due to the presence of two anomers for both **18** and the aminor, 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%.

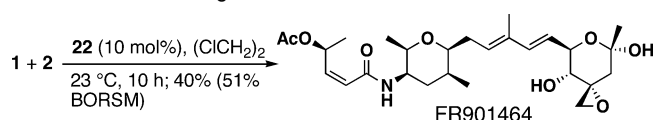
Scheme 4. Preparation of 2^a

^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.

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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>.

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