

Synthesis and Stereochemical Assignment of FR252921, a Promising Immunosuppressant**

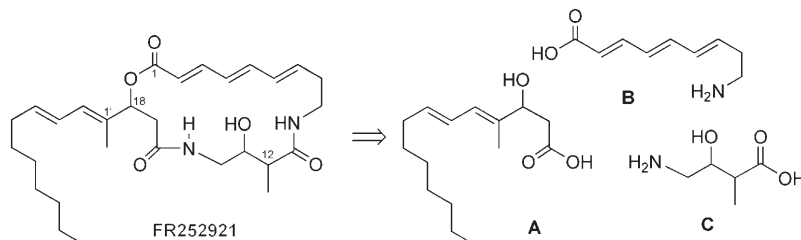
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Dedicated to Professor Koichi Narasaka

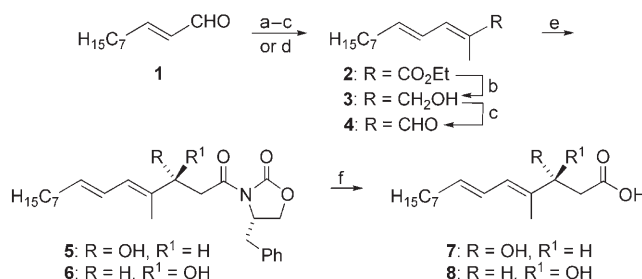
FR252921, an unusual 19-membered lactone–dilactam distinguished by a lipophilic C₁₂ appendage, was isolated from the culture broth of *Pseudomonas fluorescens* by Fujine et al., who published its structure without characterization of its three stereogenic centers.^[1] Interest in FR252921 from this^[2] and other^[3,4] laboratories stems in large part from its unique pharmacological profile. In contrast with other immunosuppressants, FR252921 potentially inhibits both lipopolysaccharide-(LPS)-stimulated and anti-CD3 mAb-stimulated splenocyte proliferation in vitro without blocking T-cell activation.^[5] It also decreases transcription regulated by activating protein 1 (AP-1) despite a null effect on the transcription factors NF-AT and NF-κB. Notably, FR252921 synergizes strongly the effects of FK506 in vitro and in vivo.^[6] To expedite current studies into the site of action of FR252921 and conclude the complete assignment of its structure, we describe herein the asymmetric total synthesis of FR252921 and three diastereomers by a versatile, convergent strategy.

Retrosynthetic analysis dissected FR252921 into segments A–C (Scheme 1), which were chosen to provide convenient access to the minimum stereocenter^[7] needed to establish the relative and absolute configuration of the natural product as well as to support future structure–activity–relationship studies.^[8,9] The segments would be assembled by following a sequence C→BC→ABC, which postponed the macrolactonization of the labile C18–OH group to the final stages of the synthesis to minimize opportunities for β elimination.

The route to both enantiomers of segment A (Scheme 2) commenced with (*E*)-2-decenal (**1**), which was homologated with commercial ethyl 2-(diethylphosphono)propionate to give a chromatographically separable 12:1 mixture of the



Scheme 1. Retrosynthetic analysis of FR252921.



Scheme 2. Synthesis of carboxylic acids **7** and **8**: a) (EtO)₂P(O)CH(CH₃)CO₂Et, NaH, THF, 0 °C, 2 h, 92% (*Z,E/E,E* 1:12); b) DIBAL-H, CH₂Cl₂, 0 °C, 20 min; c) MnO₂/MgSO₄, CH₂Cl₂, 23 °C, 2 days, 93% for two steps; d) H₃CCl₂CHO, CrCl₂, BF₃·Et₂O, THF/DMF (2:1), 23 °C, 14 h, 74% (*Z,E/E,E* 1:10); e) **9**, SmI₂, THF, –86 °C, 14 h, 91% combined; f) LiOH, H₂O₂, THF/H₂O, 4 °C, 2.5 h, 97%. DIBAL-H = diisobutylaluminum hydride, DMF = *N,N*-dimethylformamide.

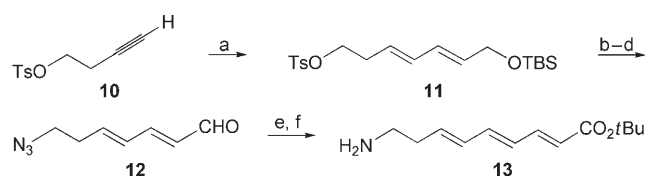
2*E*,4*E* diene ester **2** and its 2*Z*,4*E* isomer. A routine reduction–oxidation sequence from **2** with DIBAL-H and MnO₂ led to alcohol **3** and then dienal **4**^[10] in excellent overall yield. Alternatively, **4** could be obtained in a single step by the CrCl₂-induced condensation of **1** with 2,2-dichloropropanal (OMM Scientific).^[11] The samarium-mediated Reformatsky addition of (*S*)-4-benzyl-3-(2-bromoacetyl)oxazolidin-2-one^[12] (**9**) to **4** at low temperature produced predominately the 3*R* alcohol **5** (81%), which was hydrolyzed smoothly to the free acid **7**. Hydrolysis of the minor 3*S* diastereomer **6**, isolated in 10% yield, gave **8**.

Segment **B** (Scheme 3) was quickly stitched together by hydrozirconation of homopropargyl tosylate^[13] (**10**) and cross-coupling with the *E* vinylzirconocene derived from (*E*)-3-(*tert*-butyldimethylsilyloxy)-1-iodopropene^[14] (**14**) at room temperature in the presence of Pd(OAc)₂ and 2'-(dicyclohexylphosphanyl)-*N,N*-dimethyl-(1,1'-biphenyl)-2-amine^[15] (DPBA). The resulting *E,E* diene **11**, which was

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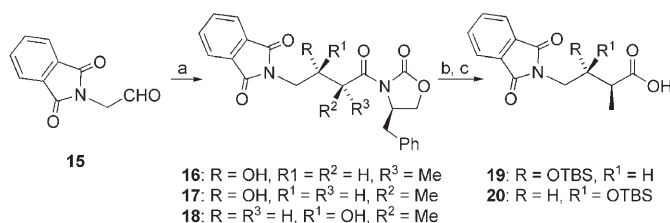
Supporting information for this article is available on the WWW under <http://www.angewandte.org> or from the author.



Scheme 3. Synthesis of amine **13**: a) 1. $[\text{Cp}_2\text{ZrClH}]$, THF, 23 °C, 1 h; 2. **14**, $\text{Pd}(\text{OAc})_2$, DPBA, THF, 23 °C, 11 h, 98%; b) NaN_3 , DMF, 23 °C, 14 h, 90%; c) $n\text{Bu}_4\text{NF}$, THF, 0 °C, 1 h; d) MnO_2 , CH_2Cl_2 , 23 °C, 14 h, 92% for two steps; e) $(\text{MeO})_2\text{P}(\text{O})\text{CH}_2\text{CO}_2\text{tBu}$, NaH, THF, 0 °C, 1 h, 95%; f) Ph_3P , THF/ H_2O (4:1), 23 °C, 20 h, 93%. TBS = *tert*-butyldimethylsilyl, Ts = *p*-toluenesulfonyl.

generated in excellent overall yield and with excellent stereoselectivity, was transformed into aldehyde **12** by treatment with sodium azide in DMF, followed by desilylation and oxidation with MnO_2 . Horner–Wadsworth–Emmons (HWE) homologation of **12** and Staudinger reduction of the azide delivered *tert*-butyl ester **13** exclusively in the all-*E* configuration, as judged by $^1\text{H}/^{13}\text{C}$ NMR spectroscopy.

On the basis of a review of the spectral data for FR252921^[8] and literature precedent for related substructures,^[4,9] we opted to prepare the *anti* 2*S*,3*R* stereoisomer and either *syn* enantiomer of segment **C** to complete the stereoarray (Scheme 4). To this end, the Evans asymmetric aldol

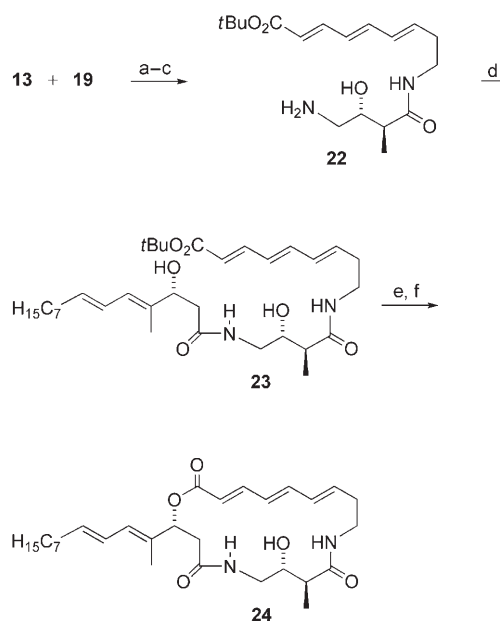


Scheme 4. Synthesis of carboxylic acids **19** and **20**: a) 1) **21**, $n\text{Bu}_2\text{BOTf}$ (2.0 equiv), $i\text{Pr}_2\text{NEt}$ (1.05 equiv), CH_2Cl_2 , 0 °C, 40 min; 2) **15**, –78 °C, 0.5 h, then 0 °C, 1 h, 90% (d.r. $\approx$ 1:1); b) TBSOTf (1.05 equiv), 2,6-lutidine, CH_2Cl_2 , 0 °C, 13 h, 99%; c) LiOH , H_2O_2 , THF/ H_2O (4:1), 4 °C, 17 h, 89%. Tf = trifluoromethanesulfonyl.

reaction^[16] emerged as the most effective of the many enantioselective procedures evaluated. In practice, the addition of (*R*)-4-benzyl-3-propionyloxazolidin-2-one^[17] (**21**) to 2-phthalimidoacetaldehyde^[18] (**15**) in the presence of equimolar amounts of $n\text{Bu}_2\text{BOTf}$ and $i\text{Pr}_2\text{NEt}$ gave the *syn* adduct **16** as a single diastereomer in 90% yield. With a two-fold excess of the boron triflate and the amine, the *anti* adduct **17** (absolute configuration confirmed by X-ray analysis, CCDC-634599) began to appear (**16**: 71%, **17**: 14%). As anticipated,^[19] the diastereoselectivity was reversed as the reagent ratio was skewed toward excess boron triflate, and a mixture of **17** and the non-Evans *syn* adduct **18** was formed. As our immediate objective was the preparation of both an *anti* isomer and a *syn* isomer, the conditions were adjusted to give an approximately 1:1 mixture of **17** and **18** in good yield. The two diastereomers were separated conveniently by fractional crystallization after rough chromatography of the crude

product mixture. Silylation of the secondary alcohol and removal of the chiral auxiliary then afforded the free acids **19** and **20** (Scheme 4).

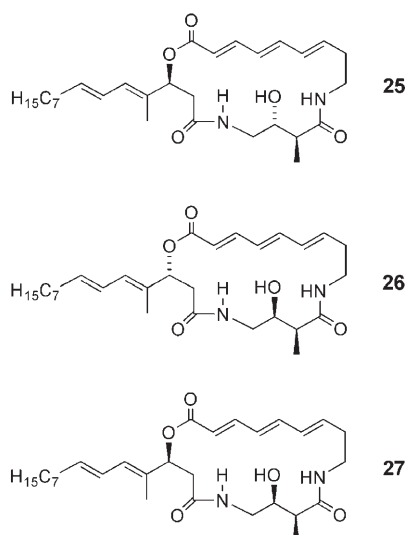
To assemble the macrocycle, amine **13** was united with the silyloxy acid **19** by using a suspension of *N*-(3-dimethylaminopropyl)-*N'*-ethylcarbodiimide (EDC) in CH_2Cl_2 (Scheme 5). Excess hydrazine and tetrabutylammonium fluo-



Scheme 5. Synthesis of macrocycle **24**: a) EDC, HOBT, CH_2Cl_2 , 23 °C, 2 h, 97%; b) $n\text{Bu}_4\text{NF}/\text{AcOH}$ (1.5:2), THF, 23 °C, 15 h, 91%; c) $\text{H}_2\text{NNH}_2 \cdot \text{H}_2\text{O}$ (50 equiv), EtOH, 45 °C, 5 min, $\approx$ 99% crude; d) **7**, EDC, HOBT, CH_2Cl_2 , 23 °C, 2.5 h, 92%; e) 1) TMSOTf, 2,6-lutidine, CH_2Cl_2 , 0 °C, 2.5 h; 2) citric acid, MeOH, 4 °C, 14 h, 91%; f) MNBA, DMAP, THF, 23 °C, 96 h, 10–15%. DMAP = 4-dimethylaminopyridine, HOBT = 1-hydroxy-1*H*-benzotriazole, TMS = trimethylsilyl.

ride were employed to remove the phthalidyl and silyl protecting groups, respectively.^[20] The product, aminoalcohol **22**, was then condensed with hydroxyacid **7** to afford the bisamide **23**. The transformation of **23** into the corresponding seco acid by cleavage of the *tert*-butyl ester according to the procedure described by Danishefsky and co-workers^[21] proceeded well and set the stage for the final ring closure.^[22] This final intermediate proved to be quite labile (perhaps because the two hydroxy groups are unprotected^[20]) and, consequently, compatible with only the mildest macrolactonization procedures.^[23] In particular, the procedure of Shiina et al.^[24] with 2-methyl-6-nitrobenzoic anhydride (MNBA) proved uniquely suitable and rewarded us with a satisfactory yield of **24**, which was isolated by reversed-phase preparatory TLC.

The remaining diastereomers **25**, **26**, and **27** (Scheme 6) were acquired uneventfully by repetition of the series of reactions in Scheme 5 with the segment combinations **8/13/19**, **7/13/20**, and **8/13/20**, respectively. Of the four macrocycles, only **24** was fully consistent with the natural material by $^1\text{H}/^{13}\text{C}$ NMR spectroscopy and HPLC, and with respect to its



Scheme 6. Diastereomers of FR252921.

melting point and optical rotation. Thus, we established that FR252921 has the configuration 12*S*,13*R*,18*R*.^[1]

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