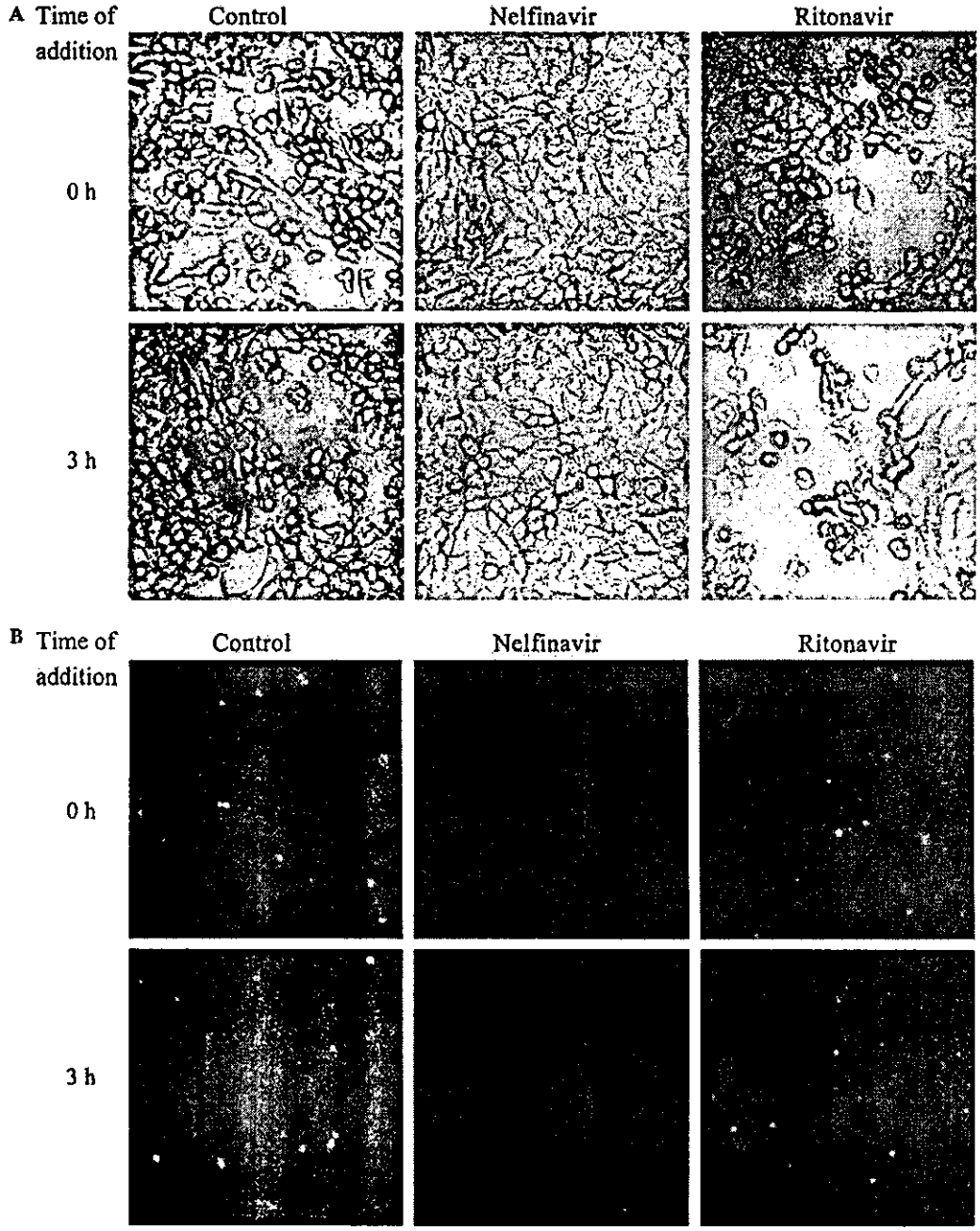


Fig. 3. Effect of various timings of drug addition on SARS-CoV replication in Vero E6 cells. (A) CPE in Vero E6 cells when drugs were added at the time of infection or 3 h after infection. The cells were treated with phosphate-buffered saline (PBS) as a control, 10 μM nelfinavir, or 10 μM ritonavir and CPE was observed 36 h after infection. (B) IFA of infected cells when drugs were added at the time of infection or 3 h after infection. The cells were treated with PBS as a control, 10 μM nelfinavir, or 10 μM ritonavir. The cells were fixed with methanol 24 h after infection and stained with serum samples from SARS patients.

(Figs. 3A and B and 4). These results indicate that the target(s) of nelfinavir may be involved in the post-entry step of SARS-CoV replication.

To investigate whether or not nelfinavir can affect the efficiency of virion entry, we quantified the copy number of SARS-CoV RNA in Vero cells immediately after the entry of virions.

Real-time RT-PCR revealed that nelfinavir did not affect the entry step of SARS-CoV infection (Fig. 5), which is consistent with our assumption that nelfinavir blocks the post-entry step of SARS-CoV replication.

The mechanisms that underlie the inhibitory action of nelfinavir on SARS-CoV replication remain to be identified. The main proteinase of SARS-CoV is one of

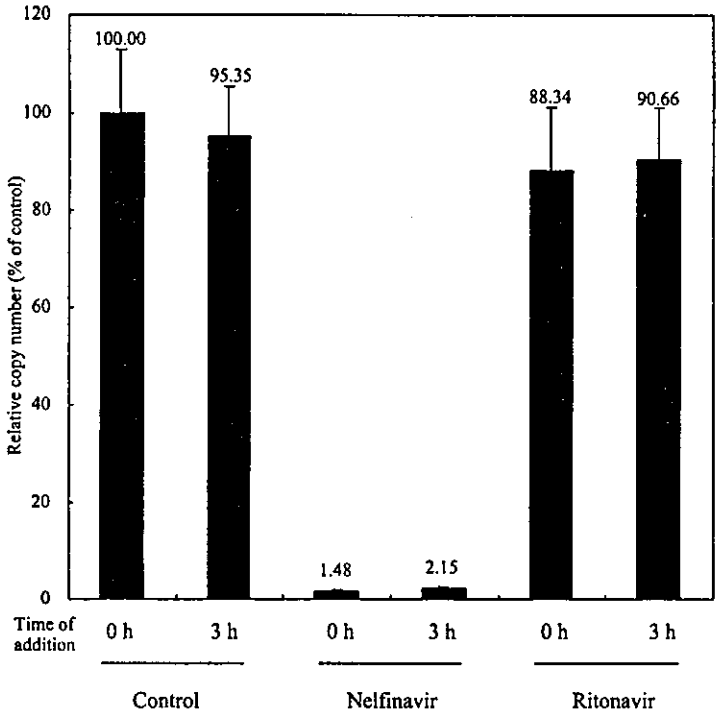


Fig. 4. Real-time RT-PCR for SARS-CoV RNA with various timings of drug addition. Nelfinavir or ritonavir was added at the time of infection or 3 h after infection at the concentration of 10 μM. Instead of these drugs PBS was added as a negative control. Viral RNA in the culture supernatant was collected 24 h after infection and quantified by the use of a fluorogenic probe. All samples were analyzed in triplicate.

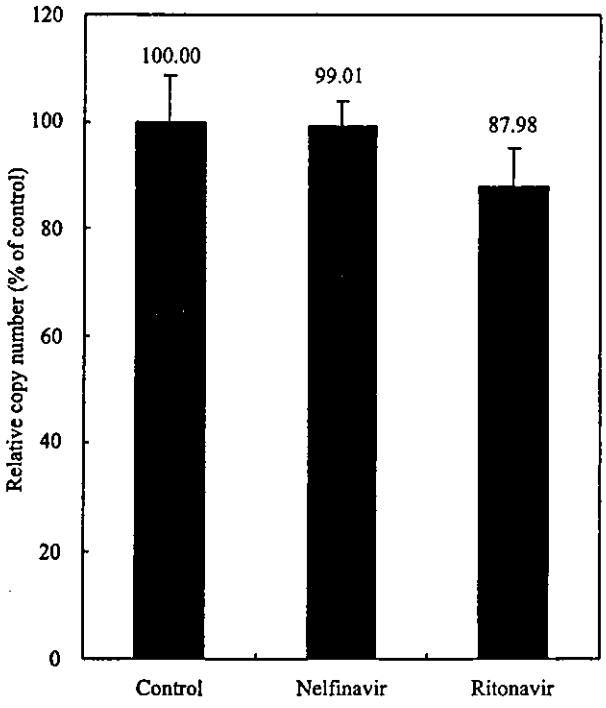


Fig. 5. Entry inhibition assay. To quantify SARS-CoV RNA which entered the cells, Vero E6 cells were pretreated with the drugs and infected with SARS-CoV. Cells were washed with PBS 3 times 3 h after infection. Subsequently viral RNA and 18S ribosomal RNA in the cells were quantified. All samples were analyzed in triplicate.

the molecules expressed after infection with its important role in viral replication [18–20], and the effect of nelfinavir on the main proteinase activity should be investigated. We have cloned, expressed, and purified SARS-CoV main proteinase in order to examine the effect of nelfinavir on this enzyme. Our preliminary study indicated that the activity of the main proteinase was blocked only partially (data not shown), which implies that nelfinavir may interact with some molecule(s) other than the main proteinase to fully inhibit SARS-CoV replication.

Nelfinavir is a very safe and widely used inhibitor of the HIV-1 protease, with strong in vivo activity in HIV-infected patients. Nelfinavir is generally used in combination with other antiretroviral medications as part of a highly active antiretroviral regimen (HAART) [21]. When used in this manner, 50–75% of patients who are naive to antiretroviral therapy have plasma HIV RNA levels below the limit of detection in association with an approximate increase of 200 mm^{-3} CD4(+) lymphocytes at 12 months of therapy [22–25]. The most common side effect of nelfinavir is mild diarrhea, which is observed in 15–20% of patients [26]. Nelfinavir is well tolerated by patients with HIV infection. Due to these characteristics, nelfinavir has become one of the most frequently prescribed first line protease inhibitors in the treatment of HIV-infected individuals.

Our studies have clearly shown that nelfinavir can strongly inhibit the replication of SARS-CoV in Vero E6 cells. The safety of this drug for humans has already been established, which constitutes the advantages of nelfinavir even for the clinical use to SARS patients. Our results suggest that nelfinavir should be examined clinically for the treatment of SARS. Moreover, nelfinavir might be a promising lead compound for anti-SARS drugs.

Acknowledgments

This work was supported by grants from the Ministry of Education, Science and Culture and the Ministry of Health, Labor and Welfare of Japan.

References

[1] N. Lee, D. Hui, A. Wu, P. Chan, P. Cameron, G.M. Joynt, A. Ahuja, M.Y. Yung, C.B. Leung, K.F. To, S.F. Lui, C.C. Szeto, S. Chung, J.J. Sung, A major outbreak of severe acute respiratory syndrome in Hong Kong, N. Engl. J. Med. 348 (2003) 1986–1994.

[2] S. M. Poutanen, D.E. Low, B. Henry, S. Finkelstein, D. Rose, K. Green, R. Tellier, R. Draker, D. Adachi, M. Ayers, A.K. Chan, D.M. Skowronski, I. Salit, A.E. Simor, A.S. Slutsky, P.W. Doyle, M. Krajden, M. Petric, R.C. Brunham, A.J. McGeer, C. National Microbiology Laboratory, T. Canadian Severe Acute Respiratory Syndrome Study, Identification of severe acute respiratory syndrome in Canada, N. Engl. J. Med. 348 (2003) 1995–2005.

[3] K.W. Tsang, P.L. Ho, G.C. Ooi, W.K. Yee, T. Wang, M. Chan-Yeung, W.K. Lam, W.H. Seto, L.Y. Yam, T.M. Cheung, P.C. Wong, B. Lam, M.S. Ip, J. Chan, K.Y. Yuen, K.N. Lai, A cluster of cases of severe acute respiratory syndrome in Hong Kong, N. Engl. J. Med. 348 (2003) 1977–1985.

[4] WHO, in, 2003.

[5] C.A. Donnelly, A.C. Ghani, G.M. Leung, A.J. Hedley, C. Fraser, S. Riley, L.J. Abu-Raddad, L.M. Ho, T.Q. Thach, P. Chau, K.P. Chan, T.H. Lam, L.Y. Tse, T. Tsang, S.H. Liu, J.H. Kong, E.M. Lau, N.M. Ferguson, R.M. Anderson, Epidemiological determinants of spread of causal agent of severe acute respiratory syndrome in Hong Kong, Lancet 361 (2003) 1761–1766.

[6] C. Drosten, S. Gunther, W. Preiser, S. van der Werf, H.R. Brodt, S. Becker, H. Rabenau, M. Panning, L. Kolesnikova, R.A. Fouchier, A. Berger, A.M. Burguiere, J. Cinatl, M. Eickmann, N. Escriou, K. Grywna, S. Kramme, J.C. Manuguerra, S. Muller, V. Rickerts, M. Sturmer, S. Vieth, H.D. Klenk, A.D. Osterhaus, H. Schmitz, H.W. Doerr, Identification of a novel coronavirus in patients with severe acute respiratory syndrome, N. Engl. J. Med. 348 (2003) 1967–1976.

[7] T.G. Ksiazek, D. Erdman, C.S. Goldsmith, S.R. Zaki, T. Peret, S. Emery, S. Tong, C. Urbani, J.A. Comer, W. Lim, P.E. Rollin, S.F. Dowell, A.E. Ling, C.D. Humphrey, W.J. Shieh, J. Guarner, C.D. Paddock, P. Rota, B. Fields, J. DeRisi, J.Y. Yang, N. Cox, J.M. Hughes, J.W. LeDuc, W.J. Bellini, L.J. Anderson, S.W. Group, A novel coronavirus associated with severe acute respiratory syndrome, N. Engl. J. Med. 348 (2003) 1953–1966.

[8] R.A. Fouchier, T. Kuiken, M. Schutten, G. van Amerongen, G.J. van Doornum, B.G. van den Hoogen, M. Peiris, W. Lim, K. Stohr, A.D. Osterhaus, Aetiology: Koch's postulates fulfilled for SARS virus, Nature 423 (2003) 240.

[9] W. Gao, A. Tamin, A. Soloff, L. D'Aiuto, E. Nwanegbo, P.D. Robbins, W.J. Bellini, S. Barratt-Boyes, A. Gambotto, Effects of a SARS-associated coronavirus vaccine in monkeys, Lancet 362 (2003) 1895–1896.

[10] L.K. So, A.C. Lau, L.Y. Yam, T.M. Cheung, E. Poon, R.W. Yung, K.Y. Yuen, Development of a standard treatment protocol for severe acute respiratory syndrome, Lancet 361 (2003) 1615–1617.

[11] K.L. Hon, C.W. Leung, W.T. Cheng, P.K. Chan, W.C. Chu, Y.W. Kwan, A.M. Li, N.C. Fong, P.C. Ng, M.C. Chiu, C.K. Li, J.S. Tam, T.F. Fok, Clinical presentations and outcome of severe acute respiratory syndrome in children, Lancet 361 (2003) 1701–1703.

[12] G. Zhaori, Antiviral treatment of SARS: can we draw any conclusions?, C.M.A.J. 169 (2003) 1165–1166.

[13] C.M. Booth, L.M. Matukas, G.A. Tomlinson, A.R. Rachlis, D.B. Rose, H.A. Dwosh, S.L. Walmsley, T. Mazzulli, M. Avendano, P. Derkach, I.E. Ephtimios, I. Kitai, B.D. Mederski, S.B. Shadowitz, W.L. Gold, L.A. Hawryluck, E. Rea, J.S. Chenkin, D.W. Cescon, S.M. Poutanen, A.S. Detsky, Clinical features and short-term outcomes of 144 patients with SARS in the greater Toronto area, Jama 289 (2003) 2801–2809.

[14] J. Cinatl, B. Morgenstern, G. Bauer, P. Chandra, H. Rabenau, H.W. Doerr, Treatment of SARS with human interferons, Lancet 362 (2003) 293–294.

[15] M.R. Loutfy, L.M. Blatt, K.A. Siminovitch, S. Ward, B. Wolff, H. Lho, D.H. Pham, H. Deif, E.A. LaMere, M. Chang, K.C. Kain, G.A. Farcas, P. Ferguson, M. Latchford, G. Levy, J.W. Dennis, E.K. Lai, E.N. Fish, Interferon alfacon-1 plus corticosteroids in severe acute respiratory syndrome: a preliminary study, Jama 290 (2003) 3222–3228.

[16] B.L. Haagmans, T. Kuiken, B.E. Martina, R.A. Fouchier, G.F. Rimmelzwaan, G. Van Amerongen, D. Van Riel, T. De Jong, S. Itamura, K.H. Chan, M. Tashiro, A.D. Osterhaus, Pegylated interferon-alpha protects type 1 pneumocytes against SARS coronavirus infection in macaques, Nat. Med. 10 (2004) 290–293.

[17] J. Cinatl, B. Morgenstern, G. Bauer, P. Chandra, H. Rabenau, H.W. Doerr, Glycyrrhizin, an active component of liquorice roots, and replication of SARS-associated coronavirus, Lancet 361 (2003) 2045–2046.

[18] V. Thiel, K.A. Ivanov, A. Putics, T. Hertzig, B. Schelle, S. Bayer, B. Weissbrich, E.J. Snijder, H. Rabenau, H.W. Doerr, A.E. Gorbalenya, J. Ziebuhr, Mechanisms and enzymes involved in SARS coronavirus genome expression, J. Gen. Virol. 84 (2003) 2305–2315.

[19] M.A. Marra, S.J. Jones, C.R. Astell, R.A. Holt, A. Brooks-Wilson, Y.S. Butterfield, J. Khattra, J.K. Asano, S.A. Barber, S.Y. Chan, A. Cloutier, S.M. Coughlin, D. Freeman, N. Girn, O.L. Griffith, S.R. Leach, M. Mayo, H. McDonald, S.B. Montgomery, P.K. Pandoh, A.S. Petrescu, A.G. Robertson, J.E. Schein, A. Siddiqui, D.E. Smailus, J.M. Stott, G.S. Yang, F. Plummer, A. Andonov, H. Artsob, N. Bastien, K. Bernard, T.F. Booth, D. Bowness, M. Czub, M. Drebot, L. Fernando, R. Flick, M. Garbutt, M. Gray, A. Grolla, S. Jones, H. Feldmann, A. Meyers, A. Kabani, Y. Li, S. Normand, U. Stroher, G.A. Tipples, S. Tyler, R. Vogrig, D. Ward, B. Watson, R.C. Brunham, M. Krajden, M. Petric, D.M. Skowronski, C. Upton, R.L. Roper, The genome sequence of the SARS-associated coronavirus, Science 300 (2003) 1399–1404.

[20] P.A. Rota, M.S. Oberste, S.S. Monroe, W.A. Nix, R. Campagnoli, J.P. Icenogle, S. Penaranda, B. Bankamp, K. Maher, M.H. Chen, S. Tong, A. Tamin, L. Lowe, M. Frace, J.L. DeRisi, Q. Chen, D. Wang, D.D. Erdman, T.C. Peret, C. Burns, T.G. Ksiazek, P.E. Rollin, A. Sanchez, S. Liffick, B. Holloway, J. Limor, K. McCaustland, M. Olsen-Rasmussen, R. Fouchier, S. Gunther, A.D. Osterhaus, C. Drosten, M.A. Pallansch, L.J. Anderson, W.J. Bellini, Characterization of a novel coronavirus associated with severe acute respiratory syndrome, Science 300 (2003) 1394–1399.

[21] J.S. Lewis 2nd, C.M. Terriff, D.R. Coulston, M.W. Garrison, Protease inhibitors: a therapeutic breakthrough for the treatment of patients with human immunodeficiency virus, Clin. Ther. 19 (1997) 187–214.
[22] G.P. Rizzardi, G. Tambussi, P.A. Bart, A.G. Chapuis, A. Lazzarin, G. Pantaleo, Virological and immunological responses to HAART in asymptomatic therapy-naive HIV-1-infected subjects according to CD4 cell count, Aids 14 (2000) 2257–2263.
[23] O. Kirk, J.D. Lundgren, C. Pedersen, L.R. Mathiesen, H. Nielsen, T.L. Katzenstein, N. Obel, J. Gerstoft, A randomized trial comparing initial HAART regimens of nelfinavir/nevirapine and ritonavir/saquinavir in combination with two nucleoside reverse transcriptase inhibitors, Antiviral Ther. 8 (2003) 595–602.
[24] R. Manfredi, L. Calza, F. Chiodo, Prospective comparison of first-line nelfinavir therapy versus nelfinavir introduction in rescue antiretroviral regimens, AIDS Patient Care Stud. 17 (2003) 105–114.
[25] S. Fleury, G.P. Rizzardi, A. Chapuis, G. Tambussi, C. Knabenhans, E. Simeoni, J.Y. Meuwly, J.M. Corpataux, A. Lazzarin, F. Miedema, G. Pantaleo, Long-term kinetics of T cell production in HIV-infected subjects treated with highly active antiretroviral therapy, Proc. Natl. Acad. Sci. USA 97 (2000) 5393–5398.
[26] S.W. Kaldor, V.J. Kalish, J.F. Davies 2nd, B.V. Shetty, J.E. Fritz, K. Appelt, J.A. Burgess, K.M. Campanale, N.Y. Chirgadze, D.K. Clawson, B.A. Dressman, S.D. Hatch, D.A. Khalil, M.B. Kosa, P.P. Lubbehusen, M.A. Muesing, A.K. Patick, S.H. Reich, K.S. Su, J.H. Tatlock, Viracept (nelfinavir mesylate, AG1343): a potent, orally bioavailable inhibitor of HIV-1 protease, J. Med. Chem. 40 (1997) 3979–3985.

Akira Otaka
Etsuko Mitsuyama
Junko Watanabe
Hideaki Watanabe
Nobutaka Fujii
Graduate School of Pharmaceutical Sciences, Kyoto University, Sakyo-ku, Kyoto 606-8501, Japan

Synthesis of Fluorine-Containing Bioisosteres Corresponding to Phosphoamino Acids and Dipeptide Units

Received 28 July 2003; accepted 1 October 2003
Published online 3 March 2004 in Wiley InterScience (www.interscience.wiley.com). DOI 10.1002/bip.10570

Abstract: It has been shown that fluorinated analogues of naturally occurring biological active compounds including amino acids often exhibit unique physiological activity. Among wide varieties of fluorine-containing amino acids, nonhydrolyzable phosphoamino acids possessing a substituent of the difluoromethylene (CF_2) unit for the phosphoryl ester oxygen are of value in the medicinal and biological fields. We have engaged in the synthesis of these classes of nonhydrolyzable phosphoamino acids corresponding to pTyr **3**, *pSer* **4**, *and pThr* **5** *with their incorporation into peptides using newly developed deprotecting procedures. In this article, stereoselective synthesis of the CF_2-substituted pThr mimetics and development of a two-step deprotecting methodology for the nonhydrolyzable analogues are reviewed. In the course of the above synthetic study, we found that γ,γ-difluoro-α,β-enoates were reduced to γ-fluoro-β,γ-enoates by organocopper reagents and then applied to the synthesis of (Z)-fluoroalkene dipeptide isosteres, which have served as potential dipeptide mimetics having structural as well as electrostatic similarity to the parent peptide bonds. Furthermore, mechanistic investigation of the organocopper-mediated reduction led us to development of a SmI_2-mediated approach toward the synthesis of the fluoroalkene isosteres.* © 2004 Wiley Periodicals, Inc. Biopolymers (Pept Sci) 76: 140–149, 2004

Keywords: fluorine-containing bioisosteres; phosphoamino acids; dipeptide units; alkene dipeptide isosteres; deprotection; stereoselective synthesis

INTRODUCTION

Bioisosteres corresponding to naturally occurring peptides or amino acids have recently received much attention due to their potential utility in medicinal and biological chemistry. Among the mimetics for amino acids or peptides, nonhydrolyzable phosphoamino acids[1–8] and (*E*)-alkene dipeptide isosteres (EADIs)[9–23] have become synthetic targets because of increasing demand for their mimetics in the development of peptide-lead drugs. In this article, we briefly review our synthetic studies on phosphatase-resistant phosphoamino acids corresponding to phosphothreonine (pThr) and its incorporation into a peptide with development of deprotecting methodologies suitable for phosphopeptides.[8] Additionally, application of an un-

Correspondence to: Akira Otaka; email: aotaka@pharm.kyoto-u.ac.jp
Contract grant sponsor: Ministry of Education, Culture, Sports, Science and Technology, Japan, the Japan Society for the Promotion of Science, and the Japan Health Science Foundation

© 2004 Wiley Periodicals, Inc.

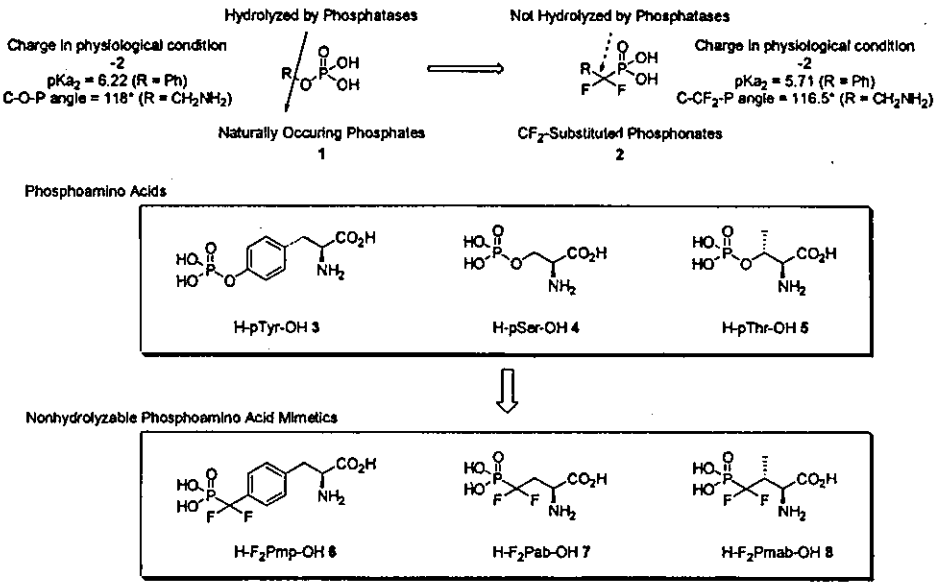


FIGURE 1 Naturally occurring phosphates and their CF_2-substituted analogues.

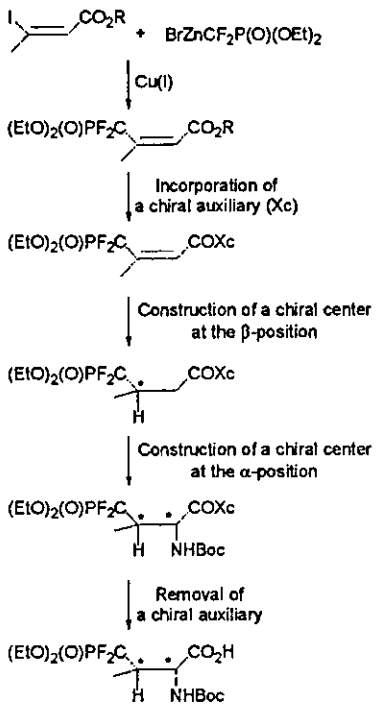
expected reaction using an organocopper reagent, which was found during the synthesis of a nonhydrolyzable phosphothreonine mimetic,[24] to the preparation of (Z)-fluoroalkene dipeptide isosteres,[25–31] is summarized.[32,33] The use of our newly found reaction for the preparation of the dipeptide isosteres provides an access to the isosteres conceptually distinct from the methodologies published in the literature.[27–31]

SYNTHESIS OF NONHYDROLYZABLE PHOSPHOAMINO ACIDS

Phosphorylation and dephosphorylation of proteins serve as posttranslational modifications that are critical for intracellular signal transduction.[34] Nonhydrolyzable phosphoamino acid-containing peptides have provided potential biochemical tools for evaluating the roles of phosphorylation events in cellular signaling.[35–40] It is worth noting that such phosphatase-resistant peptides can be used in a cellular system without addition of an inhibitor against a phosphatase, whereas naturally occurring phosphopeptides are easily hydrolyzed by the action of the phosphatases in the absence of an inhibitor. Among various nonhydrolyzable phosphoamino acid analogues, those employing the difluoromethylene unit (CF_2) as a replacement for the phosphoryl ester oxygen have shown particular utility.[41–44] (For a recent review of phosphoryltyrosyl mimetics, see Ref. 45.) Difluoromethylphosphonates **2** as a nonhydrolyzable phosphate mimetic have been well documented to be closely approximate to the phosphates **1** with regard to both bond angles (C–X–P: X = O or CF_2) and their pKa_2 values (Figure 1).[46–49]

Synthetic approaches toward CF_2-substituted phosphoamino acid mimetics by us and others have provided 4-phosphonodifluoromethyl phenylalanine (F_2Pmp **6**)[1–5] and 2-amino-4,4-difluoro-4-phosphonobutanoic acid (F_2Pab **7**)[6,7] as nonhydrolyzable pTyr and pSer mimetics, respectively. Alternatively, synthesis of 2-amino-4,4-difluoro-3-methyl-4-phosphonobutanoic acid (F_2Pmab **8**) as a CF_2-substituted pThr mimetic initially was lacking due to paucity of efficient methodologies for construction of the secondary CF_2 phosphonate unit. However, Berkowitz et al. did report a stereoselective synthesis of the F_2Pmab derivatives through the reaction of CH_3MgBr onto a keto difluoromethylphosphonate.[50] We have also examined alternative stereoselective approaches for the preparation of all four stereoisomers of the F_2Pmab derivatives.[8]

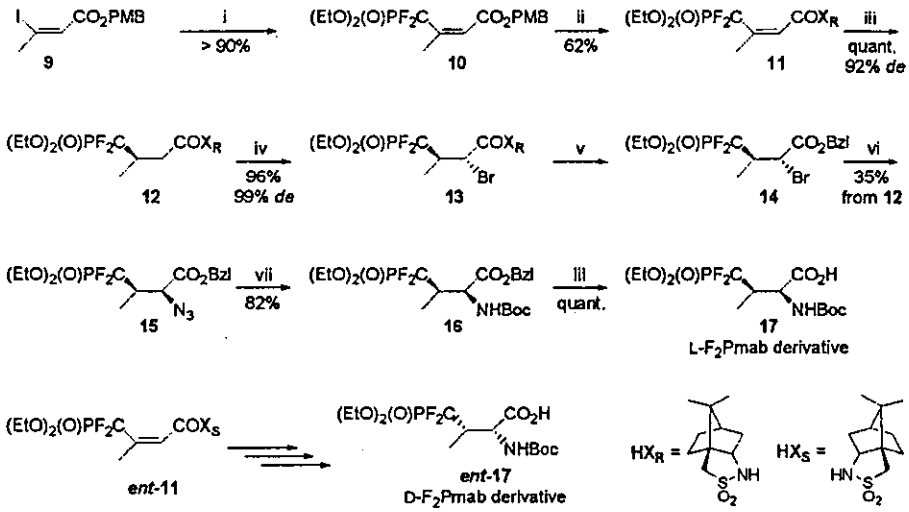
Our synthetic plan for the protected F_2Pmab derivatives is conceptually outlined in Scheme 1. The key to the synthesis of F_2Pmab derivatives was the construction of the secondary phosphate-mimicking difluoromethylphosphonate unit along with generation of two stereocenters. The construction of the secondary unit was achieved using a Cu(I)-mediated cross-coupling reaction of $BrZnCF_2P(O)(OEt)_2$ and β-iodo-



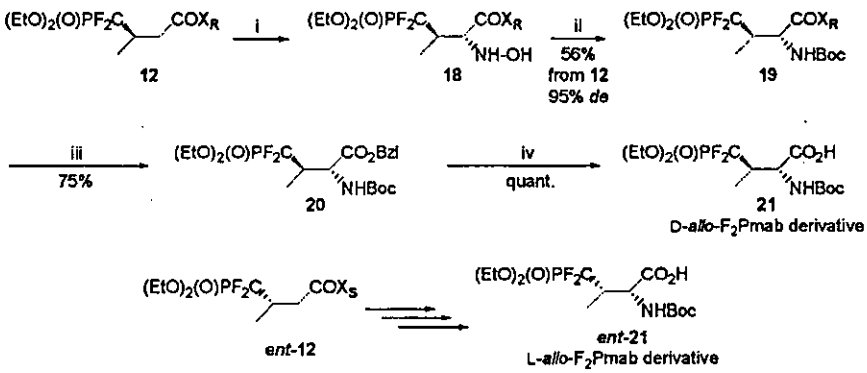
SCHEME 1 Synthetic plan for protected F_2Pmab derivatives.

α,β-unsaturated ester **9**,[51] with stereochemistry of both α- and β-stereocenters being established using bornane-10,2-sultam as a chiral auxiliary.[52,53] Synthetic routes for F_2Pmab (**17** and ***ent*-17**) and *allo*-F_2Pmab (**21** and ***ent*-21**) derivatives are shown in Scheme 2 and Scheme 3, respectively.{SCHEME2-3} Diastereoselective hydrogenation of a chiral α,β-unsaturated acylsultam **11** was applied to the generation of the β-center.[54] Subsequently, stereoselective bromination with N-bromosuccinimide (NBS) of sodium enolate resulting from β-difluoromethyl substituted acylsultam **12** was used for construction of the α-center of the *threo* derivative **17**.[55,56] Transesterification of the bromide **13** to the benzyl ester **14** followed by azide displacement of the halogen,[55–57] then reduction of the resulting azide **15**, followed by *tert*-butyloxycarbonyl (Boc) protection and finally removal of the benzyl group, afforded protected F_2Pmab derivative (**17** and ***ent*-17**) (Scheme 2).

An alternative synthetic route was employed for the generation of the β-center of *erythro* (*allo*) derivatives (**21** and ***ent*-21**) (Scheme 3), where a π-face selective electrophilic enolate amination using 1-chloro-1-nitrosocyclohexane as an NH_2^+ equivalent was utilized.[58] Deprotonation of chiral acylsultam **12** with N-sodichexamethyldisilazane (NaHMDS), followed by reaction with 1-chloro-1-nitrosocyclohexane and subsequent acid treatment, gave hydroxylamine derivative **18**. Reduction of the resulting com-



SCHEME 2 Synthesis of nonhydrolyzable pThr mimetics. Reagents: (i) $BrZnCF_2P(O)(OEt)_2$, CuBr, dimethylformamide (DMF); (ii) 95% TFAaq (TFA: trifluoroacetic acid), then LiXR, pivaloyl chloride, Et_3N, THF; (iii), H_2/Pd–C (palladium–carbon), EtOAc; (iv) N-sodiohexamethyldisilazane, NBS, THF; (v) $Ti(OiPr)_4$, Bzl-OH, toluene; (vi) tetramethylguanidinium azide, CH_3CN; (vii) Zn, AcOH (50 eq) then Et_3N, $(Boc)_2O$, CH_3CN.



SCHEME 3 Synthesis of nonhydrolyzable *allo*-pThr mimetics. Reagent: (i) NaHMDS, 1-chloro-1-nitrosocyclohexane, THF then 1 N HCl aq.; (ii) Zn, AcOH then $(Boc)_2O$, CH_3CN; (iii) $Ti(OiPr)_4$, Bzl–OH, toluene; (iv) H_2/Pd–C. EtOAc.

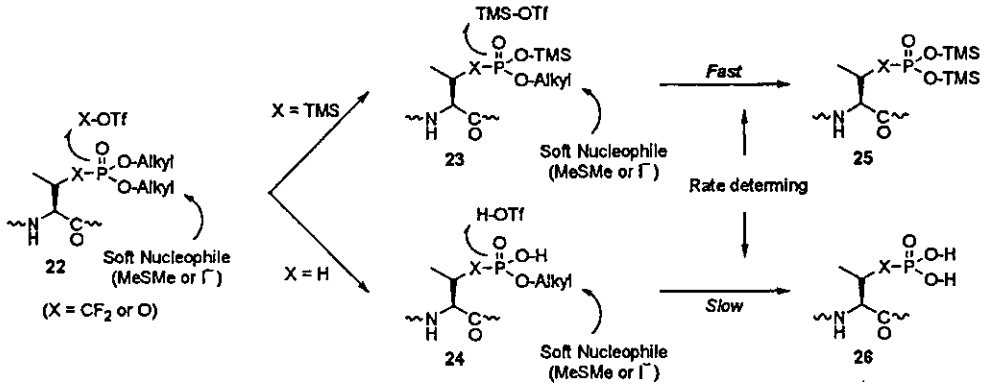
pound with Zn/AcOH, followed by a sequence of reactions consisting of Boc protection, transesterification, and hydrogenolytic debenzylation, afforded protected *allo*-F_2Pmab derivative (**21** and ***ent*-21**).

INCORPORATION OF NONHYDROLYZABLE PHOSPHOAMINO ACIDS INTO PEPTIDES USING A NEWLY DEVELOPED TWO-STEP DEPROTECTION PROTOCOL

In our synthesis of F_2Pmab derivatives, we employed side-chain ethyl protection similar to that used for F_2Pmp and F_2Pab, since the requisite difluoromethylphosphonate reagent bearing ethyl groups, $BrCF_2P(O)(OEt)_2$, is easily obtainable and cleanly converted to the corresponding Zn reagent. Furthermore, the ethyl protection is stable toward the synthetic transformations used in our study. However, ethyl phosphonate esters are difficult to deprotect using reagent systems commonly employed for peptide synthesis.[59] Increasing the acidity of deprotection reagents typically leads to facile removal of commonly used protecting groups. On the other hand, this is not the case with phosphonate- or phosphate-alkyl protecting groups (Table I).

Therefore, we developed a two-step deprotecting methodology consisting of high acidic (1*M* trimethylsilyl trifluoromethanesulfonate(TMSOTF)–thioanisole in TFA[60–62]), followed by low acidic (1*M* TMSOTf–thioanisole in TFA + TMSOTf–dimethyl sulfide) treatments. This protocol has been successfully applied to the synthesis of naturally occurring phosphopeptide using a combination of Boc chemistry and dimethyl-protected pTyr, pSer, and pThr derivatives.[63,64] Furthermore, application of this methodology to the deprotection of ethyl-protected F_2Pmp, or F_2Pab-containing, peptide resins affords fully deprotected peptides with some success. In developing the above two-step procedure, we have previously reported that operation under S_N2 conditions[65–68] is critical for removal of the phosphate or phosphonate protecting groups. Additionally, we have found that against dimethyl-protected phosphate containing peptides, replacement of TMSOTf by trifluoromethane sulfonic acid (TFMSA) results in substantial formation of incompletely deprotected peptides.[63,64] Deprotection of two alkyl groups on phosphoamino acids seems to involve the monoalkylated species (**23** or **24**) as an intermediate and the conversion of the monoalkyl intermediates to fully deprotected forms (**25** or **26**) is rate determining. Therefore, differences between TMSOTf- and TFMSA-based systems may be attributed to structural variations in intermediary monoalkyl-protected forms (**23** vs **24**). That is, the silylated intermediate **23** resulting from TMSOTf treatment may more easily undergo nucleophilic attack by sulfides to form the fully deprotected product than nonsilylated intermediate **24** derived from the TFMSA-based reagent system (structure in Table I).[69] From these assumptions, we speculated that a deprotecting system capable of silylating a monoalkyl intermediate as well as inducing nucleophilic attack, could potentially serve as highly efficient deprotecting methodology for ethyl-protected CF_2–phosphonate derivatives.[70,71] We therefore examined deprotection systems suitable for removal of the ethyl groups on F_2Pmab operating under S_N2 conditions based on silylation (silylation of monoalkyl phosphonates)–deprotection (nucleophilic deprotection with soft nucleophiles) concepts.[8] Consequently, 0.3*M* BSTFA

Table I Chemical Behavior of Alkyl-Protecting Group of Phosphoamino Acids[a]



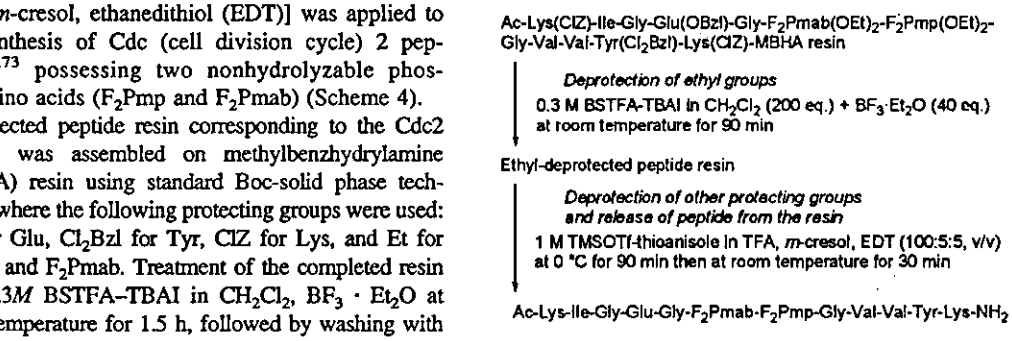
	Removal of Protecting Groups		
	Low Acidic Condition (S_N2, Hard acid)	Low Acidic Condition (S_N2, H^+ Acid)	High Acidic Condition (S_N1/S_N2 or SN1)
Alkyl groups on phosphonate	○	△	×
Protecting groups in peptide chemistry	×	×	○

[*N*,*O*-bis(trimethylsilyl) trifluoroacetamide)–TBAI (tetrabutylamminium iodide) (molar ratio 1:1) in CH_2Cl_2 in the presence of $BF_3 \cdot Et_2O$ was proved to be suitable for removal of ethyl groups on F_2Pmab residues, where BSTFA, TBAI, and BF_3 work as silylating agent, soft nucleophile, and activator of BSTFA, respectively. Alternatively, TMSOTf–DMS–*m*-cresol in CH_2Cl_2 (4:6:1:10, v/v) is also applicable to the removal of the ethyl groups. The newly developed two-step deprotection methodology consisting of a combination of a first-step reagent [0.3*M* BSTFA–TBAI in CH_2Cl_2, $BF_3 \cdot Et_2O$] followed by a second-step reagent [1*M* TMSOTf–thioanisole in TFA, *m*-cresol, ethanedithiol (EDT)] was applied to the synthesis of Cdc (cell division cycle) 2 peptide,[72,73] possessing two nonhydrolyzable phosphoamino acids (F_2Pmp and F_2Pmab) (Scheme 4).

Protected peptide resin corresponding to the Cdc2 peptide was assembled on methylbenzhydrylamine (MBHA) resin using standard Boc-solid phase technique, where the following protecting groups were used: Bzl for Glu, Cl_2Bzl for Tyr, ClZ for Lys, and Et for F_2Pmp and F_2Pmab. Treatment of the completed resin with 0.3*M* BSTFA–TBAI in CH_2Cl_2, $BF_3 \cdot Et_2O$ at room temperature for 1.5 h, followed by washing with solvent, and exposure to 1*M* TMSOTf–thioanisole in TFA, *m*-cresol, EDT at 4°C for 1.5 h with additional stirring for 0.5 h at room temperature, resulted in the release of the completely deprotected peptide. High performance liquid chromatography (HPLC) purification of the crude peptide furnished the purified Cdc2 peptide in 25% yield based on the protected peptide resin.

AN UNEXPECTED ORGANOCOPPER-MEDIATED REACTION FOUND DURING THE STUDY ON THE SYNTHESIS OF F_2Pmab DERIVATIVES

As mentioned before, F_2Pmab derivatives as nonhydrolyzable pThr mimetic have a secondary difluoro-



SCHEME 4 Deprotection for the synthesis of nonhydrolyzable Cdc2 peptide.

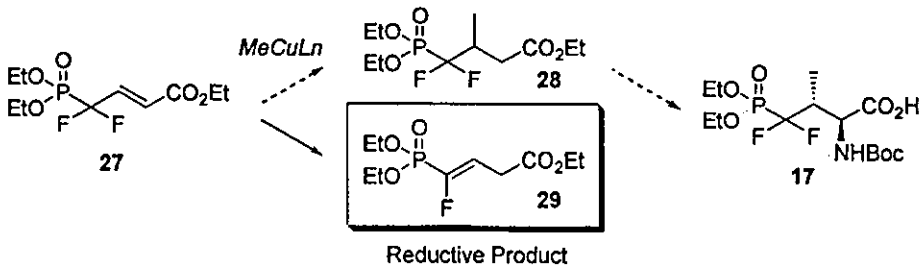


FIGURE 2 Organocopper-mediated reduction of γ-phosphono-γ,γ-difluoro-α,β-enoate.

methylphosphonate unit. In our stereoselective synthesis of F_2Pmab, the Cu(I)-mediated cross-coupling reaction of $BrZnCF_2P(O)(OEt)_2$ and β-iodo-α,β-unsaturated ester was finally used for the construction of the unit. During the course of our survey of potential synthetic procedures for the secondary unit, we attempted the conjugated addition of a methyl copper reagent to γ-phosphono-γ,γ-difluoro-α,β-enoate **27** to introduce the methyl group on the β-position (for recent reviews, see Refs. 74 and 75). The attempted reaction proceeded smoothly not to afford the desired conjugated addition product **28**, but to give an organocopper-mediated reduction product, α-fluorovinylphosphonate **29** (Figure 2).[24]

This finding is the first example of copper-mediated reduction of molecules containing the γ,γ-difluoro-α,β-enoate moiety where one fluorine works as the leaving group (Kitazume's group investigated organocopper-mediated reduction of a γ,γ,γ-trifluoro-α,β-enoate; see Ref. 76). This newly found reaction was first successfully applied to the synthesis of monofluoromethyl (CHF) -substituted nonhydrolyzable pSer mimetics.[24] We then speculated that this organocopper-mediated reaction should be applicable to molecules possessing substituents other than the phosphono group, to afford γ-fluoro-βγ-enoates. This hypothesis encouraged us to examine the feasibility of this unprecedented reduction to the synthesis of fluoroalkene dipeptide isosteres.

SYNTHESIS OF (Z)-FLUOROALKENE DIPEPTIDE ISOSTERES

Dipeptide isosteres possessing nonhydrolyzable scaffolds as replacement for scissile peptide bonds represent important constituents in peptidomimetics for medicinal and biological use.[77] Among several dipeptide mimetics, (*E*)-alkene dipeptide isosteres (EADIs) **30**, feature three-dimensional structures closely approximating the parent peptide bonds; however, certain intrinsic properties of amide bonds such as dipole interaction and hydrogen bonding are lacking. Therefore, (*Z*)-fluoroalkene dipeptide isosteres **31** have gained much attention as more suitable mimetics for alkene-type isosteres.[25,26,78–80](See Figure 3.)

Allmendinger's pioneering study on fluoroalkene isosteres employed aldol reaction of α-fluoro-α,β-unsaturated aldehydes with ester enolates, followed by the introduction of nitrogen functionality by an Overman rearrangement.[26,27] Conceptually distinct from the above-published methodology, the use of organocopper-mediated reduction of γ,γ-difluoro-α,β-enoates provides a new access to the fluoroalkene isosteres.[32,33] (Treatment of γ,γ-difluoro-α,β-enoates with R_3Al–Cu(I) reagent proceeds in an S_N2' manner to afford α-alkylated γ-fluoro-β,γ-enoates and related work; see Refs. 81–83.)

The requisite substrates **38** were synthesized by a sequence of reactions as follows (Scheme 5): (1) aldol reaction of appropriate aldehydes **32** with

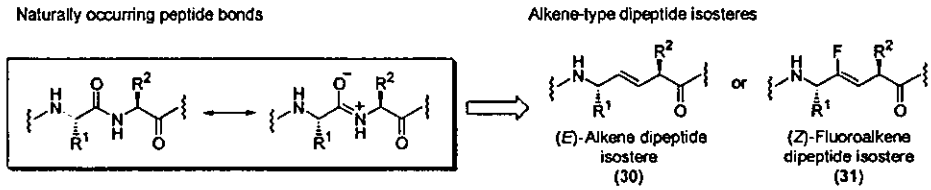
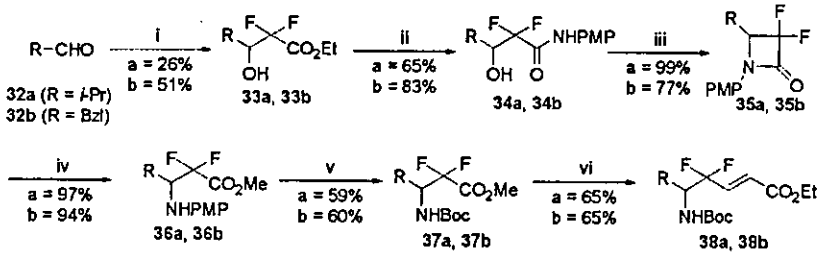


FIGURE 3 Naturally occurring peptide bond and corresponding alkene-type isosteres.



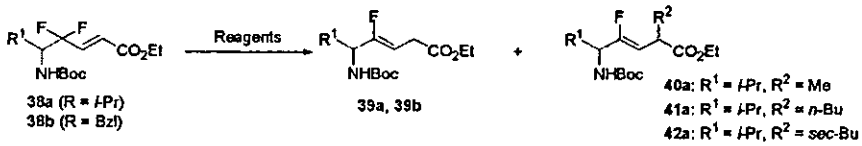
SCHEME 5 Reagents: (i) $BrZnCF_2CO_2Et$, THF; (ii) NaOH, THF–H_2O then BOP-Cl, *p*-anisidine, N,N-diisopropylethylamine (DIPEA), CH_2Cl_2; (iii) Ph_3P, diethyl azodicarboxylate (DEAD), THF; (iv) NaOH, THF–H_2O then H_2SO_4, MeOH; (v) CAN, CH_3CN-H_2O then $(Boc)_2O$, THF; (vi) diisobutylaluminium hydride(DIBAL)-H, CH_2Cl_2–toluene, then $(EtO)_2P(O)CH_2CO_2Et$, LiCl, DIPEA, CH_3CN.

$BrZnCF_2CO_2Et$; (2) conversion of the resulting esters **33** to the amide derivatives **34**; (3) Mitsunobu reaction followed by hydrolysis of the resulting lactams **35** for transformation of the hydroxyl group to the amine functionality; (4) esterification followed by exchange of the *N*-PMP (*p*-methoxyphenyl) group to a Boc group; (5) diisobutylaluminium hydride reduction of the resulting esters **37**, followed by Horner–Wadsworth–Emmons (HWE) olefination.

Reactions of the obtained δ-amino-γ,γ-difluoro-α,β-enoates **38** possessing (*E*)-geometry under several reaction conditions are summarized in Table II. First, the enoates **38** were subjected to the reaction with $Me_2Cu(CN)Li_2 \cdot 2LiCl \cdot 2LiBr$ in THF–Et_2O at −78°C for 15 min (Table II, entries 1 and 2). These reactions proceeded unequivocally to yield the desired (*Z*)-fluoroalkene dipeptide isosteres, Boc–Val–ψ[(*Z*)–CF=CH]–Gly–OH **39a** (95% isolated yield) and Boc–Phe–ψ[(*Z*)–CF=CH]–Gly–OH **39b** (85% isolated yield).[32] Although the synthetic procedure mentioned above provides a new access to fluoroalkene dipeptide isosteres, the obtained product was limited to Xaa–Gly types. We then examined the feasibility of copper-mediated procedures for the synthesis of α-substituted (*Z*)-fluoroalkene dipeptide isosteres (Table II, entries 3–6).

We explored a methodology for α-alkylation based on the reaction mechanism of the organocopper-mediated reduction. Recently, single electron transfer (SET) from an organocopper to a substrate has been reported to be involved with highly electrophilic trimethoxycarbonylethylene in the formation of the corresponding reduction product.[84,85] This work prompted us to envision that the formation of reduction products from highly electrophilic γ,γ-difluoro-α,β-enoates with organocopper reagents would be

Table II Reactions of Enoates 38 under Several Reaction Conditions



Entry	Substrate	Reagent	Condition	Product (Isolated Yield %)
1	**38a**	$Me_2Cu(CN)Li_2 \cdot 2LiBr \cdot 2LiCl$	−78°C, 10 min	**39a** (95)
2	**38b**	$Me_2Cu(CN)Li_2 \cdot 2LiBr \cdot 2LiCl$	−78°C, 10 min	**39b** (85)
3	**38a**	$Me_2Cu(CN)Li_2 \cdot 2LiBr \cdot 2LiCl$	−78°C, 4 min then −78°C, 20 min under O_2	**40a** (64)
4	**38a**	$Me_3Cu(CN)Li_3 \cdot 2LiBr \cdot 3LiCl$	−78°C, 5 min then −78°C, 30 min under O_2	**40a** (74)
5	**38a**	$n\text{-}Bu_2Cu(CN)Li_2 \cdot 2LiCl$	−78°C, 7 min then −78°C, 30 min under O_2	**41a** (54)
6	**38a**	$sec\text{-}Bu_2Cu(CN)Li_2 \cdot 2LiCl$	−78°C, 8 min then −78°C, 20 min under O_2	**42a** (45)
7	**38a**	SmI_2 + *t*-BuOH	0° C, 60 min	**39a** (85)
8	**38b**	SmI_2 + *t*-BuOH	0°C, 60 min	**39b** (92)

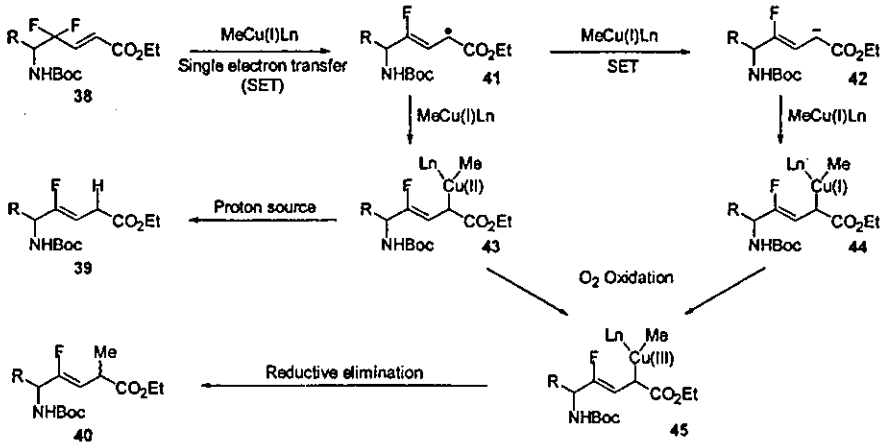


FIGURE 4 Plausible mechanisms both for the reduction and for α-alkylation via reduction-oxidative alkylation.

likely to proceed via the SET mechanism. And we speculated that involvement of the SET mechanism in the reduction allowed the reduction followed by oxidation to proceed via a mechanism shown in Figure 4 to give an α-substituted isostere. In this process, single electron transfer(s) to the substrate **38**, followed by recombination with the copper reagent, give stable Cu(II) **43** or Cu(I) **44** species quenched with proton to yield the reduction product **39**. Alternatively, oxidation of the intermediate with O_2 gives unstable Cu(III) species **45**, resulting in the formation of Me-substituted product **40** via reductive elimination.

Based on this assumption, we examined the preparation of α-substituted fluoroalkene isosteres using reduction-oxidative alkylation (R-OA) reactions with organocopper-treatment followed by O_2 oxidation. Treatment of the substrate with $Me_2Cu(CN)Li_2 \cdot 2LiCl \cdot 2LiBr$ in THF–Et_2O at −78°C under an Ar atmosphere for 4 min, followed by reaction under O_2 atmosphere at −78°C for 20 min proceeded nonstereoselectively to afford the corresponding α-methylated product **40a** in 64% yield (Table II, entry 3). The use of higher species [$Me_3Cu(CN)Li_3$] improved the yield (74%) (Table II, entry 4). Reaction with other alkyl copper reagents derived from *n*-BuLi or *sec*-BuLi, followed by O_2 oxidation, also gave α-substituted fluoroalkene isosteres (**41a** and **42a**, Table II, entry 5 and 6).[33]

Samarium (II) diiodide (SmI_2) has been well recognized as a powerful one-electron reducing agent capable of meeting the intensifying demands of synthetic organic chemistry (for some recent reviews, see Refs. 86–90). As mentioned above, taking account of our mechanistic insight into the organocopper-mediated reduction, we speculated that SmI_2 would be also applicable to the reduction of γ,γ-difluoro-α,β-enoates **38** to give fluoroalkene dipeptide isosteres. Based on this speculation, we examined the feasibility of the SmI_2-mediated reduction of **38** with application to the synthesis of (*Z*)-fluoroalkene dipeptide isosteres. Reaction of the substrate with SmI_2 in the presence of *t*-BuOH proceeded quantitatively to yield the desired (*Z*)-fluoroalkene isosteres in high chemical yields (Table II, entries 7 and 8). In this reaction, addition of a proton source such as *t*-BuOH is critical for clean conversion of the substrate to the desired isosteres. In analogy to the proposed reaction mechanism for reduction of γ,δ-epoxy-α,β-enoates with SmI_2,[91–95] a pathway via dienolates resulting from successive two-electron transfer is likely to be involved in the reduction of the γ,γ-difluoro-α,β-enaotes with SmI_2. And the proton source seems to work as kinetic trapping agents for the plausible dienolate intermediate. Additionally, replacement of the *t*-BuOH kinetic trapping agent with aldehydes or ketones provided access to α-substituted fluoroalkene isosteres through aldol reactions of Sm-dienolates with carbonyl compounds. Studies on the use of carbonyl compounds as a kinetic trapping agent with application to the synthesis of α-substituted fluoroalkene isosteres will be published in due course.[96] Furthermore, both the SmI_2-proton source and SmI_2-carbonyl compound systems have great synthetic applicability in not only the synthesis of fluoroalkene isosteres but also various kinds of amino acid derivatives.[97]

In summary, stereoselective synthesis of nonhydrolyzable pThr mimetics (F_2Pmab) and development of the two-step deprotection protocol based on the silylation–deprotection concept allowed us to prepare

F_2Pmab-containing peptides. Synthetic studies on CF_2-substituted phosphoamino acids and phosphopeptides provides an access to the compounds of value in exploring the intracellular signaling mechanism. Additionally, based on the unprecedented reaction of γ,γ-difluoro-α,β-enoates with organocopper reagents or SmI_2, we have devised facile methodologies for the synthesis of (*Z*)-fluoroalkene dipeptide isosteres which are potential dipeptide mimetics in developing peptide-lead drugs.

We thank Dr. Charles M. Deber for allowing us to present our results in this special issue. These studies were supported in part by a Grant-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science and Technology, Japan, the Japan Society for the Promotion of Science, and the Japan Health Science Foundation.

REFERENCES

1. Burke, T. R., Jr.; Smyth, M. S.; Nomizu, M.; Otaka, A.; Roller, P. P. J Org Chem 1993, 58, 1336–1340.
2. Burke, T. R., Jr.; Smyth, M. S.; Otaka, A.; Roller, P. P. Tetrahedron Lett 1993, 34, 4125–4128.
3. Smyth, M. S.; Burke, T. R., Jr.; Tetrahedron Lett 1994, 35, 551–554.
4. Wrobel, J.; Dietrich, A. Tetrahedron Lett 1993, 34, 3543–3546.
5. Solas, D.; Hale, R. L.; Patel, D. V. J Org Chem 1996, 61, 1537–1539.
6. Otaka, A.; Miyoshi, K.; Burke, T. R., Jr.; Roller, P. P.; Kubota, H.; Tamamura, H.; Fujii, N. Tetrahedron Lett 1995, 36, 927–930.
7. Berkowitz, D. B.; Shen, Q.; Maeng, J.-H. Tetrahedron Lett 1994, 35, 6445–6448.
8. Otaka, A.; Mitsuyama, E.; Kinoshita, T.; Tamamura, H.; Fujii, N. J Org Chem 2000, 65, 4888–4899.
9. Ibuka, T.; Habashita, H.; Otaka, A.; Fujii, N.; Oguchi, Y.; Uyehara, T.; Yamamoto, Y. J Org Chem 1991, 56, 4370–4382.
10. Fujii, N.; Nakai, K.; Habashita, H.; Yoshizawa, H.; Ibuka, T.; Garrido, F.; Mann, A.; Chounan, Y.; Yamamoto, Y. Tetrahedron Lett 1993, 34, 4227–4230.
11. Daly, M. J.; Ward, R. A.; Thompson, D. A.; Procter, G. Tetrahedron Lett 1995, 36, 7545–7548.
12. Kessler, H.; Kranz, M. Tetrahedron Lett 1996, 37, 5359–5362.
13. Ibuka, T.; Nakai, K.; Habashita, H.; Hotta, Y.; Fujii, N.; Mimura, N.; Miwa, Y.; Taga, T.; Yamamoto, Y. Angew Chem Int Ed Engl 1994, 33, 652–654.
14. Wipf, P.; Fritch, P. C. J Org Chem 1994, 59, 4875–4886.
15. Fujii, N.; Nakai, K.; Tamamura, H.; Otaka, A.; Mimura, N.; Miwa, Y.; Taga, T.; Yamamoto, Y.; Ibuka, T. J Chem Soc, Perkin Trans 1 1995, 1359–1371.
16. Wipf, P.; Henninger, T. C. J Org Chem 1997, 62, 1586–1587.
17. Tamamura, H.; Yamashita, M.; Muramatsu, H.; Ohno, H.; Ibuka, T.; Otaka, A.; Fujii, N. Chem Commun 1997, 2327.
18. Tamamura, H.; Yamashita, M.; Nakajima, Y.; Sakano, K.; Otaka, A.; Ohno, H.; Ibuka, T.; Fujii, N. J Chem Soc, Perkin Trans 1 1999, 2983–2996.
19. Oishi, S.; Tamamura, H.; Yamashita, M.; Hamanaka, N.; Otaka, A.; Fujii, N. J Chem Soc, Perkin Trans 1 2001, 2445–2451.
20. Henninger, T. C.; Wipf, P. In Peptidomimetics Protocols; Kazmierski, W. M., Ed.; Humana Press: Totowa, NJ, 1999, 125–136.
21. Oishi, S.; Niida, A.; Kamano, T.; Miwa, Y.; Taga, T.; Odagaki, Y.; Hamanaka, N.; Yamamoto, M.; Ajito, K.; Tamamura, H.; Otaka, A.; Fujii, N. J Chem Soc, Perkin Trans 1, 2002, 1786–1793.
22. Otaka, A.; Katagiri, F.; Kinoshita, T.; Odagaki, Y.; Oishi, S.; Tamamura, H.; Hamanaka, N.; Fujii, N. J Org Chem 2002, 67, 6152–6161.
23. Oishi, S.; Kamano, T.; Niida, A.; Odagaki, Y.; Hamanaka, N.; Yamamoto, M.; Ajito, K.; Tamamura, H.; Otaka, A.; Fujii, N. J Org Chem 2002, 67, 6162–6173.
24. Otaka, A.; Mitsuyama, E.; Watanabe, H.; Tamamura, H.; Fujii, N. Chem Commun 2000, 1081–1082.
25. Abraham, R. J.; Ellison, S. L. R.; Schonholzer, P.; Thomas, W. A. Tetrahedron 1986, 42, 2101–2110.
26. Allmendinger, T.; Furet, P.; Hungerbühler, E. Tetrahedron Lett 1990, 31, 7297–7300.
27. Allmendinger, T.; Felder, E.; Hungerbühler, E. Tetrahedron Lett 1990, 31, 7301–7304.
28. Boros, L. G.; De Corte, B.; Gimi, R. H.; Welch, J. T.; Wu, Y.; Handschumacher, R. E. Tetrahedron Lett 1994, 35, 6033–6036.
29. Bartlett, P. A.; Otake, A. J Org Chem 1995, 60, 3107–3111.
30. Welch, J. T.; Lin, J. Tetrahedron 1996, 52, 291–304.
31. Welch, J. T.; Allmendinger, T. In Peptidomimetics Protocols; Kazmierski, W. M., Ed.; Humana Press: Totowa, NJ, 1999; pp 357–384.
32. Otaka, A.; Watanabe, H.; Mitsuyama, E.; Yukimasa, A.; Tamamura, H.; Fujii, N. Tetrahedron Lett 2001, 42, 285–287.
33. Otaka, A.; Watanabe, H.; Yukimasa, A.; Oishi, S.; Tamamura, H.; Fujii, N. Tetrahedron Lett 2001, 42, 5443–5446.
34. Hunter, T. Cell 2000, 100, 113–127.
35. Burke, T. R., Jr.; Russ, P.; Lim, B. Synthesis 1991, 1019–1020.
36. Shoelson S. E.; Chatterjee, S.; Chaudhuri, M.; Burke, T. R., Jr. Tetrahedron Lett 1991, 32, 6061–6064.
37. Cushman, M.; Lee, E.-S. Tetrahedron Lett 1992, 33, 1193–1196.
38. Ojea, V.; Ruiz, M.; Shapiro, G.; Pombo-Villar, E. Tetrahedron Lett 1994, 35, 3273–3276.
39. Ruiz, M.; Ojea, V.; Shapiro, G.; Weber, H.-P.; Pombo-Villar, E. Tetrahedron Lett 1994, 35, 4551–4554.
40. Nomizu, M.; Otaka, A.; Burke, T. R., Jr.; Roller, P. P. Tetrahedron 1994, 50, 2691–2702.

41. Burke, T. R., Jr.; Smyth, M. S.; Otaka, A.; Nomizu, M.; Roller, P. P.; Wolf, G.; Case, R.; Shoelson, S. E. Biochemistry 1994, 33, 6490–6494.
42. Wange, R. L.; Isakov, N.; Burke, T. R., Jr.; Otaka, A.; Roller, P. P.; Watts, J. D.; Aebersold, R.; Samelson, L. E. J Biol Chem 1995, 270, 944–948.
43. Burke, T. R., Jr.; Kole, H. K.; Roller, P. P. Biochem Biophys Res Commun 1994, 204, 129–134.
44. Chen, L.; Wu, L.; Otaka, A.; Smyth, M. S.; Roller, P. P.; Burke, T. R., Jr.; Hertog, J.; Zhang, Z.-Y. Biochem Biophys Res Commun 1995, 216, 976–985.
45. Burke, T. R., Jr.; Lee, K. Acc Chem Res 2003, 36, 426–433.
46. Blackburn, G. M. Chem Ind (London) 1981, 134–138.
47. Chambers, R. D.; O'Hagan, D.; Lamont, R. B.; Jain, S. C. J Chem Soc, Chem Commun 1990, 1053–1054.
48. O'Hagan, D.; Rzepa, H. S. Chem Commun 1997, 645–652.
49. Smyth, M. S.; Ford, H., Jr.; Burke, T. R., Jr. Tetrahedron Lett 1992, 33, 4137–4140.
50. Berkowitz, D. B.; Eggen, M.; Shen, Q.; Shoemaker, R. K. J Org Chem 1996, 61, 4666–4675.
51. Yokomatsu, T.; Suemune, K.; Murano, T.; Shibuya, S. J Org Chem 1996, 61, 7207–7211.
52. Oppolzer, W. Pure Appl Chem 1990, 62, 1241–1250.
53. Oppolzer, W. Tetrahedron 1987, 43, 1969–2004.
54. Oppolzer, W.; Mills, R. J.; Réglier, M. Tetrahedron Lett 1986, 27, 183–186.
55. Evans, D. A.; Britton, T. C.; Ellman, J. A.; Dorow, R. L. J Am Chem Soc 1990, 112, 4011–4030.
56. Nicolás, E.; Russell, K. C.; Knollenberg, J.; Hruby, V. J. J Org Chem 1993, 58, 7565–7571.
57. Papa, A. J. J Org Chem 1966, 31, 1426–1430.
58. Oppolzer, W.; Tamura, O.; Deerberg, J. Helv Chim Acta 1992, 75, 1965–1978.
59. Otaka, A.; Burke, T. R., Jr.; Smyth, M. S.; Nomizu, M.; Roller, P. P. Tetrahedron Lett 1993, 34, 7039–7042.
60. Fujii, N.; Otaka, A.; Ikemura, O.; Akaji, K.; Funakoshi, S.; Hayashi, Y.; Kuroda, Y.; Yajima, H. J Chem Soc, Chem Commun 1987, 274–275.
61. Fujii, N.; Otaka, A.; Ikemura, O.; Hatano. M.; Okamachi, A.; Funakoshi, S.; Sakurai, M.; Shioiri, T.; Yajima, H. Chem Pharm Bull 1987, 35, 3447–3452.
62. Yajima, H.; Fujii, N.; Funakoshi, S.; Watanabe, T.; Murayama, E.; Otaka, A. Tetrahedron 1988, 44, 805–819.
63. Otaka, A.; Miyoshi, K.; Roller, P. P.; Burke, T. R., Jr.; Tamamura, H.; Fujii, N. J Chem Soc, Chem Commun 1995, 387–389.
64. Otaka, A.; Miyoshi, K.; Kaneko, M.; Tamamura, H.; Fujii, N.; Nomizu, M.; Burke, T. R., Jr.; Roller, P. P. J Org Chem 1995, 60, 3967–3974.
65. Tam, J. P.; Heath, W. F.; Merrifield, R. B. J Am Chem Soc 1986, 108, 5242–5251.
66. Tam, J. P.; Heath, W. F.; Merrifield, R. B. J Am Chem Soc 1983, 105, 6442–6455.
67. Tam, J. P.; Merrifield, R. B. In The Peptide, Analysis, Synthesis, Biology, Meienhofer, J.; Udenfriend, S., Eds.; Academic Press: New York, 1987; Vol 9, pp 185–248.
68. Tam, J. P. In Macromolecular Sequencing and Synthesis, Selected Methods and Applications; Schlesinger, D. H., Ed.; Alan R. Liss; New York, 1988; pp 153–184.
69. Takeuchi, Y.; Demachi, Y.; Yoshii, E. Tetrahedron Lett 1979, 1231–1232.
70. Gordeev, M. F.; Patel, D. V.; Barker, P. L.; Gordon, E. M. Tetrahedron Lett 1994, 35, 7585–7588.
71. Green, O. M. Tetrahedron Lett 1994, 35, 8081–8084.
72. Morgan, D. O. Nature 1995, 374, 131–134.
73. Fisher, R. P. Curr Opin Cell Biol Genet 1997, 7, 32–38.
74. Nakamura, E.; Mori, S. Angew Chem Int Ed 2000, 39, 3750–3771.
75. Taylor, R. J. K.; Ed. Organocopper Reagents, A Practical Approach; Oxford University Press: Oxford, 1994.
76. Yamazaki, T.; Shinohara, N.; Kitazume, T.; Sato, S. J. Fluorine Chem 1999, 97, 91–96.
77. Kazmierski, W. M., Ed. Peptidomimetics Protocols; Humana Press: Totowa, NJ, 1999.
78. Howard, J. A. K.; Hoy, V. J.; O'Hagan, D.; Smith, G. T. Tetrahedron 1996, 38, 12613–12622.
79. Plenio, H. Chem Rev 1997, 97, 3363–3384.
80. Hiyama, T.; Kanie, K.; Kusumoto, T.; Morizawa, Y.; Shimizu, M. In Organofluorine Compounds, Chemistry and Applications, Yamamoto, H., Ed.; Springer-Verlag: Berlin, 2000.
81. Okada, M.; Nakamura, Y.; Saito, A.; Sato, A.; Horikawa, H.; Taguchi, T. Chem Lett 2002, 28–29.
82. Nakamura, Y.; Okada, M.; Horikawa, H.; Taguchi, T. J Fluorine Chem 2002, 117, 143–148.
83. Okada, M.; Nakamura, Y.; Saito, A.; Sato, A.; Horikawa, H.; Taguchi, T. Tetrahedron Lett 2002, 43, 5845–5847.
84. Chounan, Y.; Ibuka, T.; Yamamoto, Y. J Chem Soc, Chem Commun 1994, 2003–2004.
85. Chounan, Y.; Horino, H.; Ibuka, T.; Yamamoto, Y. Bull Chem Soc Jpn 1997, 70, 1953–1959.
86. Molander, G. A. Chem Rev 1992, 92, 29–68.
87. Molander, G. A. In Organic Reactions; Paquette, L. A., et al., Eds.; John Wiley & Sons: New York, 1994; Vol 46, pp 211–367.
88. Molander, G. A.; Harris, C. R. Tetrahedron 1998, 54, 3321–3354.
89. Krief, A.; Laval, A.-M. Chem Rev 1999, 99, 745–777.
90. Steel, P. J Chem Soc, Perkin Trans 1 2001, 2727–2751.
91. Molander, G. A.; La Belle, B. E.; Hahn, G. J Org Chem 1986, 51, 5259–5264.
92. Otsubo, K.; Inanaga, J.; Yamaguchi, M. Tetrahedron Lett 1987, 28, 4437–4440.
93. Kang, S.-K.; Kim, S.-G.; Park, D.-C.; Lee, J.-S.; Yoo, W.-J.; Pak, C. S. J Chem Soc, Perkin Trans 1 1993, 9–10.
94. Kang, H. Y.; Cho, Y. S.; Koh, H. Y.; Chang, M. H. Synth Commun 1993, 23, 2977–2984.
95. Yoshida, A.; Takayama, H. Tetrahedron Lett 2001, 42, 3603–3606.
96. Otaka, A.; Watanabe, J.; Yukimasa, A.; Sasaki, Y.; Watanabe, H.; Kinoshita, T.; Oishi, S.; Tamamura, H.; Fujii, N. to be submitted.
97. Otaka, A.; Yukimasa, A.; Watanabe, J.; Sasaki, Y.; Oishi, S.; Tamamura, H.; Fujii, N. Chem Commun 2003, 1834–1835.

JOC*Article*

SmI_2-Mediated Reduction of γ,γ-Difluoro-α,β-enoates with Application to the Synthesis of Functionalized (*Z*)-Fluoroalkene-Type Dipeptide Isosteres

Akira Otaka,*[,‡] Junko Watanabe,[‡] Akira Yukimasa,[‡] Yoshikazu Sasaki,[‡] Hideaki Watanabe,[‡] Takayoshi Kinoshita,[§] Shinya Oishi,[‡] Hirokazu Tamamura,[‡] and Nobutaka Fujii[‡]

Graduate School of Pharmaceutical Sciences, Kyoto University, Sakyo-ku, Kyoto 606-8501, Japan, and Fujisawa Pharmaceutical Co., Ltd., Kashima, Yodogawa-ku, Osaka 532-8514, Japan

aotaka@pharm.kyoto-u.ac.jp

Received November 20, 2003

A samarium diiodide (SmI_2)-mediated reduction of γ,γ-difluoro-α,β-enoates (**15**, **29**, and **34**) was successfully applied to the synthesis of (*Z*)-fluoroalkene dipeptide isosteres (**23**, **30**, and **35**), which have served as potential dipeptide mimetics. Reduction of the γ,γ-difluoro-α,β-enoates by SmI_2 proceeded via successive two-electron transfers to form dienolate species which upon kinetically controlled trapping with *t*-BuOH yielded Xaa-Gly-type fluoroalkene isosteres exemplified by **23**, **30**, and **35**. Replacement of the *t*-BuOH kinetic trapping agent with aldehydes or ketones provided access to α-substituted fluoroalkene isosteres (**43** and **45**) through aldol reactions of Sm-dienolates with the carbonyl compounds. Of particular note, the use of the SmI_2–HCHO reagent system with chiral enoate **34** provided D-Phe-ψ[(*Z*)-CF=CH]-D/L-Ser isosteres (**45**), which could be converted to enantiomerically pure isosteres (**49**–**52**) that bore a variety of side chain functionalities at the α-position. This was achieved by a sequence of manipulations consisting of β-lactone formation followed by chromatographic separation and ring-opening with soft nucleophiles. Included in the present work is the first utilization of a Rh-catalyzed Reformatsky reaction of chiral imines for the stereoselective preparation of α,α-difluoro-β-amino acid derivatives (**28** and **33**). The appropriate choice of reagents (carbonyl compounds for kinetic trapping or ring-opening nucleophiles and imines for Reformatsky reactions) allows the presented methodology to yield various fluoroalkene isosteres possessing a wide range of side chain functionalities.

Introduction

(*Z*)-Fluoroalkene-type dipeptide isosteres **1** have served as potential dipeptide mimetics, where the peptide bond within a parent dipeptide is replaced by fluoroolefin units[1] (Figure 1). Both the fluoroalkene isosteres and their counterparts lacking fluorine substitution (disubstituted (*E*)-alkene isosteres **2**[2]) are stable against proteases, and resemble the parent peptide bonds[3] in their three-dimensional structure. On the other hand, when viewed from their electrostatic nature, the fluoroalkene isosteres **1** are more similar to peptide bonds than **2** due to the presence of the highly negative fluorine atom.[1a,b,4] Additionally, when incorporated into peptides, a trisubstituted alkene dipeptide isostere that possesses a substituent on the γ-position provides effects on the overall structure of peptides similar to that seen in native peptide bonds due to restriction of ϕ,ψ-dihedral angles.[5] This restriction in native peptides is known to be attributable to interactions between the carbonyl oxygen and other atoms, especially the side chain β-carbon.[6]

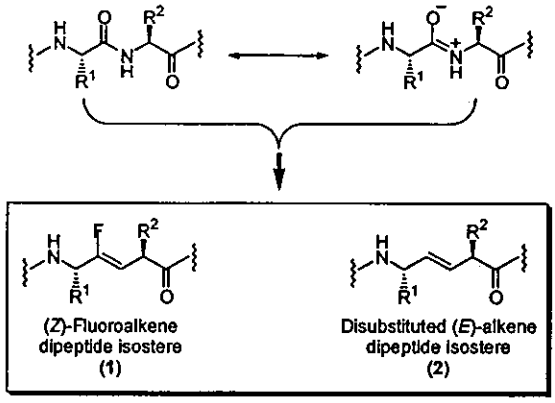


FIGURE 1. Native peptide bond and corresponding alkene isosteres.

* Address correspondence to this author. Phone: 81-75-753-4571. Fax: 81-75-753-4570.
‡ Kyoto University.
§ Fujisawa Pharmaceutical Co., Ltd.

(1) For fluoroalkene dipeptide isosteres, see: (a) Abraham, R. J.; Ellison, S. L. R.; Schonholzer, P.; Thomas, W. A. *Tetrahedron* **1986**, *42*, 2101–2110. (b) Allmendinger, T.; Furet, P.; Hungerbühler, E. *Tetrahedron Lett.* **1990**, *31*, 7297–7300. (c) Allmendinger, T.; Felder, E.; Hungerbühler, E. *Tetrahedron Lett.* **1990**, *31*, 7301–7304. (d) Boros, L. G.; De Corte, B.; Gimi, R. H.; Welch, J. T.; Wu, Y.; Handschumacher, R. E. *Tetrahedron Lett.* **1994**, *35*, 6033–6036. (e) Bartlett, P. A.; Otake, A. *J. Org. Chem.* **1995**, *60*, 3107–3111. (f) Welch, J. T.; Lin, J. *Tetrahedron* **1996**, *52*, 291–304. (g) Welch, J. T.; Allmendinger, T. In *Peptidomimetics Protocols*; Kazmierski, W. M., Ed.; Humana Press: Totowa, NJ, 1999; pp 357–384.

10.1021/jo035709d CCC: $27.50 © 2004 American Chemical Society
Published on Web 02/06/2004

Because fluorine and oxygen are quite similar in their van der Waals radii (F, 1.35 Å; O, 1.40 Å) as well as being isoelectronic with each other ($2S^22P^6$),[4d] the similarity in restriction-effects mentioned above is to be expected. These qualities of fluoroalkene dipeptide isosteres have contributed to the fact that a substance P analogue containing a Phe-ψ[(Z)-CF=CH]-Gly unit has been shown to exhibit potency comparable to that of the natural ligand whereas the disubstituted (*E*)-alkene counterpart was significantly less potent.[1c]

Syntheses of this class of compounds have been achieved by the use of aldol reactions of α-fluoro-α,β-unsaturated aldehydes with ester enolates, followed by introduction of nitrogen functionality, or by fluoroolefination reactions of aldehydes or ketones with α-fluoroacetate derivatives.[1b-g] These synthetic methods are based on a strategy wherein the construction of fluoroalkene units is followed by derivatization of functional groups. This requires relatively long reaction sequences. During the course of our synthetic studies on nonhydrolyzable phosphoamino acids,[7] we found that γ-phosphono-γ,γ-difluoro-α,β-enoate **3** was reduced to the corresponding γ-phosphono-γ-fluoro-β,γ-enoate **4** with organocopper reagents.[8] This organocopper-mediated reduction[9] was then applied to the synthesis of (*Z*)-fluoroalkene dipeptide isosteres **6** (Figure 2).[10,11] Mechanistic investigation of the organocopper-mediated reduction of the highly electrophilic δ-amino-γ,γ-difluoro-α,β-enoates **5** led us to envision that single electron transfer (SET) from an organocopper to the substrate is responsible for the reduction.[12,13]

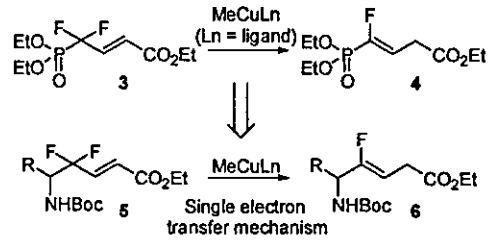


FIGURE 2. Organocopper-mediated reduction of γ-phosphono-γ,γ-difluoro-α,β-enoate.

Samarium diiodide (SmI_2)[14] has been well-recognized as a powerful one-electron reducing agent capable of meeting the diverse demands of synthetic organic chemistry.[15] Taking account of our mechanistic insight into the organocopper-mediated reduction, we speculated that SmI_2 would also be applicable to the reduction of γ,γ-difluoro-α,β-enoates to give γ-fluoro-β,γ-enoates, which could open the avenue to the synthesis of fluoroalkene dipeptide isosteres with δ-amino-γ,γ-difluoro-α,β-enoates as key intermediates (Figure 2, **5** to **6**). We present herein an examination of the feasibility of the SmI_2-mediated reduction of γ,γ-difluoro-α,β-enoates and its application to the syntheses of fluoroalkene dipeptide isosteres, including the synthesis of chiral α-functionalized (*Z*)-fluoroalkene dipeptide isosteres.

Results and Discussion

Prior to examination of the synthetic applicability of SmI_2 to preparation of the fluoroalkene isosteres, δ-siloxy-γ,γ-difluoro-α,β-enoates such as **10** were subjected to SmI_2- or organocopper-mediated reduction for comparison of these reagent systems. Requisite substrates

(2) (*E*)-Alkene isostere: (a) Ibuka, T.; Habashita, H.; Otaka, A.; Fujii, N.; Oguchi, Y.; Uyehara, T.; Yamamoto, Y. *J. Org. Chem.* **1991**, *56*, 4370−4382. (b) Fujii, N.; Nakai, K.; Habashita, H.; Yoshizawa, H.; Ibuka, T.; Garrido, F.; Mann, A.; Chounan, Y.; Yamamoto, Y. *Tetrahedron Lett.* **1993**, *34*, 4227−4230. (c) Daly, M. J.; Ward, R. A.; Thompson, D. A.; Procter, G. *Tetrahedron Lett.* **1995**, *36*, 7545−7548. (d) Kessler, H.; Kranz, M. *Tetrahedron Lett.* **1996**, *37*, 5359−5362. (e) Ibuka, T.; Nakai, K.; Habashita, H.; Hotta, Y.; Fujii, N.; Mimura, N.; Miwa, Y.; Taga, T.; Yamamoto, Y. *Angew. Chem., Int. Ed. Engl.* **1994**, *33*, 652−654. (f) Wipf, P.; Fritch, P. C. *J. Org. Chem.* **1994**, *59*, 4875−4886. (g) Fujii, N.; Nakai, K.; Tamamura, H.; Otaka, A.; Mimura, N.; Miwa, Y.; Taga, T.; Yamamoto, Y.; Ibuka, T. *J. Chem. Soc., Perkin Trans. 1* **1995**, 1359−1371. (h) Wipf, P.; Henninger, T. C. *J. Org. Chem.* **1997**, *62*, 1586−1587. (i) Tamamura, H.; Yamashita, M.; Muramatsu, H.; Ohno, H.; Ibuka, T.; Otaka, A.; Fujii, N. *Chem. Commun.* **1997**, 2327. (j) Tamamura, H.; Yamashita, M.; Nakajima, Y.; Sakano, K.; Otaka, A.; Ohno, H.; Ibuka, T.; Fujii, N. *J. Chem. Soc., Perkin Trans. 1* **1999**, 2983−2996. (k) Oishi, S.; Tamamura, H.; Yamashita, M.; Hamanaka, N.; Otaka, A.; Fujii, N. *J. Chem. Soc., Perkin Trans. 1* **2001**, 2445−2451. (l) Henninger, T. C.; Wipf, P. In ref 1g, pp 125−136.

(3) For some arguments about planarity of peptide bonds, see: (a) MacArthur, M. W.; Thornton, J. M. *J. Mol. Biol.* **1996**, *264*, 1180−1195. (b) Edison, A. S. *Nature Struct. Biol.* **2001**, *8*, 201−202.

(4) (a) Howard, J. A. K.; Hoy, V. J.; O'Hagan, D.; Smith, G. T. *Tetrahedron* **1996**, *38*, 12613−12622. (b) O'Hagan, D.; Rzepa, H. S. *Chem. Commun.* **1997**, 645−652. (c) Plenio, H. *Chem. Rev.* **1997**, *97*, 3363−3384. (d) Hiyama, T.; Kanie, K.; Kusumoto, T.; Morizawa, Y.; Shimizu, M. In *Organofluorine Compounds. Chemistry and Applications*; Yamamoto, H., Ed.; Springer-Verlag: Berlin, Germany, 2000.

(5) For some recent examples including tetrasubstituted isosteres, see: (a) Gardner, R. R.; Liang, G.-B.; Gellman, S. H. *J. Am. Chem. Soc.* **1995**, *117*, 3280−3281. (b) Schopfer, U.; Stahl, M.; Brandl, T.; Hoffmann, R. W. *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 1745−1747. (c) Wipf, P.; Henninger, T. C.; Geib, S. T. *J. Org. Chem.* **1998**, *63*, 6088−6089. (d) Oishi, S.; Niida, A.; Kamano, T.; Miwa, Y.; Taga, T.; Odagaki, Y.; Hamanaka, N.; Yamamoto, M.; Ajito, K.; Tamamura, H.; Otaka, A.; Fujii, N. *J. Chem. Soc., Perkin Trans. 1* **2002**, 1786−1793. (e) Otaka, A.; Katagiri, F.; Kinoshita, T.; Odagaki, Y.; Oishi, S.; Tamamura, H.; Hamanaka, N.; Fujii, N. *J. Org. Chem.* **2002**, *67*, 6152−6161. (f) Oishi, S.; Kamano, T.; Niida, A.; Odagaki, Y.; Hamanaka, N.; Yamamoto, M.; Ajito, K.; Tamamura, H.; Otaka, A.; Fujii, N. *J. Org. Chem.* **2002**, *67*, 6162−6173. (g) Wang, X. J.; Hart, S. A.; Xu, B.; Mason, M. D.; Goodell, J. R.; Etzkorn, F. A. *J. Org. Chem.* **2003**, *68*, 2343−2349.

(6) Head-Gordon, T.; Head-Gordon, M.; Frisch, M. J.; Brooks, C. L., III; Pople, J. A. *J. Am. Chem. Soc.* **1991**, *113*, 5989−5997.

(7) Otaka, A.; Mitsuyama, E.; Kinoshita, T.; Tamamura, H.; Fujii, N. *J. Org. Chem.* **2000**, *65*, 4888−4899.

(8) (a) Otaka, A.; Mitsuyama, E.; Watanabe, H.; Tamamura, H.; Fujii, N. *Chem. Commun.* **2000**, 1081−1082. Kitazume's group investigated organocopper-mediated reduction of a γ,γ,γ-trifluoro-α,β-enoate, see: (b) Yamazaki, T.; Shinohara, N.; Kitazume, T.; Sato, S. *J. Fluorine Chem.* **1999**, *97*, 91−96.

(9) For copper-mediated reduction of γ-substituted-α,β-enoates, see: (a) Ibuka, T.; Aoyagi, T.; Yamamoto, Y. *Chem. Pharm. Bull.* **1986**, *34*, 2417−2427. (b) Fujii, N.; Habashita, H.; Shigemori, N.; Otaka, A.; Ibuka, T.; Tanaka, M.; Yamamoto, Y. *Tetrahedron Lett.* **1991**, *32*, 4969−4972. (c) Ibuka, T.; Yoshizawa, H.; Habashita, H.; Fujii, N.; Chounan, Y.; Tanaka, M.; Yamamoto, Y. *Tetrahedron Lett.* **1992**, *33*, 3783−3786.

(10) Otaka, A.; Watanabe, H.; Mitsuyama, E.; Yukimasa, A.; Tamamura, H.; Fujii, N. *Tetrahedron Lett.* **2001**, *42*, 285−287.

(11) Treatment of γ,γ-difluoro-α,β-enoates with R_3Al−Cu(I) reagent proceeds in an S_N2' manner to afford α-alkylated γ-fluoro-β,γ-enoates and related work, see: (a) Okada, M.; Nakamura, Y.; Saito, A.; Sato, A.; Horikawa, H.; Taguchi, T. *Chem. Lett.* **2002**, 28−29. (b) Nakamura, Y.; Okada, M.; Horikawa, H.; Taguchi, T. *J. Fluorine Chem.* **2002**, *117*, 143−148. (c) Okada, M.; Nakamura, Y.; Saito, A.; Sato, A.; Horikawa, H.; Taguchi, T. *Tetrahedron Lett.* **2002**, *43*, 5845−5847.

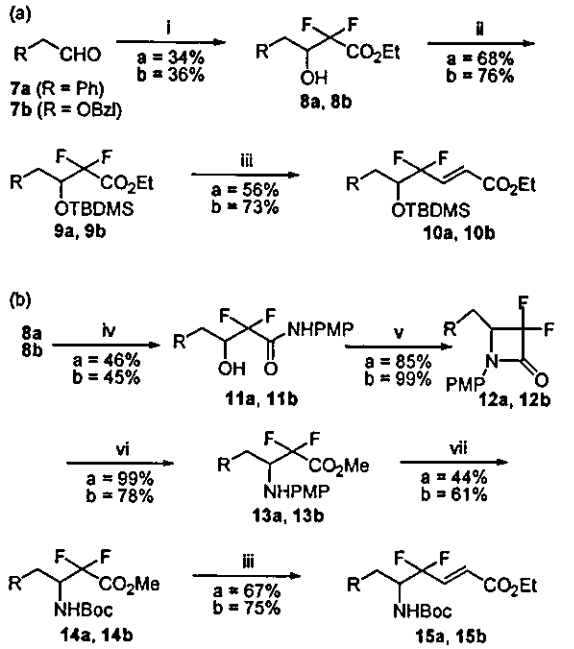
(12) Otaka, A.; Watanabe, H.; Yukimasa, A.; Oishi, S.; Tamamura, H.; Nobutaka, F. *Tetrahedron Lett.* **2001**, *42*, 5443−5446.

(13) (a) Chounan, Y.; Ibuka, T.; Yamamoto, Y. *J. Chem. Soc., Chem. Commun.* **1994**, 2003−2004. (b) Chounan, Y.; Horino, H.; Ibuka, T.; Yamamoto, Y. *Bull. Chem. Soc. Jpn.* **1997**, *70*, 1953−1959.

(14) Girard, P.; Namy, J. K.; Kagan, H. B. *J. Am. Chem. Soc.* **1980**, *102*, 2693−2698.

(15) For some recent reviews, see: (a) Molander, G. A. *Chem. Rev.* **1992**, *92*, 29−68. (b) Molander, G. A. In *Organic Reactions*; Paquette, L. A., et al., Eds.; John Wiley & Sons: New York, 1994; Vol. 46, pp 211−367. (c) Molander, G. A.; Harris, C. R. *Tetrahedron* **1998**, *54*, 3321−3354. (d) Krief, A.; Laval, A.-M. *Chem. Rev.* **1999**, *99*, 745−777. (e) Steel, P. *J. Chem. Soc., Perkin Trans. 1* **2001**, 2727−2751.

SCHEME 1[a]



[a] Reagents and conditions: (i) $BrZnCF_2CO_2Et$, THF; (ii) TBDMSOTf, 2,6-lutidine, CH_2Cl_2; (iii) DIBAL-H, CH_2Cl_2–toluene, then $(EtO)_2P(O)CH_2CO_2Et$, LiCl, $(i\text{-}Pr)_2NEt$, MeCN; (iv) NaOH, THF–H_2O, then BOP-Cl, *p*-anisidine, $(i\text{-}Pr)_2NEt$, CH_2Cl_2; (v) Ph_3P, DEAD, THF; (vi) NaOH, THF–H_2O, then H_2SO_4, MeOH; (vii) CAN, MeCN–H_2O, then $(Boc)_2O$, THF.

were synthesized as follows (Scheme 1a): (1) Reformatsky reaction of aldehydes **7** with $BrZnCF_2CO_2Et$, followed by TBDMS protection of the resulting hydroxyl group, and (2) DIBAL-H reduction of the esters **9** to the corresponding aldehydes, followed by carbon chain elongation with Horner–Emmons olefination. Reaction of **10a** or **10b** with a cyano Gilman reagent ($Me_2CuLi{\cdot}LiCN$)[16,17] afforded the corresponding (*Z*)-δ-siloxy-γ-fluoro-β,γ-enoates[18] **16a** or **16b** with an accompanying significant amount of fluorodiene **17a** or **17b**, respectively (Table 1, entries 1 and 2). This undesirable fluorodiene formation may be attributable to the presence of hydrogen atoms that are easily abstracted by the organocopper reagent at the position adjacent to the δ-siloxy group. Initial attempts to reduce **10b** with SmI_2 in THF gave the desired (*Z*)-fluoroalkene in low yield (17%) with accompanying unidentified products (Table 1, entry 3). The use of SmI_2 in the presence of protic solvent has been well-documented to suppress the formation of side products resulting from unproductive pathways. Reports have indicated that γ,δ-epoxy-α,β-enoates undergo reductive epoxide ring opening with SmI_2 in THF in the presence of a proton source to give δ-hydroxy-β,γ-enoates.[19] Addition of EtOH as a proton source in the reduction of the γ,γ-difluoro-α,β-enoates **10** proved to improve the chemical yields (67% for **10a** and 78% for **10b**); nonetheless, a nonnegligible amount of 1,4-reduction products **18** was formed.[20]

In analogy to the proposed reaction mechanism for γ,δ-epoxy-α,β-enoates,[19a,b,d] a pathway via dienolates[21] **21** resulting from successive two-electron transfers is likely to be involved in the reduction of the γ,γ-difluoro-α,β-enoates **10** with SmI_2 in THF in the presence of a proton source (Figure 3). This proposed mechanism is probably adequate in light of the experimental observation that transfer of two electrons into the π-electron system adjacent to a trifluoromethyl group induces the pushing out of one of the fluorine atoms of the CF_3 group.[22] Formation of the 1,4-reduction products **18** would seem to be attributable to protonation at the β-position of the possible intermediate **20** to yield enolates **22**. On the basis of such a mechanism for 1,4-reductive product formation, we speculated that the use of more sterically crowded alcohols with lower acidity (e.g. *t*-BuOH) as proton sources could prevent the protonation at the β-carbon to give the dienolates **21**, which could be converted to the desired reductive product **16** by protonation at the α-carbon. Reaction of **10a** or **10b** with SmI_2 in THF in the presence of *t*-BuOH as the proton source (THF:*t*-BuOH = 7.3 or 7.7:0.5, v/v) at 0 °C for 60 min proceeded quantitatively to yield the desired γ-fluoro-β,γ-enoates **16a** or **16b** in 92% or 90% isolated yield, respectively, without any detectable formation of 1,4-reductive product or fluorodiene (Table 1, entries 6 and 7).[23] Addition of a stoichiometrical amount of *t*-BuOH was sufficient for the clean SmI_2-mediated conversion of **10** to **16**. Having ascertained the reductive conditions for use of SmI_2 with γ,γ-difluoro-α,β-enoates, we next applied the SmI_2-mediated reductive protocol to the synthesis of

(16) For recent reviews, see: (a) Nakamura, E.; Mori, S. *Angew. Chem., Int. Ed.* **2000**, *39*, 3750–3771. (b) Taylor, R. J. K., Ed. In *Organocopper Reagents, A Practical Approach*; Oxford University Press: Oxford, UK, 1994.

(17) For recent discussion about the structures, see: Kronenburg, C. M. P.; Jastrzebski, J. T. B. H.; Spek, A. L.; van Koten, G. *J. Am. Chem. Soc.* **1998**, *120*, 9688–9689 and references cited herein.

(18) Fluoroalkene compounds obtained in this study have coupling constants ($^3J_{HF}$ = 35.3–37.0 Hz). Those values are well consistent with those of compounds possessing (*Z*)-fluoroalkene units. See: Waschüsch, R.; Carran, J.; Savignac, P. *Tetrahedron* **1996**, *52*, 14199–14216.

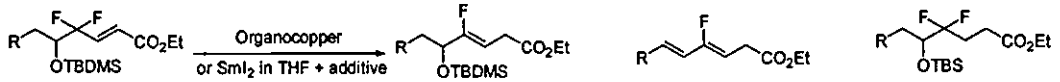
(19) For reductive C–O cleavage at the γ-position to α,β-unsaturated carbonyls (a–e) or the α-position to carbonyls (f–k) by SmI_2 and reductive C–N cleavage by SmI_2 (*l*), see: (a) Molander, G. A.; La Belle, B. E.; Hahn, G. *J. Org. Chem.* **1986**, *51*, 5259–5264. (b) Otsubo, K.; Inanaga, J.; Yamaguchi, M. *Tetrahedron Lett.* **1987**, *28*, 4437–4440. (c) Kang, S.-K.; Kim, S.-G.; Park, D.-C.; Lee, J.-S.; Yoo, W.-J.; Pak, C. S. *J. Chem. Soc., Perkin Trans. 1* **1993**, 9–10. (d) Kang, H. Y.; Cho, Y. S.; Koh, H. Y.; Chang, M. H. *Synth. Commun.* **1993**, *23*, 2977–2984. (e) Yoshida, A.; Takayama, H. *Tetrahedron Lett.* **2001**, *42*, 3603–3606. (f) Mikami, K.; Yoshida, A. *Angew. Chem., Int. Ed. Engl.* **1997**, *109*, 892–894. (g) Nakamura, Y.; Takeuchi, S.; Ohgo, Y.; Yamaoka, M.; Yoshida, A.; Mikami, K. *Tetrahedron Lett.* **1997**, *38*, 2709–2712. (h) Yoshida, A.; Mikami, K. *Synlett* **1997**, 1375–1376. (i) Yoshida, A.; Hanamoto, T.; Inanaga, J.; Mikami, K. *Tetrahedron Lett.* **1998**, *39*, 1777–1780. (j) Mikami, K.; Yamaoka, M.; Yoshida, A.; Nakamura, Y.; Takeuchi, S.; Ohgo, Y. *Synlett* **1998**, 607–608. (k) Nakamura, Y.; Takeuchi, S.; Ohgo, Y.; Yamaoka, M.; Yoshida, A.; Mikami, K. *Tetrahedron* **1999**, *55*, 4595–4620. (l) Molander, G. A.; Stengel, P. J. *Tetrahedron* **1997**, *53*, 8887–8912.

(20) (a) Inanaga, J.; Sakai, S.; Handa, Y.; Yamaguchi, M.; Yokoyama, Y. *Chem. Lett.* **1991**, 2117–2118. (b) Cabrera, A.; Alper, H. *Tetrahedron Lett.* **1992**, *33*, 5007–5008. (c) Yanada, R.; Bessho, K.; Yanada, K. *Synlett* **1995**, 443–444. (d) Fujita, Y.; Fukuzumi, S.; Otera, J. *Tetrahedron Lett.* **1997**, *38*, 2121–2124.

(21) (a) Yang, S.-M.; Fang, J.-M. *Tetrahedron Lett.* **1997**, *38*, 1589–1592. (b) Reutrakul, V.; Saeeng, R.; Pohmakotr, M.; Kongsaeree, P. *Tetrahedron Lett.* **1999**, *40*, 1019–1020. (c) Concellón, J. M.; Bernad, P. L.; Rodríguez-Solla, H. *Angew. Chem., Int. Ed.* **2001**, *40*, 3897–3899.

(22) For reductive C–F bond cleavage with Mg (a, b) or transition-metal complexes (c), see: (a) Uneyama, K.; Mizutani, G.; Maeda, K.; Kato, T. *J. Org. Chem.* **1999**, *64*, 6717–6723. (b) Mae, M.; Amii, H.; Uneyama, K. *Tetrahedron Lett.* **2000**, *41*, 7893–7896. (c) Kraft, B. M.; Jones, W. D. *J. Am. Chem. Soc.* **2002**, *124*, 8681–8689.

TABLE 1. Reduction of δ-Siloxy-γ,γ-difluoro-α,β-enoates with Organocopper or SmI_2



10a (R = Ph) **10b** (R = OBzl) **16a** (R = Ph) **16b** (R = OBzl) **17a** (R = Ph) **17b** (R = OBzl) **18a** (R = Ph) **18b** (R = OBzl)

entry	substrate	reagent (equiv, solvent, additive)	condition	products (isolated yield %)
1	**10a**	Me_2CuLi·LiCN[a] (4 equiv, THF:Et_2O = 4:1)	−78 °C, 10 min	**16a** (59), **17a** (10)[b]
2	**10b**	Me_2CuLi·LiCN[a] (4 equiv, THF:Et_2O = 4:1)	−78 °C, 10 min	**16b** (36), **17b** (31)
3	**10b**	SmI_2 (6 equiv, THF)	0 °C, 60 min	**16b** (17)[c]
4	**10a**	SmI_2 (6 equiv, THF:EtOH = 7.7:0.5)	0 °C, 60 min	**16a** (67), **18a** (25)[b]
5	**10b**	SmI_2 (6 equiv, THF:EtOH = 7.3:0.5)	0 °C, 60 min	**16b** (78), **18b** (15)[b]
6	**10a**	SmI_2 (6 equiv, THF:*t*-BuOH = 7.7:0.5)	0 °C, 60 min	**16a** (92)
7	**10b**	SmI_2 (6 equiv, THF:*t*-BuOH = 7.3:0.5)	0 °C, 60 min	**16b** (90)
8	**10a**	SmI_2 (6 equiv, THF + 1.5 equiv. *t*-BuOH)	0 °C, 60 min	**16a** (94)
9	**10b**	SmI_2 (6 equiv, THF + 1.5 equiv. *t*-BuOH)	0 °C, 60 min	**16b** (86)

[a] In the presence of Li salts (LiCl and LiBr). [b] Obtained as mixtures. The ratios were determined by ^1H NMR. [c] No starting material. Several unidentified compounds were formed.

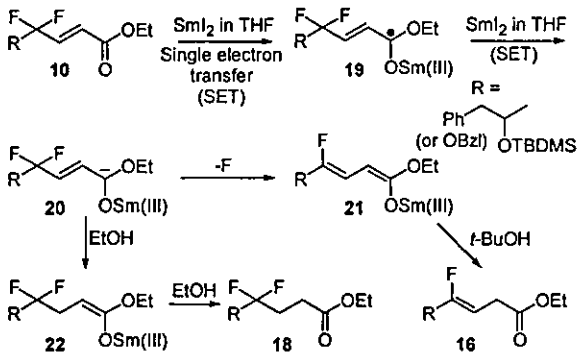


FIGURE 3. Possible mechanism of the reaction of γ,γ-difluoro-α,β-enoates with SmI_2.

fluoroalkene dipeptide isosteres in comparison with the organocopper-mediated method.

Requisite substrates **15a** and **15b** were synthesized in a nonstereoselective manner as follows (Scheme 1b): (1) hydrolysis of **8a** or **8b** and subsequent amide formation with *p*-anisidine with the aid of bis(2-oxo-3-oxazolidinyl)-phosphinic chloride (BOP-Cl);[24] (2) conversion of the hydroxyl group to amine functionality utilizing a sequence of reactions consisting of intramolecular Mitsunobu reaction,[25] hydrolysis of the resulting β-lactams **12**, and esterification of the carboxylic acids; (3) deprotection of the PMP (*p*-methoxyphenyl) group of **13** followed by Boc-reprotection; and (4) carbon chain elongation by DIBAL-H reduction of the Boc-protected β-amino acid esters **14** and subsequent Horner−Emmons olefination with triethyl phosphonoacetate. Reduction of **15a** or **15b** with SmI_2 or organocopper for the preparation of the fluoroalkene isosteres is summarized in Table 2. Both the reaction with the cyano Gilman reagent (Me_2CuLi·LiCN at −78 °C for 10 min) and that with SmI_2-*t*-BuOH (7.7 or 7.3:0.5, v/v, 0 °C for 60 min) proceeded quantitatively to yield the desired (*Z*)-fluoroalkene isosteres in high chemical yields (slightly higher yields in the case of the SmI_2-mediated method). The resulting fluoroalkene isosteres were obtained as racemates due to the nonstereoselective synthesis of precursor γ,γ-difluoro-α,β-enoates **15**. On the basis of these preliminary experiments, synthesis of chiral fluoroalkene isosteres was envisioned via stereoselective preparation of the α,α-difluoro-β-amino acids as outlined in the following.

Recent growing interest in both the medicinal and the synthetic organic chemistry of fluororinated β-amino acids[26] has led to the development of methodologies for their asymmetric synthesis.[27,28] Published methods utilizing the addition of a Reformatsky-type reagent ($BrZnCF_2CO_2Et$) to chiral imines[27] prompted us to use a similar type of reaction for the construction of our chiral units. Recently, Honda et al. reported a one-pot preparation of chiral β-amino esters by a rhodium-catalyzed three-component coupling reaction, where ethyl bromoacetate is added to chiral aldimines with complete diastereoselectivity under mild reaction conditions (0 °C) in the presence of Wilkinson's catalyst and diethylzinc.[29,30] The chiral aldimines were prepared from aldehydes and an *O*-protected phenylglycinol derivative in situ in the presence of molecular sieves. Honda's protocol was of note because simple operations with mild reaction conditions give β-amino acid esters in contrast to the fact that

(23) Proton sources have been reported to exert an influence on the regiochemical and stereochemical outcome of SmI_2-mediated reduction. see: (a) Yoshida, A.; Mikami, K. *Synlett.* **1997**, 1375−1376. (b) Yoshida, A.; Hanamoto, T.; Inanaga, J.; Mikami, K. *Tetrahedron Lett.* **1998**, *39*, 1777−1780. In addition to the steric hindrance of *t*-BuOH, the low acidic nature of *t*-BuOH as compared with that of EtOH would also contribute to slow protonation at the β-carbon of **20**, which results in the preferential conversion of **20** to **16** via the dienolate **21**.

(24) Diago-Meseguer, J.; Palomo-Coll, A. L.; Fernandez-Lizarbe, J. R.; Zugaza-Bilbao, A. *Synthesis* **1980**, 547−551.

(25) Nakayama, K.; Kawato, H. C.; Inagaki, H.; Nakajima, R.; Kitamura, A.; Someya, K.; Ohta, T. *Org. Lett.* **2000**, 2, 977−980.

(26) For reviews on fluorinated amino acids, see: (a) Welch, J. T. *Tetrahedron* **1987**, *43*, 3123−3197. (b) Kukhar', V. P.; Soloshonok, V. A., Eds. *Fluorine-containing Amino Acids*; John Wiley & Sons: New York, 1994.

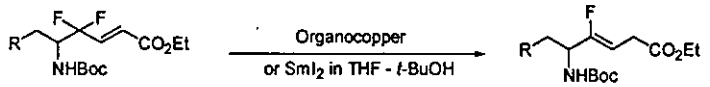
(27) (a) Marcotte, S.; Pannecoucke, X.; Feasson, C.; Quirion, J.-C. *J. Org. Chem.* **1999**, *64*, 8461−8464. (b) Staas, D. D.; Savage, K. L.; Hormnick, C. F.; Tsou, N. N.; Ball, R. G. *J. Org. Chem.* **2002**, *67*, 8276−8279.

(28) For reports on use of difluoroketenesilylacetal, see: (a) Iseki, K.; Kuroki, Y.; Asada, D.; Takahashi, M.; Kishimoto, S.; Kobayashi, Y. *Tetrahedron* **1997**, *53*, 10271−10280. (b) Iseki, K. *Tetrahedron* **1998**, *54*, 13887−13914.

(29) Honda, T.; Wakabayashi, H.; Kanai, K. *Chem. Pharm. Bull.* **2002**, *50*, 307−308.

(30) Adrian, J. C., Jr.; Snapper, M. L. *J. Org. Chem.* **2003**, *68*, 2143−2150.

TABLE 2. Reduction of δ-Amino-γ,γ-difluoro-α,β-enoates with Organocopper or SmI_2



15a (R = Ph)
15b (R = OBzl)

23a (R = Ph)
23b (R = OBzl)

entry	substrate	reagent (equiv, solvent)	condition	product (isolated yield %)
1	**15a**	Me_2CuLi·LiCN[a] (4 equiv, THF:Et_2O = 4:1)	−78 °C, 10 min	**23a** (84)
2	**15b**	Me_2CuLi·LiCN[a] (4 equiv, THF:Et_2O = 4:1)	−78 °C, 10 min	**23b** (89)
3	**15a**	SmI_2 (6 equiv, THF:*t*-BuOH = 7.7:0.5)	0 °C, 60 min	**23a** (92)
4	**15b**	SmI_2 (6 equiv, THF:*t*-BuOH = 7.3:0.5)	0 °C, 60 min	**23b** (94)

[a] In the presence of Li salts (LiCl and LiBr).

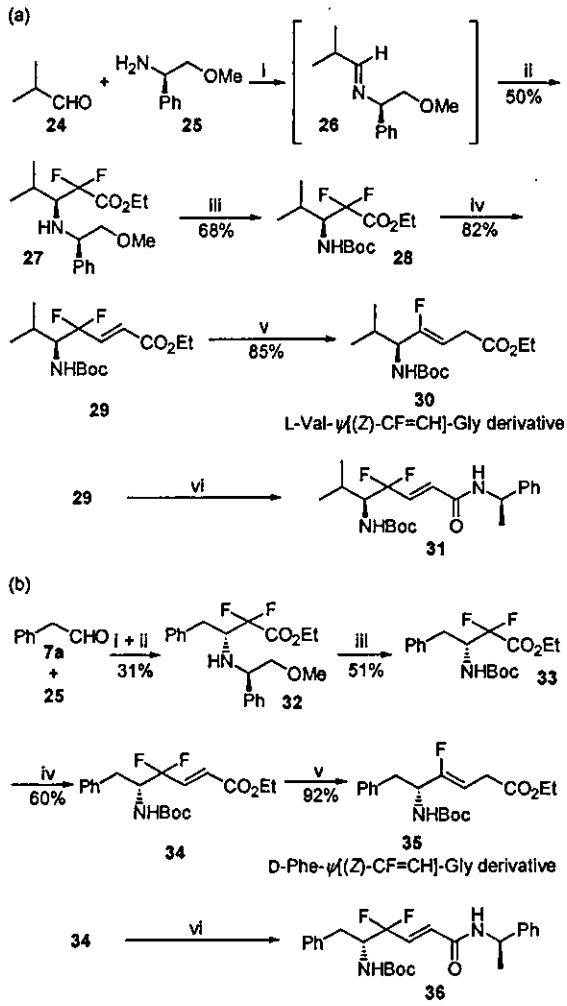
addition of Reformatsky reagents to imines usually yields β-lactam derivatives.[27a] Therefore, we attempted to apply this method to the asymmetric synthesis of requisite α,α-difluoro-β-amino acid derivatives such as **14** by replacement of ethyl bromoacetate with ethyl bromodifluoroacetate. Initially the stereoselective synthesis of a L-Val-Gly-type fluoroalkene dipeptide isostere **30** was envisioned (Scheme 2-(a)).

Successive addition of Wilkinson's catalyst, ethyl bromodifluoroacetate, and diethylzinc to the chiral aldimine **26** prepared from isobutyraldehyde **24** and the methyl ether of (*R*)-phenyl glycinol[31] **25** with the aid of activated molecular sieves resulted in the diastereoselective formation of the (*S*)-α,α-difluoro-β-amino ester **27** in moderate yield.[32] The chiral auxiliary was then removed by hydrogenolysis with $Pd(OH)_2/C$-H_2 in EtOH. The resulting amine compound was Boc protected with $(Boc)_2O$ in THF under refluxing conditions to yield derivative **28**, which was converted to γ,γ-difluoro-α,β-enoate **29** as a substrate for the SmI_2-mediated reduction in a manner identical with that employed for the synthesis of **15**. Establishment of chirality as being of the (*S*)-configuration was achieved as follows: (1) hydrolysis of ethyl ester **29**; (2) condensation with (*R*)-methylbenzylamine; and (3) X-ray analysis of crystallized sample **31**. Reaction of **29** with SmI_2 in THF in the presence of *t*-BuOH proceeded quantitatively to afford L-Val-Gly-type fluoroalkene isostere **30** in 85% isolated yield.

Transition state model **37**[29,33] can be used to rationalize the reaction outcome of the addition of the Reformatsky-type reagent to the chiral aldimine **26** (Figure 4). According to this model, the zinc enolate attacks from the less-hindered *Re* face of the imine to furnish the (*S*)-α,α-difluoro-β-amino ester **27**.

Next, the synthesis of the chiral Phe-Gly-type isostere **35** was undertaken, which required the condensation of phenylacetaldehyde **7a** and the chiral amine **25** (Scheme 2b). The analogous *p*-toluenesulfinyl imine has been reported to favor the corresponding tautomeric form (enamine) rather than the aldimine.[34,35] In addition, competitive α-deprotonation can be problematic during addition of organometallic reagents to imines prepared from phenylacetaldehyde. A one-pot reaction consisting

SCHEME 2[a]



[a] Reagents and conditions: (i) molecular sieves 3Å, THF; (ii) $BrCF_2CO_2Et$, Et_2Zn, $RhCl(PPh_3)_3$, THF−hexane; (iii) $Pd(OH)_2$, H_2, EtOH, then $(Boc)_2O$, THF; (iv) DIBAL-H, CH_2Cl_2−toluene, then $(EtO)_2P(O)CH_2CO_2Et$, LiCl, $(i\text{-}Pr)_2NEt$, MeCN; (v) SmI_2, *t*-BuOH, THF; (vi) 1 M LiOH (aq), THF, then (*R*)-methylbenzylamine, 1-hydroxybenzotriazole, 1-ethyl-3-(3′-dimethylaminopropyl)carbodiimide, $(i\text{-}Pr)_2NEt$, THF.

(31) Smith, A. B., III; Yager, K. M.; Phillips, B. W.; Taylor, C. M. *Org. Synth.* **1998**, *75*, 19−30.

(32) In our experiments, flash chromatographical purification of crude **27** (or **32**) did not afford another diastereomer even though the possibility of formation of a diastereomer cannot be completely ruled out.

(33) Mokhallalati, M. K.; Wu, M.-J.; Pridgen, L. N. *Tetrahedron Lett.* **1993**, *34*, 47−50.

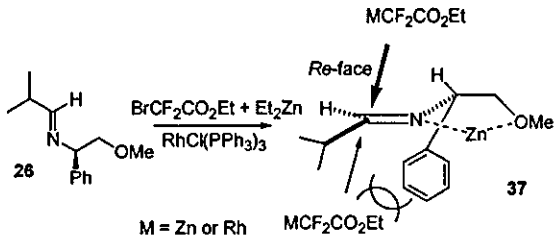


FIGURE 4. Proposed mechanism of addition to chiral imine **26**.

of a chiral aldimine (prepared from **25** and **7a**) ethyl bromodifluoroacetate, Wilkinson's catalyst, and diethylzinc in THF at 0 °C for 30 min proceeded with high diastereoselectivity to give diastereomerically pure α,α-difluoro-β-amino acid derivative **32** in 31% isolated yield.[32] Compound **32** was converted to the requisite substrate **34** for the SmI_2-mediated reduction in a manner identical with that used for transformation of **27** to **29**. To determine the absolute configuration at the δ-carbon of **34**, compound **34** was converted to the crystalline amide **36** by a similar sequence of reactions used for the preparation of **31**. X-ray crystallographic analysis of **36** showed that the absolute configuration is *R*. This stereo preference is opposite to what was observed when isobutyraldehyde was used. Although the origin of reversal of the stereoselectivity was not thoroughly examined, the following factors could explain this phenomenon (Figure 5).

As mentioned above, imines prepared from phenylacetaldehyde have a tendency to exist in tautomeric equilibrium between imines **38** and enamines **39** or **40** due to the presence of the phenylacetaldehyde aromatic nucleus. The imine−enamine equilibrium could facilitate the interconversion between two possible reactive imine conformers **38*E*** and **38*Z*** (giving zinc-chelated imines **41** and **42**, respectively).[36] Even though other factors cannot be excluded, one possible explanation for the observed reaction outcome is that addition of the incoming Zn-enolate to the *Si* face of intermediate **42** is likely to be favored either kinetically or thermodynamically to yield the compound possessing the observed absolute configuration. At present, it is unclear whether the reaction proceeds under kinetic or thermodynamic control.

Enantiomerically pure γ,γ-difluoro-α,β-enoate **34** was subjected to reaction with SmI_2-*t*-BuOH in THF at 0 °C for 60 min to afford chiral D-Phe-Gly-type fluoroalkene dipeptide isostere (D-Phe-ψ[(Z)-CF=CH]-Gly) **35** in 92% isolated yield. Taken together, the above-described SmI_2-*t*-BuOH reduction protocols were useful for the synthesis of chiral Xaa-Gly-type fluoroalkene isosteres. Introduction of an α-substituent into the isosteres is planned based on the same SmI_2-mediated reduction.

As mentioned above, formation of dienolate species in the SmI_2-mediated reduction of γ,γ-difluoro-α,β-enoates appears to be involved. Trapping of the dienolates with electrophiles, with the exception of proton electrophiles, is envisioned to result in the formation of α-substituted isosteres. In one instance, Molander et al. reported the trap of the Sm-dienolate derived from a γ,δ-epoxy-α,β-enoate with MeI as an electrophile, albeit in relatively low yields (39%).[19a] We also carried out the reaction of γ,γ-difluoro-α,β-enoate **34** with SmI_2 in THF in the presence of BzlBr; however, no α-alkylated isostere was obtained except for reduction product and several unidentified compounds (Table 3, entry 1). Thereupon, examination of electrophiles as possible trapping agents was extended to include carbonyl compounds such as aldehydes and ketones (Table 3, entries 2−4). One representative example of the SmI_2-mediated intermolecular coupling with this type of substrate combination (alkenes and aldehydes or ketones) is the reductive coupling between carbonyl compounds (aldehydes or ketones) and activated alkenes such as α,β-enoates.[37] In this reaction, ketyls derived from reduction of aldehydes or ketones with SmI_2 couple to the β-position of the α,β-enoates to yield γ-lactone derivatives. Kinetic electrophilic trapping of intermediates resulting from γ,γ-difluoro-α,β-enoates with SmI_2 can result in clean conversion of substrates to the desired reduction products. With this in mind, we attempted the SmI_2-mediated reduction of the γ,γ-difluoro-α,β-enoate **34** in the presence of aldehydes or ketones with the intention of nucleophilic attack of the α-carbon in the intermediary conjugated Sm-dienolates onto the carbonyl compounds.[38,39] This would allow coupling of ketyls derived from carbonyl compounds onto the β-carbon.

Reaction of enoate **34** with SmI_2 (3 equiv) in THF in the presence of acetone (3 equiv) at 0 °C for 60 min proceeded smoothly to yield α-substituted aldol compound **43** as a mixture of α-carbon diastereomers in 82% combined yield. No β-substituted product was observed. Although an in-depth mechanistic investigation of the above coupling reaction was not pursued, the fact that α-aldol coupling products were unambiguously obtained in high yields supports the proposed reaction mechanism, where the γ,γ-difluoro-α,β-enoates rather than carbonyl compounds are preferentially reduced with SmI_2 in THF to form dienolates such as **21**, which react with ketones at their α-position.[38,40] The use of diethyl dicarbonate as an electrophile allowed a Claisen-type condensation to give the α-ethoxycarbonyl fluoroalkene isostere **44**.

Synthesis of α-hydroxymethylated fluoroalkene isosteres is of synthetic value with respect to the prepara-

(34) Davis, F. A.; Reddy, R. E.; Szewczyk, J. M.; Reddy, G. V.; Portonovo, P. S.; Zhang, H.; Fanelli, D.; Reddy, R. T.; Zhou, P.; Carroll, P. J. *J. Org. Chem.* **1997**, *62*, 2555−2563.

(35) Successful example of addition to the imine prepared from phenylacetaldehyde, see: Cogan, D. A.; Liu, G.; Ellman, J. *Tetrahedron* **1999**, *55*, 8883−8904.

(36) d'Angelo, J.; Desmaële, D.; Dumas, F.; Guingant, A. *Tetrahedron: Asymmetry* **1992**, *3*, 459−505 and references cited herein.

(37) (a) Fukuzawa, S.; Nakanishi, A.; Fujinami, T.; Sakai, S. *J. Chem. Soc., Chem. Commun.* **1986**, 624−625. (b) Fukuzawa, S.; Nakanishi, A.; Fujinami, T.; Sakai, S. *J. Chem. Soc., Perkin Trans. 1* **1988**, 1669−1675. (c) Molander, G. A.; Kenny, C. *J. Am. Chem. Soc.* **1989**, *111*, 8236−8246. (d) Fukuzawa, S.; Seki, K.; Tatsuzawa, M.; Mutoh, K. *J. Am. Chem. Soc.* **1997**, *119*, 1482−1483. (e) Xu, M.-H.; Wang, W.; Xia, L.-J.; Lin, G.-Q. *J. Org. Chem.* **2001**, *66*, 3953−3962.

(38) Presumably, because the π-electron density is higher at the α-carbon than the γ-carbon, extended dienolate normally reacts with alkylating agents to produce α-substituted-β,γ-unsaturated compounds. see: (a) Caine, D. In *Comprehensive Organic Synthesis*; Trost, B. M., Fleming, I., Eds.; Pergamon Press: Oxford, UK, 1991; Vol. 3, pp 1−63. (b) Fleming, I. *Frontier Orbitals and Organic Chemical Reactions*; John Wiley & Sons: New York, 1976.

(39) SmI_2-mediated conjugated reduction/intramolecular aldol reactions of compounds possessing α,β-enoate and ketone moieties have recently been reported, see: (a) Hutton, T. K.; Muir, K.; Procter, D. J. *Org. Lett.* **2002**, *4*, 2345−2347. (b) Hutton, T. K.; Muir, K.; Procter, D. J. *Org. Lett.* **2003**, *5*, 4811−4814.