Available online at www.sciencedirect.com
SCIENCE DIRECT®
Tetrahedron 60 (2004) 2971–2977
Tetrahedron

# Synthesis of α,α-disubstituted 4-phosphonophenylalanine analogues as conformationally-constrained phosphotyrosyl mimetics☆

Shinya Oishi,[a] Sang-Uk Kang,[a] Hongpeng Liu,[b] Manchao Zhang,[b] Dajun Yang,[b] Jeffrey R. Deschamps[c] and Terrence R. Burke, Jr.[a,*]

[a]*Laboratory of Medicinal Chemistry, Center for Cancer Research, National Cancer Institute, National Institutes of Health, NCI-Frederick, PO Box B, Bldg. 376 Boyles St., Frederick, MD 21702-1201, USA*
[b]*Department of Hematology/Oncology, University of Michigan Medical School, Ann Arbor, MI 48109, USA*
[c]*Laboratory for the Structure of Matter, Naval Research Laboratory, Washington, DC 20375, USA*

Received 1 December 2003; revised 4 February 2004; accepted 4 February 2004

**Abstract**—Syntheses of *N*-Boc (*S*)-4-(diethylphosphono)-(α-methyl)phenylalanine [Boc-(α-Me)Phe(4-$PO_3Et_2$)-OH] (**9**) and *N*-Boc (*S*)-2-amino-6-(diethylphosphono)tetralin-2-carboxylic acid [Boc-Atc(6-$PO_3Et_2$)-OH] (**18**) are reported as conformationally-constrained phosphotyrosyl mimetics suitably protected for peptide synthesis. Both syntheses proceeded through chiral arylhalides that are converted to arylphosphonates by palladium-catalyzed cross coupling with diethylphosphite. These amino acid analogues may be useful in the study of cellular signal transduction processes.


## 1. Introduction

The biological dependence of peptides on tertiary structure has made restriction of conformational flexibility an important component of peptide mimetic design.[1–6] A variety of non-proteinogenic constrained α-amino acids have been developed for this purpose. In the case of tyrosine (Tyr, **1a**), analogues have been reported that limit rotational freedom either through the addition of substituents at the α- or β-positions or through side chain inclusion in an appended ring structure (Fig. 1).[7] One example is α-methyl-Tyr (**2a**), which is a member of the broader class of α-methyl-containing amino acids known to promote turn geometries.[8] Another example is 2-amino-6-hydroxy-tetralin-2-carboxylic acid (**3a**), which can induce turn conformations similar to **2a** as well as severely restrict $\chi_1$ and $\chi_2$ angles.[9,10]

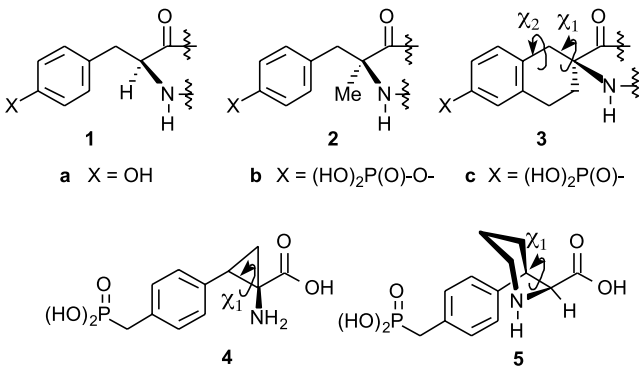


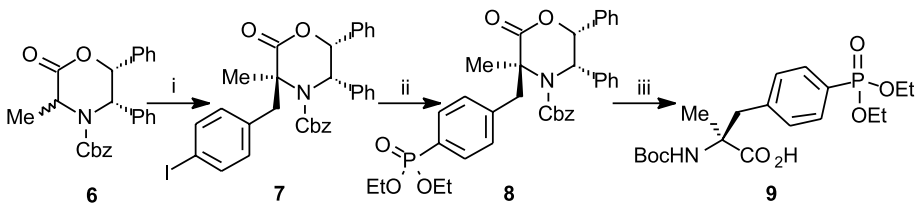

**Figure 1.** Structures of pTyr and pTyr mimetics.

The biological activities of proteins are also highly influenced by post-translational modifications. This is exemplified by the conversion of Tyr residues to phosphotyrosyl residues (pTyr, **1b**) via protein-tyrosine kinases (PTKs). In aberrant cellular signal transduction this process can be critical to several diseases, including certain cancers.[11,12] Derivatives of pTyr such as α-methyl-pTyr (**2b**) that include elements of conformational constraint can be useful in the design of pTyr-dependent signaling antagonists.[13,14] This type of analogue is of limited use in cellular systems where the phosphoryl ester bond is labile to protein-tyrosine phosphatases (PTPs). Accordingly, the development of PTP-stable phosphonate-based congeners has been undertaken, as exemplified by **1c**[15,16] and (α-methyl)-*p*-phosphonophenylalanine ((α-methyl)-Ppp, **2c**).[17] These compounds can retain much of the biological potency of the parent phosphate-containing analogues.[17,18] Although

☆ Supplementary data associated with this article can be found in the online version, at doi: 10.1016/j.tet.2004.02.005

*Keywords*: Enantioselective; Phosphotyrosyl mimetic; Conformational constraint.
* Corresponding author. Tel.: +1-301-846-5906; fax: +1-301-846-6033; e-mail address: tburke@helix.nih.gov

0040–4020/$ - see front matter © 2004 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tet.2004.02.005



**Scheme 1.** (i) 4-Iodobenzyl bromide, KHMDS (86% yield); (ii) $(EtO)_2PHO$, $Et_3N$, $Pd(PPh_3)_4$ (74% yield); (iii) (a) $H_2$, Pd·C, (b) $Boc_2O$, $Et_3N$ (61% yield).

phosphonate-containing $\chi_1$-constrained pTyr mimetics such as **4**[19] and **5**[20] have been disclosed, to date the $\chi_1$, $\chi_2$-restricted pTyr mimetic **3c** has yet to be reported, in spite of its potential usefulness. Work was therefore undertaken to prepare **3c** in its enantio-pure form, suitably protected for incorporation into peptides using standard methodologies. Efforts were also devoted to develop a new enantioselective synthesis of orthogonally-protected **2c**.

### 1.1. Synthesis of *N*-Boc (*S*)-4-(diethylphosphono)-(α-methyl)phenylalanine

The synthesis of (α-methyl)-Ppp (**2c**) in racemic form as the diethyl phosphonate ester bearing *N*-Fmoc protection has been achieved previously[17] via a Schiff base approach.[19] In the current work, enantioselective synthesis of the (*S*)-*N*-Boc variant of this compound (*N*-Boc (α-methyl)-Ppp$(OEt)_2$, **9**) was accomplished using the 3-methyl-substituted Williams oxazinone **6**[21] in a protocol similar to that used to prepare non-phosphorus-containing (α-methyl)-pTyr mimetics (Scheme 1).[14] Reaction of **6** with 4-iodobenzyl bromide in the presence of KHMDS afforded the fully-protected (α-methyl)-(4-iodo)phenylalanine (**7**),[22] which represents a potential common intermediate for the enantioselective synthesis of a variety of 4-substituted phenylalanines. While attempted $Pd(PPh_3)_4$-mediated replacement of iodine using $(Bu^tO)_2P(O)H$ failed, product **8** could be obtained in good yield using the sterically less demanding $(EtO)_2P(O)H$. Hydrogenolytic deprotection and reaction with Boc anhydride provided the desired target compound **9** in 39% overall yield.

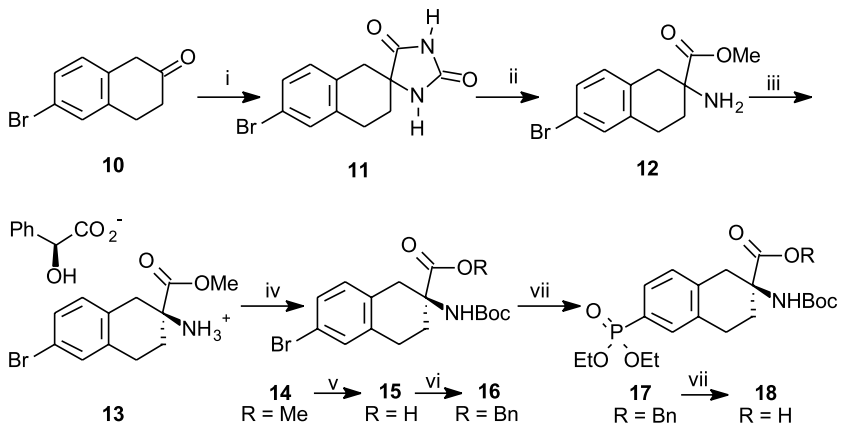
### 1.2. Synthesis of *N*-Boc (*S*)-2-amino-6-(diethylphosphono)tetralin-2-carboxylic acid

The route to cyclic α,α-substituted analogue **18** involved the synthesis of racemic 2-amino-6-bromo-tetralin-2-carboxylic acid methyl ester (**12**), which was obtained in classic fashion[23,24] from commercially available 6-bromo-2-tetralone **10** through the intermediacy of spirohydantoin **11** (Scheme 2). Resolution of (±)-**12** as the (L)-(+)-mandelic acid salt according to literature procedures[23] provided the ammonium salt **13** bearing the L-configuration as determined by single crystal X-ray crystallography. The optical purity of **13** was verified by HPLC analysis of **14** using a chiral stationary phase (ee >98%). Amino ester **13** was converted in two steps to the *N*-Boc amino acid **15** (77% yield), which represents a versatile intermediate for the preparation of a variety of 6-substituted analogues using cross-coupling chemistries. Transient protection of **15** as its benzyl ester (**16**) allowed $Pd(PPh_3)_4$-mediated introduction of the 6-$(EtO)_2PO$-group, which provided the conformationally-constrained pTyr mimetic **18** following hydrogenolytic de-esterification.

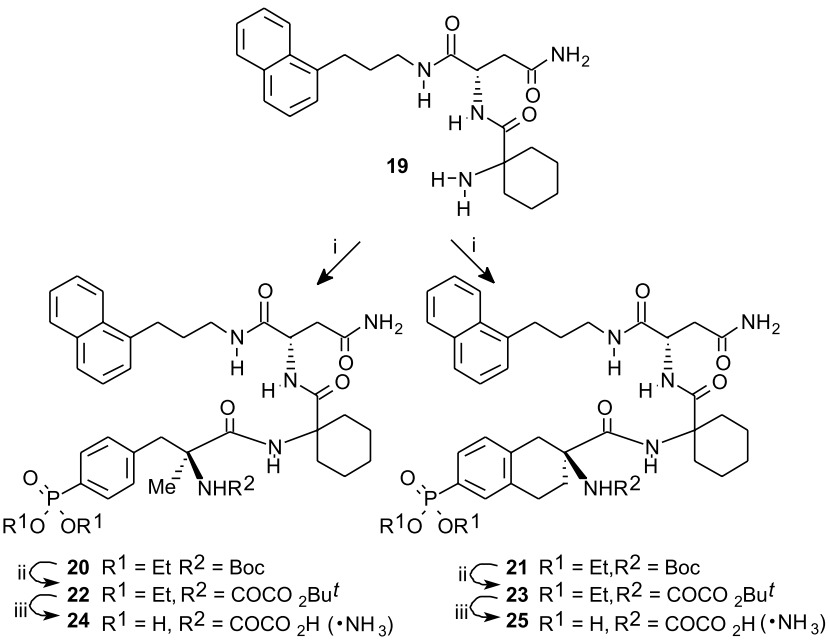
### 1.3. Incorporation of pTyr mimetics 9 and 18 in Grb2 SH2 domain-directed peptides

In order to demonstrate the suitability of conformationally-constrained pTyr mimetics **9** and **18** for peptide synthesis, Grb2 SH2 domain-directed tripeptides **24** and **25** were prepared, respectively (Scheme 3). This platform has been used extensively to investigate a variety of pTyr mimetics, including the $\chi_1$-constrained analogue **5**.[20] Coupling of the α,α-disubstituted residues with the sterically-crowded amino group of dipeptide **19**[25] was achieved using tetramethylfluoroformamidinium hexafluorophosphate (TFFH).[26] Final products **24** and **25** were obtained as their ammonium salts following deprotection using trimethylsilyl iodide (TMSI) and TFA.

Although the primary purpose in preparing peptides **24** and



**Scheme 2.** (i) KCN, $(NH_4)_2CO_3$ (81% yield); (ii) (a) $Ba(OH)_2$, (b) $SOCl_2$, MeOH (54% yield); (iii) L-(+)-mandelic acid (25% yield); (iv) $Boc_2O$, $Et_3N$ (79% yield); (v) LiOH (98% yield); (vi) BnBr, $Pr^i_2EtN$ (97% yield); (vii) $(EtO)_2PHO$, $Et_3N$, $Pd(PPh_3)_4$ (93% yield); (iii) (a) $H_2$, Pd·C (99% yield).




**Scheme 3.** (i) **9** or **18**, TFFH, $Pr^i_2EtN$; (ii) (a) TFA, anisole, (b) $Bu^tO_2CCOCl$, $Pr^i_2EtN$; (iii) (a) TMSI, (b) TFA–$H_2O$ (95:5).

**25** was to verify the suitability of amino acid analogues **9** and **18** for peptide synthesis, once in hand, it was also of interest to determine the Grb2 SH2 domain-binding potency of these peptides. In an ELISA-based extracellular binding assay, the unconstrained $N^{\alpha}$-oxalyl Pmp-containing variant of **24** and **25** had previously been shown to exhibit low nanomolar Grb2 SH2 domain-binding potency.[25] Therefore, it was surprising that in a similar ELISA-based Grb2 SH2 domain binding assay,[27] peptides **24** and **25** displayed poor affinity ($IC_{50}$ >10 μM). This was reminiscent of a report that conformational constraint of the pTyr-mimicking residue is deleterious to SH2 domain-binding potency.[20] However, it was in marked contrast to the findings of a recent study employing a cyclopropane-based pTyr mimetic that was equipotent to the unconstrained parent.[28]

## 2. Conclusions

Reported herein are the syntheses of two conformationally-constrained pTyr mimetics in protected forms suitable for incorporation into peptides using standard methodologies. Chiral arylhalide intermediates in both approaches represent potential starting points for the cross-coupling synthesis of additional constrained phenylalanyl analogues.

## 3. Experimental

### 3.1. General synthetic

Reactions were carried out under argon in oven-dried glassware using standard gas-tight syringes, cannulas and septa. Anhydrous solvents were purchased from Aldrich Chemical Corporation and used without further drying. Melting points were measured using a MEL-TEMP II apparatus and are uncorrected. Elemental analyses were obtained from Atlantic Microlab, Inc., Norcross, GA. $^1H$ NMR data were recorded on a Varian 400 MHz spectrometer and are reported in ppm relative to TMS and referenced to the solvent in which they were run. Fast atom bombardment mass spectra (FABMS) were acquired with a VG analytical 7070E mass spectrometer under the control of a VG 2035 data system. HPLC separations were conducted using a Cosmosil $5C_{18}$-ARII (20×250 mm) with a solvent system of 0.1% aqueous $NH_3$ (v/v, solvent A)/0.1% $NH_3$ in MeCN (v/v, solvent B) or a CHIRALCEL OD (10×250 mm) using hexanes (solvent C) and *i*-PrOH (solvent D).

**3.1.1. (3*S*,5*S*,6*R*)-4-(Benzyloxycarbonyl)-5,6-diphenyl-3-(4-iodo)benzyl-3-methyl-2,3,5,6-tetrahydro-4*H*-1,4-oxazin-2-one (7).** To a stirred solution of **6**[21] (2.00 g, 4.98 mmol) in dry THF (40 mL) was added a solution of KHMDS in toluene (0.5 M, 12.0 mL, 6.0 mmol) at −78 °C under argon. After 5 min, a solution of 4-iodobenzyl bromide (1.84 g, 6.22 mmol) was added dropwise at −78 °C, and stirring was continued for 30 min, followed by quenching with a saturated $NH_4Cl$ solution. The mixture was extracted with EtOAc, and the extract was washed with water and dried over $MgSO_4$. Concentration under reduced pressure followed by flash chromatography over silica gel with *n*-hexanes–EtOAc (20:1) provided **7** as colorless crystals (2.67 g, 86% yield): mp 98–100 °C; $[\alpha]_D^{21}$ +156.0 (*c* 0.51, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$) δ 2.01 (s, 3H), 3.08 (d, *J*=13.2 Hz, 1H), 3.88 (m, 1H), 4.22 (d, *J*=13.4 Hz, 1H), 5.01 (m, 1H), 5.10 (d, *J*=12.2 Hz, 1H), 5.23 (d, *J*=12.2 Hz, 1H), 6.60–7.63 (m, 19H). $^{13}C$ NMR (400 MHz, $CDCl_3$) δ 25.3, 43.8, 59.9, 66.3, 67.6, 78.8, 93.1, 126.2, 128.1, 128.2, 128.6, 128.9, 132.0, 134.3, 135.6, 136.0, 136.3, 137.9, 153.5, 172.8. Anal. calcd for $C_{32}H_{28}INO_4$: C, 62.24; H, 4.57; N, 2.27. Found: C, 62.30; H, 4.63; N, 2.26. FABMS *m/z* 618 ($MH^+$).

**3.1.2. (3*S*,5*S*,6*R*)-4-(Benzyloxycarbonyl)-5,6-diphenyl-3-(4-diethylphosphono)benzyl-3-methyl-2,3,5,6-tetrahydro-4*H*-1,4-oxazin-2-one (8).** A mixture of **7** (2.53 g, 4.09 mmol), diethyl phosphite (0.580 mL, 4.50 mmol),

$Et_3N$ (0.627 mL, 4.50 mmol) and $Pd(PPh_3)_4$ (236 mg, 0.204 mmol) in dry toluene (5 mL) was stirred at reflux for 6 h at 90 °C under argon. The whole was extracted with EtOAc, and the extract was washed with $H_2O$ and brine, and dried over $MgSO_4$. Concentration under reduced pressure followed by flash chromatography over silica gel with *n*-hexanes–EtOAc (1:1) provided **8** as a colorless oil (1.90 g, 74% yield): $[\alpha]_D^{22}$ +189.9 (*c* 0.28, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.16 (t, *J*=7.0 Hz, 3H), 1.27 (t, *J*=7.0 Hz, 3H), 1.57 (s, 9H), 2.02 (s, 3H), 3.17 (d, *J*=13.4 Hz, 1H), 3.80 (m, 1H), 3.83–4.16 (m, 4H), 4.37 (d, *J*=13.4 Hz, 1H), 5.02 (m, 1H), 5.14 (d, *J*=12.3 Hz, 1H), 5.24 (d, *J*=12.1 Hz, 1H), 6.64 (d, *J*=7.2 Hz, 2H), 6.86 (d, *J*=6.8 Hz, 2H), 6.96–7.56 (m, 13H), 7.72 (m, 2H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 16.2, 16.3, 5.4, 44.2, 59.6, 62.1, 62.2, 66.4, 67.7, 78.7, 125.8, 128.0, 128.1, 128.2, 128.4, 128.6, 128.8, 130.2, 132.2, 134.2, 135.4, 135.9, 141.3, 153.5, 172.6. FABMS *m*/*z* 628 ($MH^+$). Anal. calcd for $C_{36}H_{38}NO_7P$: C, 68.89; H, 6.10; N, 2.23. Found: C, 68.49; H, 6.24; N, 2.41.

**3.1.3. (2*S*)-2-(*tert*-Butyloxycarbonyl)amino-3-(4-diethylphosphono)phenylpropionic acid [Boc-(α-Me)Phe(4-$PO_3Et_2$)-OH] (9).** Oxazinone **8** (4.70 g, 7.48 mmol) was treated using Pd·C (10%, 1.0 g) in THF–EtOH (1:1, 300 mL) under $H_2$. After filtration through Celite, the solution was concentrated under reduced pressure to yield the amino acid. This was taken up in DMF–$H_2O$ (4:1, 25 mL) and reacted with $Boc_2O$ (2.45 g, 11.23 mmol) and $Et_3N$ (3.13 mL, 22.4 mmol) at 0 °C and with stirring for 4 days at room temperature. The mixture was acidified with saturated citric acid solution and extracted with EtOAc. The extract was washed with $H_2O$ and brine, and dried over $MgSO_4$. Concentration under reduced pressure followed by flash chromatography over silica gel using $CH_2Cl_2$–MeOH (10:1) provided **9** as colorless gum (1.92 g, 61% yield): $[\alpha]_D^{22}$ +22.0 (*c* 0.95, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.31 (m, 6H), 1.45 (s, 9H), 1.74 (s, 3H), 3.28 (d, *J*=13.3 Hz, 1H), 3.69 (m, 1H), 4.00–4.25 (m, 4H), 5.56 (s, 1H), 7.35 (dd, *J*=8.2, 4.2 Hz, 2H), 7.66 (dd, *J*=13.3, 8.0 Hz, 2H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 16.1, 16.2, 24.2, 28.4, 41.1, 60.4, 62.5, 62.7, 79.1, 124.2, 126.1, 130.2, 131.5, 142.8, 154.3, 175.3. FABMS *m*/*z* 416 ($MH^+$). Anal. calcd for $C_{19}H_{30}NO_7P$: C, 54.93; H, 7.28; N, 3.37. Found: C, 55.24; H, 7.42; N, 3.22.

**3.1.4. 3′,4′-Dihydro-6′-bromo-spiro[imidazolidine-4,2′(1′*H*)-naphthalene]-2,5-dione (11).** A mixture of 6-bromo-2-tetralone **10** (20.3 g, 90.2 mmol), KCN (7.63 mL, 117 mmol), $(NH_4)_2CO_3$ (78.0 g, 811 mmol) in 50% aqueous EtOH (540 mL) was stirred at reflux for 1 h. After evaporation of EtOH, the suspension was filtrated to provide **11** as brown powder (21.7 g, 81% yield): mp 285–287 °C (decomp.); $^1H$ NMR (400 MHz, DMSO-$d_6$) $\delta$ 1.80 (m, 1H), 1.92 (m, 1H), 2.75 (d, *J*=17.0 Hz, 1H), 2.90 (m, 2H), 3.04 (d, *J*=17.3 Hz, 1H), 7.05 (d, *J*=8.0 Hz, 1H), 7.30 (dd, *J*=8.0, 2.2 Hz, 1H), 7.35 (d, *J*=2.2 Hz, 1H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 24.5, 29.5, 36.3, 60.3, 118.8, 128.5, 130.9, 131.0, 132.1, 137.7, 156.2, 177.9. FABMS *m*/*z* 295 ($MH^+$). Anal. calcd for $C_{12}H_{11}BrN_2O_2$: C, 48.84; H, 3.76; N, 9.49. Found: C, 49.27; H, 3.79; N, 9.20.

**3.1.5. Methyl 2-amino-1,2,3,4-tetrahydro-6-bromo-2-naphthalenecarboxylate (12).** A suspension of **11** (21.5 g, 72.9 mmol) and $Ba(OH)_2$ in $H_2O$ (750 mL) was stirred at reflux for 36 h. The mixture was acidified with 6 N $H_2SO_4$, and filtered and the filter pad was washed with MeOH repeatedly. The combined filtrate was concentrated under reduced pressure to yield a suspension, which was adjusted to pH 6.0 with $NH_4OH$ and the resulting solid was collected by filtration to provide the crude amino acid (18.7 g, 95%). A mixture of amino acid (17.7 g) and $SOCl_2$ (14.3 mL, 197 mmol) in MeOH (350 mL) was stirred at reflux for 3 h. After filtration, the filtrate was concentrated under reduced pressure to give the methyl ester as an HCl salt. This was neutralized with excess $NaHCO_3$ in toluene–$H_2O$ (5:9, 840 mL) and the organic phase was separated and dried over $Na_2SO_4$. Concentration under reduced pressure followed by flash chromatography over silica gel with *n*-hexanes–EtOAc (1:1) gave **12** as a pale yellow oil (10.7 g, 57% yield): $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.62 (s, 2H), 1.87 (m, 1H), 2.12 (ddd, *J*=13.4, 9.6, 6.1 Hz, 1H), 2.68 (d, *J*=16.5 Hz, 1H), 2.78 (dt, *J*=17.2, 5.5 Hz, 1H), 2.98 (m, 1H), 3.21 (d, *J*=16.4 Hz, 1H), 3.74 (s, 3H), 6.94 (d, *J*=8.0 Hz, 1H), 7.20–7.28 (m, 2H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 25.3, 31.6, 38.8, 52.4, 56.2, 119.6, 129.0, 130.9, 131.4, 132.7, 137.1, 177.0. FABMS *m*/*z* 284 ($MH^+$). Anal. calcd for $C_{12}H_{14}BrNO_2$; C: 50.72; H: 4.97; N: 4.93. Found: C, 50.90; H, 5.00; N, 4.93.

**3.1.6. (*S*)-2-Amino-1,2,3,4-tetrahydro-6-bromo-2-naphthalenecarboxylic acid methyl ester (*S*)-mandelic acid salt (13).** To a solution of amino ester **12** (10.4 g, 36.8 mmol) in $Et_2O$–MeOH (3:1, 150 mL) was added L-(+)-mandelic acid (5.55 g, 36.5 mmol). Repeated recrystalization from $Et_2O$–MeOH (3:1) provided salt **13** as colorless crystals (4.07 g, 25% yield): mp 138–140 °C; Anal. calcd for $C_{20}H_{22}BrNO_5$: C, 55.06; H, 5.08; N, 3.21. Found: C, 54.77; H, 5.08; N, 3.21.

**3.1.7. Methyl (*S*)-2-(*tert*-butyloxycarbonyl)amino-1,2,3,4-tetrahydro-6-bromo-2-naphthalenecarboxylate (14).** To a stirred suspension of **13** (50 mg, 0.114 mmol) in $CHCl_3$ (0.160 mL) were added $Boc_2O$ (55 mg, 0.252 mmol) and $Et_3N$ (0.070 mL, 0.504 mmol) at 0 °C, and stirring was continued for 2 days at room temperature. The mixture was extracted with EtOAc, and the extract was successively washed with 5% citric acid solution, brine, 5% $NaHCO_3$ solution and brine, and dried over $MgSO_4$. Concentration under reduced pressure followed by flash chromatography over silica gel with *n*-hexanes–EtOAc (7:1) provided **14** as colorless crystals (35 mg, 79% yield): mp 115–117 °C; $[\alpha]_D^{21}$ +30.6 (*c* 0.68, $CHCl_3$). Enantiomeric purity, determined by HPLC analysis using a CHIRALCEL column (isocratic, 10% D in C), was >98%. $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.41 (s, 9H), 2.10 (m, 1H), 2.46 (m, 1H), 2.82 (dd, *J*=8.3, 4.8 Hz, 2H), 2.92 (d, *J*=16.8 Hz, 1H), 3.22 (d, *J*=16.7 Hz, 1H), 3.76 (s, 3H), 4.72 (s, 1H), 6.92 (d, *J*=8.0 Hz, 1H), 7.23–7.29 (m, 2H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 25.1, 28.2, 37.6, 52.5, 57.6, 120.0, 129.2, 130.9, 131.4, 131.6, 137.2, 154.9, 174.2. FABMS *m*/*z* 384 ($MH^+$). Anal. calcd for $C_{17}H_{22}BrNO_4$: C, 53.14; H, 5.77; N, 3.65. Found: C, 53.27; H, 5.78; N, 3.70.

**3.1.8. (*S*)-2-(*tert*-Butyloxycarbonyl)amino-1,2,3,4-tetrahydro-6-bromo-2-naphthalenecarboxylic acid (15).** To a

stirred solution of amino ester **14** (2.00 g, 5.20 mmol), in THF (25 mL) was added 1 N LiOH (15.6 mL, 15.6 mmol) at 0 °C, and stirring was continued overnight at room temperature. The mixture was acidified with saturated citric acid solution, concentrated under reduced pressure and extracted with EtOAc. The extract was washed with $H_2O$ and brine, and dried over $MgSO_4$. Concentration and recrystalization from $Et_2O$–MeOH (5:1) gave **15** as colorless crystals (1.90 g, 98% yield): mp 183–185 °C; $[\alpha]_D^{22}$ +22.8 (*c* 0.49, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.42 (s, 9H), 2.10 (ddd, *J*=13.6, 10.5, 6.8 Hz, 1H), 2.53 (m, 1H), 2.84 (m, 2H), 2.94 (d, *J*=17.1 Hz, 1H), 3.31 (d, *J*=16.8 Hz, 1H), 4.79 (br, 1H), 6.95 (d, *J*=8.0 Hz, 1H), 7.27 (m, 2H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 24.6, 28.1, 56.6, 77.9, 118.3, 128.2, 130.5, 131.3, 133.5, 137.7, 155.0,175.4. FABMS *m*/*z* 370 ($MH^+$). Anal. calcd for $C_{16}H_{20}BrNO_4$; C, 51.90; H, 5.44; N, 3.78. Found: C, 51.90; H, 5.47; N, 3.66.

**3.1.9. Benzyl (*S*)-2-(*tert*-butyloxycarbonyl)amino-1,2,3,4-tetrahydro-6-bromo-2-naphthalenecarboxylate (16).** To a stirred solution of **15** (1.88 g, 5.07 mmol) in DMF (5.5 mL) were added BnBr (0.664 mL, 5.58 mmol) and $Pr^i_2EtN$ (1.06 mL, 6.09 mmol) at 0 °C and stirring was continued for 24 h at room temperature. The mixture was extracted with EtOAc, and the extract was washed with saturated citric acid solution, $H_2O$, 5% $NaHCO_3$ solution and brine, and dried over $Na_2SO_4$. Concentration under reduced pressure followed by flash chromatography over silica gel with *n*-hexane–EtOAc (7:1) provided **16** as colorless crystals (2.27 g, 97% yield): mp 89–91 °C; $[\alpha]_D^{22}$ +16.9 (*c* 0.92, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.37 (s, 9H), 2.12 (ddd, *J*=13.6, 9.7, 7.5 Hz, 1H), 2.46 (m, 1H), 2.79 (m, 2H), 2.92 (d, *J*=16.5 Hz, 1H), 3.26 (d, *J*=16.6 Hz, 1H), 4.75 (br, 1H), 5.18 (s, 2H), 6.91 (d, *J*=8.0 Hz, 1H), 7.21–7.27 (m, 2H), 7.29–7.38 (m, 5H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 25.1, 28.2, 37.6, 57.7, 67.2, 120.0, 128.1, 128.2, 128.5, 129.2, 130.9, 131.5, 135.6, 137.2, 149.1, 154.8, 173.5. FABMS *m*/*z* 460 ($MH^+$). Anal. calcd for $C_{23}H_{26}BrNO_4$: C, 60.01; H, 5.69; N, 3.04. Found: C, 59.64; H, 5.70; N, 2.92.

**3.1.10. (*S*)-2-(*tert*-Butyloxycarbonyl)amino-1,2,3,4-tetrahydro-6-(diethylphosphono)-2-naphthalenecarboxylic acid benzyl ester (17).** Treatment of **16** (2.21 g, 4.80 mmol) with diethyl phosphite using a procedure similar to that described for the preparation **8** from **7**, gave **17** as a colorless oil (1.91 g, 93% yield): $[\alpha]_D^{22}$ +9.30 (*c* 1.13, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.33 (t, *J*=7.0 Hz, 6H), 1.38 (s, 9H), 2.15 (ddd, *J*=13.6, 9.2, 7.3 Hz, 1H), 2.45 (m, 1H), 2.86 (m, 2H), 3.04 (d, *J*=17.3 Hz, 1H), 3.40 (d, *J*=17.0 Hz, 1H), 4.11 (m, 4H), 4.79 (s, 1H), 5.19 (s, 2H), 7.15 (dd, *J*=7.8, 4.3 Hz, 1H), 7.28–7.38 (m, 5H), 7.49–7.60 (m, 2H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 16.3, 25.1, 28.2, 28.9, 38.0, 57.7, 62.0, 67.2, 125.2, 127.1, 128.1, 128.2, 128.5, 129.1, 129.5, 132.4, 135.3, 135.6, 137.7, 154.8, 173.5. HR-FABMS *m*/*z* calcd for $C_{27}H_{37}NO_7P$ ($MH^+$) 518.2308, found: 518.2264.

**3.1.11. (*S*)-2-(*tert*-Butyloxycarbonyl)amino-1,2,3,4-tetrahydro-6-(diethylphosphono)-2-naphthalenecarboxylic acid [Boc-Atc(6-$PO_3Et_2$)-OH] (18).** Benzyl ester **17** (1.84 g, 3.55 mmol) was treated using Pd·C (10%, 300 mg) in EtOAc (100 mL) under an $H_2$ atmosphere. After filtration through Celite and concentration under reduced pressure, purification by flash chromatography over silica gel with $CH_2Cl_2$–MeOH (10:1) provided **18** as a colorless powder (1.51 g, 99% yield): mp 75–77 °C; $[\alpha]_D^{22}$ +7.19 (*c* 0.67, $CHCl_3$); $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.33 (t, *J*=7.0 Hz, 6H), 1.42 (s, 9H), 2.14 (m, 1H), 2.51 (m, 1H), 2.91 (m, 2H), 3.07 (d, *J*=17.3 Hz, 1H), 3.43 (d, *J*=17.3 Hz, 1H), 4.13 (m, 4H), 4.87 (br, 1H), 7.17 (dd, *J*=7.8, 4.1 Hz, 1H), 7.53 (dd, *J*=12.9, 7.8 Hz, 1H), 7.59 (d, *J*=14.1 Hz, 1H). $^{13}C$ NMR (400 MHz, $CDCl_3$) $\delta$ 16.3, 25.1, 28.2, 37.8, 57.6, 62.3, 80.6, 124.7, 126.6, 129.1, 129.6, 132.4, 135.4, 138.0, 155.6, 177.0. FABMS *m*/*z* 428 ($MH^+$). Anal. calcd for $C_{20}H_{30}NO_7P$: C, 56.20; H, 7.07; N, 3.28. Found: C, 56.07; H, 7.10; N, 3.22.

**3.1.12. Boc-(α-Me)Phe(4-$PO_3Et_2$)-$Ac_6$c-Asn-$(CH_2)_3$-(1-naphthyl) (20).** To a stirred solution of **9** (215 mg, 0.518 mmol) in dry DMF (2 mL) was added TFFH (136 mg, 0.518 mmol) at room temperature. After 10 min, amine **19**[25] (200 mg, 0.471 mmol) and $Pr^i_2EtN$ (0.180 mL, 1.03 mmol) were added to the above mixture at room temperature, and stirring was continued for 24 h at 50 °C. The mixture was extracted with EtOAc, and the extract was washed successively with saturated citric acid solution, brine, saturated $NaHCO_3$ solution and brine, and dried over $Na_2SO_4$. Concentration followed by flash chromatography over silica gel with $CH_2Cl_2$–MeOH (100:0 to 10:1) gave **20** as colorless semisolid (282 mg, 71% yield): $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.20–1.53 (m, 21H), 1.58–1.77 (m, 3H), 1.82 (m, 1H), 1.90–2.11 (m, 4H), 2.17 (m, 1H), 2.65 (dd, *J*=15.3, 5.1 Hz, 1H), 2.90 (d, *J*=13.6 Hz, 1H), 3.04 (dd, *J*=15.3, 5.6 Hz, 1H), 3.12 (m, 2H), 3.35 (m, 1H), 3.41 (m, 1H), 3.51 (d, *J*=13.6 Hz, 1H), 4.14 (m, 4H), 4.66 (s, 1H), 4.71 (m, 1H), 5.35 (br, 1H), 6.22 (br, 1H), 7.04 (m, 3H), 7.34 (m, 2H), 7.43 (m, 2H), 7.53 (t, *J*=5.6 Hz, 1H), 7.66 (m, 3H), 7.79 (m, 1H), 7.85 (d, *J*=8.3 Hz, 1H), 7.93 (d, *J*=8.3 Hz, 1H). FABMS *m*/*z* 822 ($MH^+$). Anal. calcd for $C_{43}H_{60}N_5O_9P{\cdot}0.5H_2O$: C, 62.15; H, 7.40; N, 8.43. Found: C, 62.00; H, 7.38; N, 8.37.

**3.1.13. Boc-Atc(6-$PO_3Et_2$)-$Ac_6$c-Asn-$(CH_2)_3$-(1-naphthyl) (21).** Using a procedure similar to that described for the preparation of peptide **20** from **19**, coupling of **18** (200 mg, 0.471 mmol) with **19** provided **21** as a colorless semisolid (282 mg, 71% yield): $^1H$ NMR (400 MHz, $CDCl_3$) $\delta$ 1.21–1.43 (m, 18H), 1.52–1.84 (m, 5H), 1.85–2.09 (m, 5H), 2.22 (m, 1H), 2.39 (m, 1H), 2.57–2.91 (m, 5H), 3.08 (m, 2H), 3.25 (d, *J*=17.0 Hz, 1H), 3.34 (m, 2H), 4.13 (m, 4H), 4.79 (dt, *J*=8.2, 5.8 Hz, 1H), 4.81 (s, 1H), 5.35 (br, 1H), 6.36 (br, 1H), 7.02 (dd, *J*=7.8, 4.1 Hz, 1H), 7.20 (br, 1H), 7.30 (m, 2H), 7.40 (m, 2H), 7.50 (dd, *J*=17.9, 7.5 Hz, 1H), 7.55 (m, 2H), 7.64 (m, 1H), 7.80 (m, 2H), 8.01 (d, *J*=8.3 Hz, 1H). FABMS *m*/*z* 834 ($MH^+$). Anal. calcd for $C_{44}H_{60}N_5O_9P{\cdot}H_2O$: C, 62.03; H, 7.34; N, 8.22. Found: C, 62.29; H, 7.30; N, 8.32.

**3.1.14. *tert*-Bu$^t$O-$(CO)_2$-(α-Me)Phe(4-$PO_3Et_2$)-$Ac_6$c-Asn-$(CH_2)_3$-(1-naphthyl) (22).** Protected peptide **20** (93 mg, 0.113 mmol) was treated with TFA-anisole (10:1, 5.5 mL) for 2 h at room temperature then the reaction mixture was concentrated and dissolved in dry DMF (1 mL). To this were added *tert*-butyl oxalyl chloride

(24 mg, 0.169 mmol) and $Pr^i_2EtN$ (0.059 mL, 0.339 mmol) at 0 °C, and stirring was continued for 2 h at 50 °C. The mixture was extracted with EtOAc, and the extract was washed successively with saturated citric acid solution, brine, saturated $NaHCO_3$ solution and brine, and dried over $Na_2SO_4$. Concentration followed by flash chromatography over silica gel with $CH_2Cl_2$–MeOH (100:0 to 10:1) gave **20** as colorless semisolid (56 g, 58% yield): $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 1.18–1.45 (m, 12H), 1.49 (s, 9H), 1.56–1.82 (m, 5H), 1.99 (m, 3H), 2.29 (m, 1H), 2.71 (dd, $J$=14.6, 5.3 Hz, 1H), 2.82 (dd, $J$=14.6, 6.3 Hz, 1H), 3.03 (d, $J$=13.6 Hz, 1H), 3.11 (m, 2H), 3.34 (m, 2H), 3.46 (d, $J$=13.4 Hz, 1H), 4.12 (m, 4H), 4.65 (m, 1H), 5.65 (br, 1H), 6.89 (br, 1H), 7.23 (dd, $J$=8.0, 3.6 Hz, 2H), 7.28 (s, 1H), 7.32–7.38 (m, 2H). 7.40–7.50 (m, 3H), 7.52 (m, 1H), 7.66 (dd, $J$=6.5, 2.6 Hz, 1H), 7.70–7.84 (m, 4H), 8.04 (d, $J$=8.0 Hz, 1H). FABMS $m/z$ 850 ($MH^+$). Anal. calcd for $C_{44}H_{60}N_5O_{10}P{\cdot}H_2O$: C, 60.89; H, 7.20; N, 8.07. Found: C, 61.11; H, 7.03; N, 8.09.

**3.1.15. *tert*-BuO-(CO)$_2$-Atc(6-PO$_3$Et$_2$)-Ac$_6$c-Asn-(CH$_2$)$_3$-(1-naphthyl) (23).** Using a procedure similar to that described for the preparation of peptide **22** from **20**, coupling of **21** (265 mg, 0.317 mmol) with *tert*-butyl oxalyl chloride gave **23** as colorless semisolid (183 mg, 66% yield): $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 0.83 (m, 2H), 1.14 (m, 1H), 1.33 (m, 6H), 1.36–1.62 (m, 12H), 1.65 (m, 2H), 1.86–2.06 (m, 3H), 2.24 (m, 3H), 2.78 (m, 4H), 3.10 (m, 3H), 3.23 (m, 1H), 3.37 (m, 1H), 3.61 (d, $J$=17.3 Hz, 1H), 4.11 (m, 4H), 4.66 (m, 1H), 5.52 (br, 1H), 6.74 (br, 1H), 6.93 (s, 1H), 7.20 (dd, $J$=4.3, 3.9 Hz, 1H), 7.35 (m, 3H), 7.44 (m, 3H), 7.60 (m, 3H), 7.68 (dd, $J$=7.0, 2.2 Hz, 1H), 7.82 (m, 1H), 8.04 (d, $J$=8.2 Hz, 1H). FABMS $m/z$ 862 ($MH^+$). Anal. calcd for $C_{45}H_{60}N_5O_{10}P{\cdot}H_2O$: C, 61.42; H, 7.10; N, 7.96. Found: C, 61.58; H, 7.13; N, 7.90.

**3.1.16. HO-(CO)$_2$-(α-Me)Phe(4-PO$_3$H$_2$)-Ac$_6$c-Asn-(CH$_2$)$_3$-(1-naphthyl) (24).** To a stirred solution of protected peptide **22** (43 mg, 0.050 mmol) in MeCN (1 mL) were added thioanisole (0.100 mL) and TMSI (0.711 mL) at 0 °C and the mixture was stirred for 30 min at 0 °C and for an additional 1 h at room temperature. After concentration, the residue was dissolved in 95% TFA (10 mL), and stirring was continued for 2 h at room temperature. The mixture was concentrated and extracted with 0.1% $NH_4OH$, and the extract was washed with $Et_2O$. The aqueous solution was purified by preparative HPLC (linear gradient 3–13% B in A over 30 min) to give **24** as colorless powder (39 mg, 98% yield): $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 1.18 (s, 3H), 1.21 (m, 2H), 1.40–1.60 (m, 4H), 1.66 (m, 1H), 1.74–1.96 (m, 4H), 2.29 (m, 1H), 2.37 (m, 1H), 2.90 (m, 2H), 3.05 ($t$=8.0 Hz, 2H), 3.18 (m, 2H), 3.30 (d, $J$=12.6 Hz, 1H), 4.25 (m, 1H), 6.62 (br, 1H), 7.29 (m, 2H), 7.37 (m, 2H), 7.47 (m, 3H), 7.61 (m, 2H), 7.73 (d, $J$=7.8 Hz, 1H), 7.87 (m, 1H), 7.96 (d, $J$=7.5 Hz, 1H), 8.07 (m, 2H), 8.30 (br, 1H), 8.35 (s, 1H). FABMS $m/z$ 736 [$(M-H)^-$].

**3.1.17. HO-(CO)$_2$-Atc(6-PO$_3$H$_2$)-Ac$_6$c-Asn-(CH$_2$)$_3$-(1-naphthyl) (25).** Using a procedure similar to that described for the preparation of **24** from **22**, treatment of **23** (49 mg, 0.0568 mmol) gave **25** as a colorless powder (34 mg, 74% yield): $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 1.17 (m, 2H), 1.38–1.66 (m, 5H), 1.70–1.98 (m, 6H), 2.20–2.52 (m, 4H), 2.84–3.34 (m, 7H), 4.25 (m, 1H), 6.66 (s, 1H), 7.06 (m, 1H), 7.30–7.54 (m, 7H), 7.74 (m, 1H), 7.82 (d, $J$=7.3 Hz, 1H), 7.86–7.96 (m, 2H), 8.09 (m, 1H), 8.26 (br, 1H). FABMS $m/z$ 748 [$(M-H)^-$].

## 4. Supplementary Material

Single crystal X-ray crystallographic data for salt **13**, including a thermal ellipsoid plot at the 50% confidence interval and tables of atomic coordinates and parameters are provided (10 pages). Supplementary material can be found in the online version of this paper.

## Acknowledgements

Appreciation is expressed to Dr. James Kelley of the LMC for mass spectral analysis. Gratitude is also expressed to the Japan Society for the Promotion of Science for Research Abroad for Postdoctoral Fellowship funding of S.O. Work was supported in part by the Office of Naval Research (ONR) and the National Institute on Drug Abuse (NIDA) (for J.R.D.) and by the Susan G. Komen Breast Cancer Foundation (for M.Z. and D.Y.).

## References and notes

1. Mutter, M. *Angew. Chem. Int. Ed. Engl.* **1985**, *24*, 639–653.
2. Babine, R. E.; Bender, S. L. *Chem. Rev.* **1997**, *97*, 1359–1472.
3. Ripka, A. S.; Rich, D. H. *Curr. Opin. Chem. Biol.* **1998**, *2*, 441–452.
4. Hruby, V. J.; Balse, P. M. *Curr. Med. Chem.* **2000**, *7*, 945–970.
5. Bursavich, M. G.; Rich, D. H. *J. Med. Chem.* **2002**, *45*, 541–558.
6. Freidinger, R. M. *J. Med. Chem.* **2003**, *46*, 5553–5566.
7. Gibson, S. E.; Guillo, N.; Tozer, M. J. *Tetrahedron* **1999**, *55*, 585–615.
8. Prasad, B. V. V.; Balaram, P. *CRC Crit. Rev. Biochem.* **1984**, *14*, 307–348.
9. Rastogi, S. N.; Bindra, J. S.; Anand, N. *Ind. J. Chem.* **1971**, *9*, 1175–1182.
10. Obrecht, D.; Altorfer, M.; Bohdal, U.; Daly, J.; Huber, W.; Labhardt, A.; Lehmann, C.; Muller, K.; Ruffieux, R.; Schonholzer, P.; Spiegler, C.; Zumbrunn, C. *Biopolymers* **1997**, *42*, 575–626.
11. Tsatsanis, C.; Spandidos, D. A. *Int. J. Mol. Med.* **2000**, *5*, 583–590.
12. Renhowe, P. A. *Ann. Rep. Med. Chem.* **2001**, *36*, 109–118.
13. Kzardenings, A. K.; Gordeev, M. F.; Patel, D. V. *Tetrahedron Lett.* **1996**, *37*, 3635–3638.
14. Liu, W. Q.; Vidal, M.; Gresh, N.; Rogues, B. P.; Garbay, C. *J. Med. Chem.* **1999**, *42*, 3737–3741.
15. Petrakis, K. S.; Nagabhushan, T. L. *J. Am. Chem. Soc.* **1987**, *109*, 2831–2833.
16. Bayle-Lacoste, M.; Moulines, J.; Collignon, N.; Boumekouez, A.; De Tinguy-Moreaud, E.; Neuzil, E. *Tetrahedron* **1990**, *46*, 7793–7802.
17. Garbay, C.; Liu, W.-Q.; Vidal, M.; Roques, B. P. Preparation of pseudopeptides having an inhibiting activity with respect to

paths activated by proteins with active tyrosine kinase activity and pharmaceutical compositions containing same. US Pat. Appl. Publ. No. US 2002/0055463 A1 (May 9, 2002) for US Appl. No. 09/977,349, filed October 16, 2001. Priority Fr 98/16459 (December 24, 1998).
18. Stankovic, C. J.; Surendran, N.; Lunney, E. A.; Plummer, M. S.; Para, K. S.; Shahripour, A.; Fergus, J. H.; Marks, J. S.; Herrera, R.; Hubbell, S. E.; Humblet, C.; Saltiel, A. R.; Stewart, B. H.; Sawyer, T. K. *Bioorg. Med. Chem. Lett.* **1997**, *7*, 1909–1914.
19. Liu, W. Q.; Carreaux, F.; Meudal, H.; Roques, B. P.; GarbayJaureguiberry, C. *Tetrahedron* **1996**, *52*, 4411–4422.
20. Liu, D.-G.; Wang, X.-Z.; Gao, Y.; Li, B.; Yang, D.; Burke, T. R., Jr. *Tetrahedron* **2002**, *58*, 10423–10428.
21. Williams, R. M.; Im, M. N. *J. Am. Chem. Soc.* **1991**, *113*, 9276–9286.
22. Compound **7** was diastereomerically pure to H NMR.
23. Ishizumi, K.; Ohashi, N.; Tanno, N. *J. Org. Chem.* **1987**, *52*, 4477–4485.
24. Obrecht, D.; Spiegler, C.; Schoenholzer, P.; Mueller, K.; Heimgartner, H.; Stierli, F. *Helv. Chim. Acta* **1992**, *75*, 1666–1696.
25. Yao, Z. J.; King, C. R.; Cao, T.; Kelley, J.; Milne, G. W. A.; Voigt, J. H.; Burke, T. R. *J. Med. Chem.* **1999**, *42*, 25–35.
26. Carpino, L. A.; El-Faham, A. *J. Am. Chem. Soc.* **1995**, *117*, 5401–5402.
27. Gao, Y.; Luo, J.; Yao, Z.-J.; Guo, R.; Zou, H.; Kelley, J.; Voigt, J. H.; Yang, D.; Burke, T. R., Jr. *J. Med. Chem.* **2000**, *43*, 911–920.
28. Davidson, J. P.; Lubman, O.; Rose, T.; Waksman, G.; Martin, S. F. *J. Am. Chem. Soc.* **2002**, *124*, 205–215.