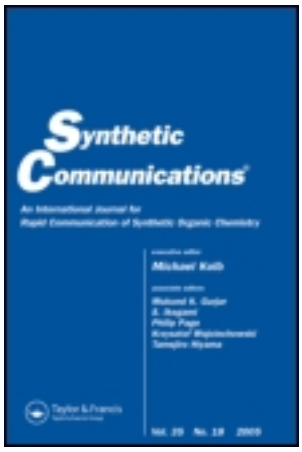


Synthetic Communications: An International Journal for Rapid Communication of Synthetic Organic Chemistry

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/lsyc20

SYNTHESIS OF (S)-(+)-2-AMINO-6-(AMINOOXY)-HEXANOIC ACID

Maciej Adamczyk [a] & Rajarathnam E. Reddy [a]

[a] Department of Chemistry (9NM, Bldg AP20-2), Diagnostics Division, Abbott Laboratories, 100 Abbott Park Road, Abbott Park, IL, 60064-6016, U.S.A.

Version of record first published: 16 Aug 2006.

To cite this article: Maciej Adamczyk & Rajarathnam E. Reddy (2001): SYNTHESIS OF (S)-(+)-2-AMINO-6-(AMINOOXY)-HEXANOIC ACID, Synthetic Communications: An International Journal for Rapid Communication of Synthetic Organic Chemistry, 31:4, 579-586

To link to this article: http://dx.doi.org/10.1081/SCC-100000585

SYNTHETIC COMMUNICATIONS, 31(4), 579–586 (2001)

SYNTHESIS OF (*S*)-(+)-2-AMINO-6-(AMINOOXY)-HEXANOIC ACID

Maciej Adamczyk* and Rajarathnam E. Reddy

Department of Chemistry (9NM, Bldg AP20-2), Diagnostics Division, Abbott Laboratories, 100 Abbott Park Road, Abbott Park, IL 60064-6016, USA

ABSTRACT

(*S*)-(+)-2-Amino-6-(aminooxy)hexanoic acid (AAHA, **3**), a non-proteinogenic amino acid, and its derivative, (*S*)-(−)-6-{[(*tert*-butoxycarbonyl)amino]oxy}-2-{[(9H-fluoren-9-ylmethoxy)-carbonyl]amino}hexanoic acid (**4**), were synthesized from (*S*)-(−)-6-amino-2-{[(benzyloxy)carbonyl] amino}hexanoic acid (**5**) in good overall yield.

Synthesis of non-proteinogenic amino acids is of considerable interest currently because of their importance in a variety of medicinal and biotechnological applications (1–5). The aminooxy group ($-ONH_2$) exhibits greater nucleophilicity than the corresponding primary amino group, and the resulting *O*-alkyl oximes, which are formed by reaction of aminooxy group with carbonyl compounds, are more stable than the imines (6,7). Thus, proteins (8,9), oligonucleotides (10,11), and hapten-label conjugates (12,13) have been chemoselectively ligated through

*Corresponding author. E-mail: maciej.adamczyk@abbott.com

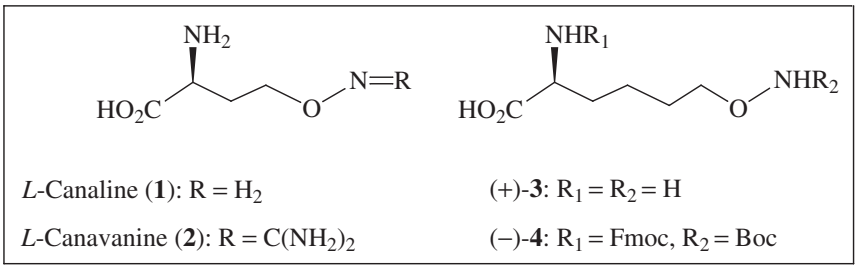
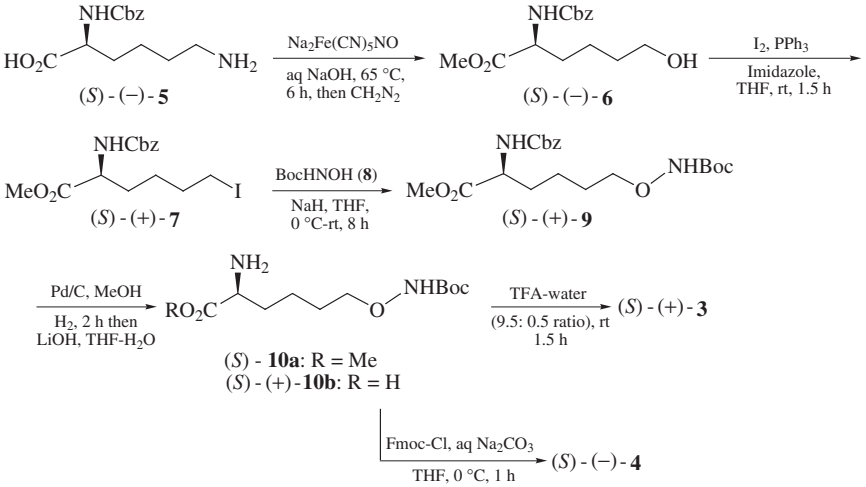


Figure.

the oxime bond from aminooxy and carbonyl functional groups, thereby avoiding the activation of carboxylic acid and side-chain protection. The aminooxy group is also present in natural products, e.g., *L*-canaline (**1**) (Figure), which was isolated from leguminous plants as a degradation product of *L*-canavanine (**2**) exhibiting insecticidal and antitumor activities (14,15). Recently, for our program on the synthesis of novel peptidimemtics, we needed an amino acid building block, *L*-lysine derivative with aminooxy functionality. In this paper, we describe the synthesis of (*S*)-(+)-2-amino-6-(aminooxy)hexanoic acid (AAHA, **3**) and its derivative, (*S*)-(−)-6-{[(*tert*-butoxycarbonyl)amino]oxy}-2-{[(*9H*-fluoren-9-ylmethoxy)carbonyl]amino} hexanoic acid (**4**) from (*S*)-(−)-6-amino-2-{[(benzyloxy)carbonyl]amino}hexanoic acid (**5**).

The strategy for the synthesis of (*S*)-(+)-AAHA (**3**), which is depicted in Scheme 1, involves the utilization of a commercially available *L*-lysine derivative, (*S*)-(−)-6-amino-2-[(benzyloxy)carbonyl]amino}hexanoic acid (**5**), in which the ε-amine could be transformed to the desired aminooxy functional group. Thus, (*S*)-(−)-**5** was first heated with sodium nitroprusside (16) in water at pH 9.5 and the resulting crude hydroxy acid [(*S*)-**6a**] was treated with ethereal-diazomethane in ethyl acetate to form (*S*)-(−)-**6b** in an improved yield (43%) for two steps on a 15-g scale. The hydroxy functionality in (*S*)-(+)-**6b** was then transformed to the corresponding iodide (*S*)-(+)-**7** in excellent yield (84%) by treatment with iodine, triphenylphosphine, and imidazole in THF (17). The aminooxy functionality was introduced via alkylation of *tert*-butyl−*N*-hydroxycarbamate (**8**) with iodide (*S*)-(−)-**7** using sodium hydride in THF. Purification of the crude product by preparative HPLC afforded (*S*)-(+)-**9** in 76% yield. The Cbz group in (*S*)-(+)-**9** was removed by hydrogenolysis and the crude amine (*S*)-**10a** was treated with lithium hydroxide in THF-water to hydrolyze the methyl ester. The crude compound was purified by preparative HPLC to give the acid (*S*)-(+)-**10b** in 86% yield. Finally, the Boc group in (*S*)-(+)-**10b** was hydrolyzed by treatment with TFA-water and purified the crude product by preparative HPLC to afford

MARCEL DEKKER, INC.
270 Madison Avenue, New York, New York 10016



Scheme 1.

(*S*)-(+)-2-amino-6-(aminooxy)hexanoic acid (AAHA, **3**) in good (83%) yield as its TFA salt. The Fmoc derivative, (*S*)-(−)-6-{[(*tert*-butoxycarbonyl)-amino]oxy}-2-{[(9H-fluoren-9-ylmethoxy)carbonyl]-amino}hexanoic acid (**4**), which is needed for the solid phase peptide synthesis, was also prepared from (*S*)-(+)-**10b** by treatment with Fmoc-Cl in aq sodium carbonate-THF in 88% yield after purification by HPLC.

In summary, (*S*)-(−)-amino-6-(aminooxy)hexanoic acid (AAHA, **3**) and its derivative, (*S*)-(−)-6-{[(*tert*-butoxycarbonyl)amino]oxy}-2-{[(9*H*-fluoren-9-yl methoxy)carbonyl]amino}hexanoic acid (**4**), were synthesized from (*S*)-(−)-6-amino-2-{[(benzyloxy)carbonyl]amino}hexanoic acid (**5**) in good overall yield.

EXPERIMENTAL

General Methods and Materials

^{1}H and ^{13}C NMR spectra were recorded on a Varian Gemini spectrometer (300 MHz) and the chemical shifts (δ) were reported in ppm relative to TMS and coupling constants (J) were reported in Hz. Electrospray ionization mass spectrometry (ESI-MS) was carried out on a Perkin-Elmer (Norwalk, CT) Sciex API 100 Benchtop system employing Turbo Ionspray ion source, and HRMS were

obtained on a Nermang 3010 MS-50, JEOL SX102-A mass spectrometer. THF was freshly distilled from a purple solution of sodium and benzophenone. (*S*)-(−)-6-Amino-2-{[(benzyloxy)carbonyl]amino}-hexanoic acid (**5**) was purchased from Novabiochem (San Diego, CA) and all other reagents were from Aldrich Chemical Co. (Milwaukee, WI) or Sigma Chemical Co. (St. Louis, MO) and used without purification. All the solvents employed were of HPLC grade purchased from EM Science (Gibbstown, NJ) and used as received. Analytical reversed phase (RP) HPLC was performed using a Waters, RCM, C18, Symmetry, 7.0 μm (8 × 100 mm) (solvents ratio v/v reported). Preparative reversed phase (RP) HPLC was performed using Waters RCM, C18, Symmetry, 7.0μm (40 × 100 mm), (solvents ratio v/v reported). Optical rotations were measured on Autopol III polarimeter from Rudolph Research (Flanders, NJ).

(*S*)-(−)-Methyl-2-{[(benzyloxy)carbonyl]amino}-6-hydroxyhexanoate (6b)

(*S*)-(−)-6-Amino-2-{[(benzyloxy)carbonyl]amino}-hexanoic acid (**5**, 15.75 g, 56.19 mmol) was dissolved in water (225 mL) and the pH adjusted to 9.5 using 4M aq. NaOH. The mixture was heated to 60–65°C in an oil bath. Sodium nitroprusside (30.0 g, 100.67 mmol, 1.8 equiv.) was added portionwise over a 35-min period while maintaining the pH of the reaction mixture between 9–10 using 4N NaOH and a pH meter. The resulting brown mixture was heated for an additional 6 h while maintaining the pH between 9–10 with occasional addition of 4M aq. NaOH. The reaction mixture was cooled to room temperature and filtered through celite powder (5 mm bed). The pH of the filtrate was adjusted to 1.5 using 6M HCl and extracted with EtOAc (3 × 250 mL). The combined organic layers were washed with brine (100 mL), dried (Na_2SO_4), and concentrated on a rotary evaporator. The resulting crude hydroxy-acid (*S*)-**6a** (11.8 g) was dissolved in EtOAc (55 mL) and cooled with an ice bath. A freshly generated ethereal-diazomethane solution [100 mL, generated from *N*-nitroso-*N*-methylurea (17.36 g, 168.59 mmol, 3.0 equiv.), KOH (18.87 g, 337.0 mmol, 6.0 equiv.), ether (100 mL) and water (80 mL)] was added at 0–5°C and allowed the mixture to warm to room temperature. The mixture was then stirred for 18 h, and carefully concentrated on a rotary evaporator below 30°C bath temperature. The crude product was purified by silica gel column chromatography (60% EtOAc in hexanes) to afford 7.1 g of (*S*)-(−)-**6b** in 43% yield for two steps as a colorless viscous oil. R_f: 0.37 (60% EtOAc in hexanes); Analytical RP HPLC: MeCN:0.1% aq trifluoroacetic acid/50:50, 2.0 mL/min at 225 nm, t_R: 2.62 min, 97%; ESI-MS (m/z): 296 $(M + H)^+$, 313 $(M + NH_4)^+$, 318 $(M + Na)^+$, 613 $(2 \times M + Na)^+$ and identical in other physical properties to the reported values (16).

MARCEL DEKKER, INC.
270 Madison Avenue, New York, New York 10016

(*S*)-(+)-Methyl-2-{[(benzyloxy)carbonyl]amino}-6-iodohexanoate (7)

Triphenylphosphine (5.895 g, 22.5 mmol, 1.5 equiv.), imidazole (1.63 g, 24.0 mmol, 1.6 equiv.) and iodine (5.69 g, 22.5 mmol, 1.5 equiv.) were added sequentially to a solution of (*S*)-(−)-**6b** (4.44 g, 15.0 mmol) dissolved in THF (150 mL) at room temperature under nitrogen. After stirring the mixture for 1.5 h, the solvent was removed on a rotary evaporator to dryness and the crude product was purified by silica gel column chromatography (20–25% EtOAc in hexanes) to afford 5.08 g of iodide (*S*)-(+)-**7** in 84% yield as colorless thick oil. R_f: 0.43 (25% EtOAc in hexanes); Analyticl RP HPLC: MeCN:0.1% aq trifluoroacetic acid/50:50, 2.0 mL/min at 225 nm, t_R: 16.34 min, >99%; $[\alpha]^{23}_D$ + 14.2 (c 1.31, $CHCl_3$); ^{1}H NMR ($CDCl_3$): δ 7.38–7.32 (m, 5 H), 5.29 (d, 1 H, J = 7.5 Hz), 5.12 (s, 2 H), 4.40 (q, 1 H, J = 12.6, 7.2 Hz), 3.76 (s, 3 H), 3.16 (t, 2 H, J = 6.9 Hz), 1.92–1.78 (m, 3 H), 1.75–1.60 (m, 1 H), 1.54–1.40 (m, 2 H); ^{13}C NMR ($CDCl_3$): δ 172.7, 155.8, 136.2, 128.6, 128.2, 128.1, 67.1, 53.6, 52.5, 32.7, 31.6, 26.0, 6.1; ESI-MS (m/z): 406 $(M + H)^+$, 423 $(M + NH_4)^+$, 428 $(M + Na)^+$, 833 $(2 \times M + Na)^+$; HRMS (FAB, m/z): calcd. for $C_{15}H_{21}NO_4I$, 406.0515 $(M + H)^+$; observed, 406.0509.

(*S*)-(+)-Methyl-2-{[(benzyloxy)carbonyl]amino}-6-{[*tert*-butoxy carbonyl)-amino]oxy}hexanoate (9)

NaH (dry 95%, 0.598 g, 24.94 mmol, 2.0 equiv.) was added in three portions to a 0°C cooled solution of *tert*-butyl-*N*-hydroxycarbamate (**8**, 4.14 g, 31.17 g, 2.5 equiv.) dissolved in THF (100 mL) under nitrogen. After stirring the mixture for 30 min, a solution of (*S*)-(+)-**7** (5.02 g, 12.47 mmol) dissolved in THF (80 mL) was added via double-ended needle at 0°C over a 5-min period. After stirring the mixture for 3 h, the cooling bath was removed and the mixture allowed to warm to room temperature. Stirring was continued for an additional 12 h. The mixture was diluted with EtOAc (1 L) and washed with brine (3 × 100 mL), dried ($MgSO_4$), and concentrated on a rotary evaporator. The crude compound was dissolved in MeCH-0.1% aq trifluoroacetic acid (100 mL, 70:30 ratio) and purified by preparative RP HPLC (MeCN:0.1% aq trifluoroacetic acid/50:50, 45.0 mL/min at 225). The product was lyophilized to afford 3.71 g of (*S*)-(+)-**9** in 73% yield as a colorless viscous oil. Analytical RP HPLC: MeCN:aq trifluoroacetic acid/50:50, 2.0 mL/min at 225, t_R: 9.61 min, >99%; $[\alpha]^{23}_D$ + 1.54 (c 0.78, $CHCl_3$); ^{1}H NMR ($CDCl_3$): δ 7.38–7.32 (m, 5 H), 5.41 (d, 1 H, J = 8.4 Hz), 5.11 (s, 2 H), 4.41–4.36 (m, 1 H), 3.83 (t, 2 H, J = 6.3 Hz), 3.75 (s, 3 H), 3.02 (br s, 1 H), 1.92–1.40 (m, 6 H), 1.47 (s, 9 H); ^{13}C NMR ($CDCl_3$): δ 172.9, 157.0, 156.0, 136.2, 128.5, 128.2, 128.1, 81.8, 76.1, 67.0, 53.5, 52.4, 32.3, 28.2, 27.3, 21.6; ESI-MS (m/z): 411

MARCEL DEKKER, INC.
270 Madison Avenue, New York, New York 10016

$(M + H)^+$; 428 $(M + NH_4)^+$, 433 $(M + Na)^+$, 838 $(2 \times M + NH_4)^+$, 843, $(2 \times M + Na)^+$; HRMS (FAB, m/z): calcd. for $C_{20}H_{31}N_2O_7$, 411.2131 $(M + H)^+$; observed, 411.2142.

(*S*)-(+)-2-Amino-6-{[(*tert*-butoxycarbonyl)amino]oxy}hexanoic Acid (10b)

Ten percent Pd/C [(wet, Deguassa type E101 NE/W), 0.371 g] was added to a solution of (*S*)-(+)-**9** (3.71 g, 9.05 mmol) dissolved in methanol and stirred under H_2 atmosphere using a balloon at room temperature. after 1.25 h, the mixture was filtered through celite powder (5 mm bed), washed with methanol (20 mL), and the filtrate was concentrated on a rotary evaporator. The resulting crude amino-ester (**10a**, 2.62 g) was dissolved in THF (82 mL). To this mixture, LiOH (monohydrate, 0.760 g, 18.1 mmol, 2.0 equiv.) and water (28 mL) were added sequentially at room temperature and stirred for 1.25 h. Most of the THF from the reaction mixture was removed on a rotary evaporator, diluted with water (25 mL), and adjusted the pH to 4.0 using 6N HCl. The mixture was diluted with MeCN (25 mL) and purified by preparative RP HPLC (MeCN:0.1% aq trifluoroacetic acid/20:80, 45.0 mL/min at 225). Lyophilization of the product gave 2.53 g of (*S*)-(+)-**10b**-TFA salt in 74% yield as a colorless glassy material. Analytical RP HPLC: MeCN:aq trifluoroacetic acid/20:80, 2.0 mL/min at 225 nm; t_R: 3.4 min, 98%; $[\alpha]^{23}{}_D + 7.4$ (c 1.12, MeOH); ^{1}H NMR (CD_3OD): δ 3.91 (dd, 1 H, $J = 7.2$, 6.0 Hz), 3.80 (t, 2 H, $J = 6.0$ Hz), 2.04–1.82 (m, 2 H), 1.72–1.50 (m, 4 H), 1.46 (s, 9 H); ^{13}C NMR ($CDCl_3$): δ 172.2, 159.2, 82.1, 76.8, 54.1, 31.3, 28.6, 28.5, 22.6; ESI-MS (m/z): 263 $(M + H)^+$, 525 $(2 \times M + H)^+$; HRMS (FAB, m/z): calcd for $C_{11}H_{23}N_2O_5$, 263.1607 $(M + H)^+$; observed, 263.1604; Anal calcd. for $C_{11}H_{23}F_3N_2O_7$: C, 41.49; H, 6.16; F, 15.15; N, 7.44; Found: C, 41.39; H, 6.17; F, 15.36; N, 7.35.

(*S*)-(+)-2-Amino-6-(aminooxy)hexanoic Acid (3)

Trifluoroacetic acid (19 mL) and water (1.0 mL) were added to (*S*)-(+)-**10b**-TFA salt (0.094 g, 0.25 mmol) at room temperature and stirred for 1.5 h. The mixture was concentrated on a rotary evaporator, dissolved in 0.1% aq trifluoroacetic acid (10.0 mL), and purified by preparative RP HPLC (MeCN:0.1% aq trifluoroacetic acid/5:95, 25.0 mL/min at 225 nm). Lyophilization of the product gave 0.081 g of (*S*)-(+)-**3** as its TFA salt in 83% yield as a colorless gummy material. Analytical RP HPLC: MeCN:0.5 M triethylammonium acetate/5:95, 2.0 mL/min at 225 nm; t_R: 2.42 min, 97%; $[\alpha]^{23}{}_D + 11.7$ (c 0.58, MeOH); ^{1}H NMR (CD_3OD): δ 4.05 (t, 3 H, $J = 6.0$ Hz), 4.00–3.93 (m, 1 H), 2.02–1.82 (m, 2 H), 1.81–1.44 (m, 4 H), ^{1}H NMR (CD_3CN + 5 drops of CF_3CO_2D): δ 4.05 (t, 2 H, $J = 6.0$ Hz), 3.98 (t, 1 H, $J = 6.3$ Hz), 1.96–1.80 (m, 2 H), 1.71–1.62

MARCEL DEKKER, INC.
270 Madison Avenue, New York, New York 10016

(m, 2 H), 1.59–1.34 (m, 2 H); ^{13}C NMR (CD_3CN + 5 drops of (CF_3CO_2D). δ 170.2, 76.2, 54.0, 30.0, 27.6, 21.8; ESI-MS (m/z): 163 $(M + H)^+$, 185 $(M + Na)^+$, 325 $(M + H)^+$ HRMS (FAB, m/z): calcd for $C_6H_{14}N_2O_3$, 163.1083 $(M + H)^+$; observed, 163.1086.

(*S*)-(−)-6-{[(*tert*-Butoxycarbonyl)amino]oxy}-2-{[(*9H*-fluoren-9-yl methoxy)-carbonyl]amino}hexanoic Acid (4)

Aqueous Na_2CO_3 [(0.334 g, 3.15 mmol, 3.0 equiv., dissolved in water (6 mL)] was added to a solution of (*S*)-(+)-**10b**-TFA salt (0.393 g, 1.05 mmol) dissolved in THF (6 mL) and the mixture was cooled to 0°C. 9-Fluorenylmethyl chloroformate (Fmoc-Cl, 0.299 g, 1.16 g, 1.1 equiv.) was added in two portions over a 5-min period and the mixture stirred for 1 h. The mixture was diluted with water (15 mL), adjusted the pH to 4.0 using 0.1N HCl and purified by preparative HPLC (MeCN:0.1% aq trifluoroacetic acid/65:35, 45.0 mL/min at 225 nm). Lyophilization of the product gave 0.443 g of (*S*)-(−)-**4** in 88% yield as white powder. Analytical RP HPLC: MeCN:0.1% aq trifluoroacetic acid/65:35, 2.0 mL/min at 225 nm; t_R: 4.02 min, >99%; $[\alpha]^{23}_D$ − 3.2 (c 0.66, MeOH); ^{1}H NMR (CD_3OD): δ 7.78 (d, 2 H, J = 7.5 Hz), 7.67 (t, 2 H, J = 6.6 Hz), 7.41–7.27 (m, 4 H), 4.42–4.30 (m, 2 H), 4.20 (t, 1 H, J = 6.9 Hz), 4.15–4.11 (m, 1 H), 3.78 (t, 2 H, J = 6.3 Hz), 1.92–1.42 (m, 6 H), 1.45 (s, 9 H); ^{13}C NMR (CD_3OD): δ 175.9, 159.1, 158.8, 145.4, 145.2, 142.6, 128.8, 128.2, 126.3, 120.9, 81.9, 77.3, 68.0, 55.3, 32.4, 28.6, 27.7, 23.5; ESI-MS (m/z): 485 $(M + H)^+$, 502 $(M + NH_4)^+$, 986 $(2 \times M + NH_4)^+$; HRMS (FAB, m/z): calcd for $C_{26}H_{32}N_2O_7$, 485.2288 $(M + H)^+$; observed, 485.2282; Anal calcd. for $C_{26}H_{34}N_2O_8$ (monohydrate): C, 62.14; H, 6.82; 5.57; Found: C, 62.62; H, 6.10; N, 5.89.

REFERENCES

1. Williams, R.M. In *Organic Chemistry Series*; Baldwin, J.E.; Magnus, P.D., Eds.; Pergman Press: Oxford, 1989; Vol. 1.
2. Duthaler, O.R. Tetrahedron **1994**, *50*, 1539–1650.
3. Robi, J.A. Tetrahedron Lett. **1994**, *35*, 393–397.
4. Adamczyk, M.; Reddy, R.E. Tetrahedron Asymmetry **1999**, *9*, 3919–3921.
5. Adamczyk, M.; Reddy, R.E.; Rege, S.D. Synthetic Commun. **2000**, *30*, 1389–1400.
6. Jencks, W.P. Prog. Phys. Org. Chem. **1964**, *2*, 63–128.
7. Isaacs, N. *Physical Organic Chemistry*, 2nd Ed.; Longman: Essex; 276–267.
8. Waker, M.A. Angew. Chem. Int. Ed. Engl. **1997**, *36*, 1069–1071.

MARCEL DEKKER, INC.
270 Madison Avenue, New York, New York 10016

9. Tam, J.P.; Spetzler, J.C. Biomedica Peptides, Proteins and Nucleic Acids **1995**, *1*, 123–132.
10. Salo, H.; Virta, P.; Hakala, H.; Prakash, T.P.; Kawasaki, A.M.; Manoharan, M.; Lonnberg, H. Bioconjugate Chem. **1999**, *10*, 815.
11. Boturyn, D.; Boudali, A.; Constant, J-F., Defrancq, E.; Lhomme, J. Tetrahedron **1997**, *53*, 5485–5492.
12. Adamczyk, M.; Chen, Y-Y.; Grote, J.; Mattingly, P.G. Steroids **1999**, *64*, 283–290.
13. Adamczyk, M.; Mattingly, P.G.; Moore, J.A.; Pan, Y. Org. Lett. **1999**, *1*, 779–781.
14. Rosenthal, G.A. Life Sci. **1978**, *23*,93–98.
15. Na Phuket, S.R.; Trifonov, L.S.; Crooks, P.A.; Rosenthal, G.A.; Freeman, J.W.; Strodel, W.E. Drug Development. Res. **1997**, *40*, 325–332.
16. Bambal, R.; Hanzlik, R.P. J. Org. Chem. **1994**, *59*, 729–732.
17. Adamczyk, M.; Johnson, D.D.; Reddy, R.E. Tetrahedron **1999**, *55*, 63–88.

Received in the USA May 4, 2000

MARCEL DEKKER, INC.
270 Madison Avenue, New York, New York 10016