Derivatives of L-pyrimidylalanine

Gury Zvilichovsky* and Vadim Gurvich

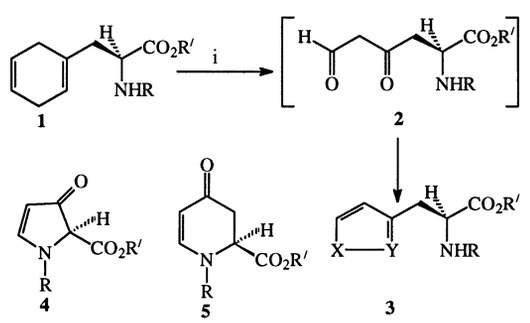
Department of Organic Chemistry, The Hebrew University of Jerusalem, Jerusalem 91904, Israel

1 PERKIN

Optically active derivatives of L-isoxazolo[2,3-*a*]pyrimidin-4-ylalanine are prepared by the combination of Birch reduction of the phenyl ring of L-phenylalanine, followed by ozonolysis and condensation with 3-amino-5-oxo-4-phenyl-2,5-dihydroisoxazole. Ring cleavage of the five-membered ring by reduction, oxidation and nucleophiles gives pyrimidylalanine derivatives.

Introduction

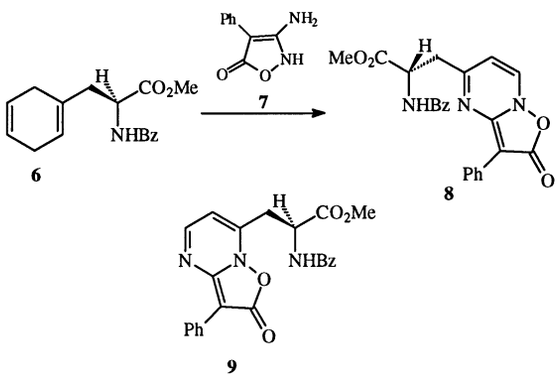
Amino acids are enjoying unprecedented renaissance in virtually all disciplines. Many nonproteinogenic α-amino acids have been found to have biological and pharmacological activities[1–9] and are also incorporated in semisynthetic penicillins,[10] cephalosporins and biologically active peptides.[11,12] Several pyrimidylalanine derivatives occur in plants and possess antitumour activity by virtue of their ability to inhibit an enzyme system in the biosynthesis of purines.[13–16] DL-2-, 4- and 5-pyrimidylalanines were prepared by Haggerty in 1965 from the corresponding methylpyrimidine derivatives.[17] An approach to the transformation of L-phenylalanine into other optically active amino acids with the retention of optical activity was shown recently.[18,19] It consists of the combination of Birch reduction of the phenyl ring, ozonolysis of the resulting hexadienyl derivative **1** to yield the dicarbonyl intermediate **2**, and condensation of this with dinucleophiles to give heterocyclic amino acid derivatives **3**. In the case of phenylalanine as the starting material it was shown that the chiral centre was not affected. It was described as a one-pot reaction, mainly because of the tendency of the dicarbonyl intermediate to undergo cyclization to a cyclic derivative **5** when isolation was attempted.[20] In the case of the conversion of phenylglycine derivatives the cyclization product which was isolated was the pyrrole derivative **4**.[18]



Scheme 1 *Reagent:* i, O_3

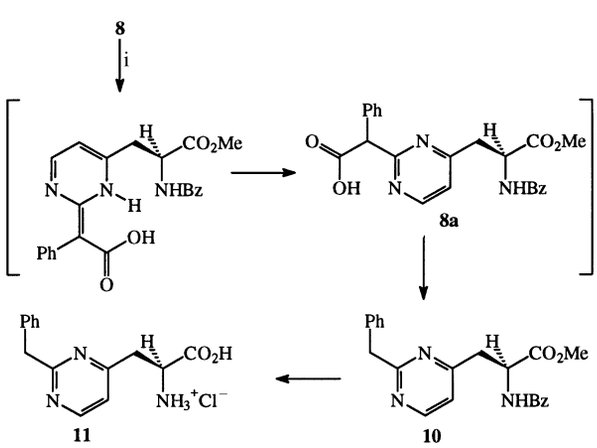
Results and discussion

In the present work 3-aminoisoxazole derivative **7** was introduced as a dinucleophile into the ozonolysis mixture of the ester of the protected cyclohexadienylalanine **6**, resulting in the bicyclic system **8**. This condensation proceeds under acid catalysis. It was assumed that the aminopropionic chain is linked to the pyrimidine ring at position 5 rather than at position 7, excluding the regioisomer **9**. The regioselectivity of this condensation was demonstrated earlier, where the structure was confirmed by single-crystal X-ray analysis.[21] The NMR spectra of the product which was obtained here were compatible with structure **8** rather than structure **9**. The orange–yellow crystalline isoxazolo[2,3-*a*]pyrimidylalanine derivative **8** is stable on the shelf, provided it is protected from light. Upon reduction of the N–O bond in the bicyclic system with zinc in acetic acid the isoxazole ring is cleaved. The intermediate acid **8a** which is formed is unstable and undergoes spontaneous decarboxylation, yielding the L-(2-benzylpyrimidin-4-yl)alanine derivative **10**. Removal of both the benzoyl and ester groups from compound **10** was achieved by acid hydrolysis, and gave the unprotected amino acid **11**.

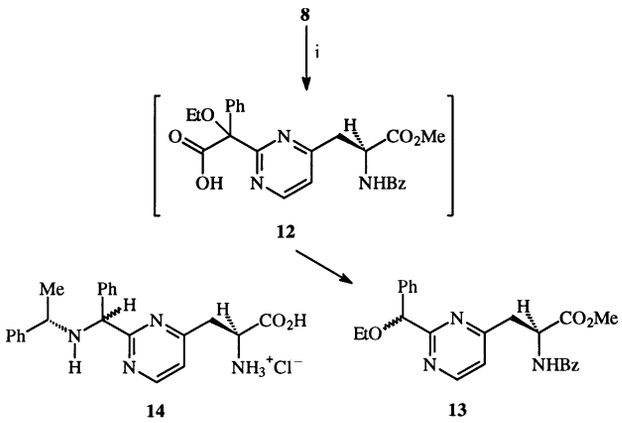


Scheme 2

Heating of the isoxazolopyrimidylalanine derivative **8** in ethanol gave the benzyl ether derivative **13**. The mechanism for these rearrangements was described earlier.[6] The nucleophilic ethanol opens the five-membered ring to yield an unstable carboxylic acid derivative **12** which undergoes spontaneous decarboxylation to yield product **13**. This ether was obtained as a 5:4 diasteroisomeric mixture.



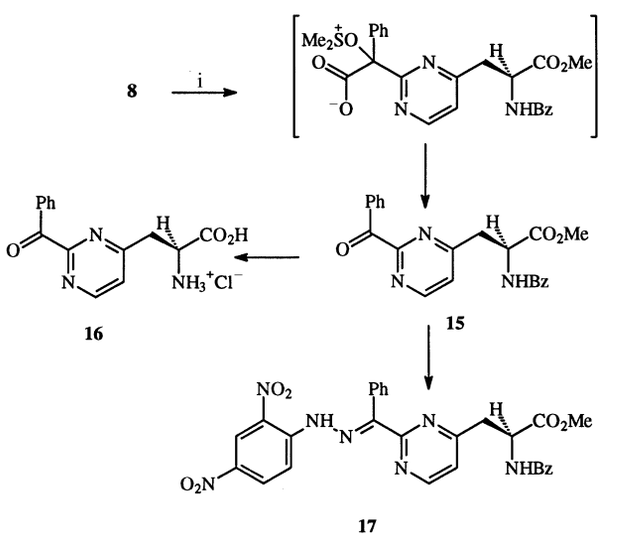
Scheme 3 *Reagent:* i, [H]



Scheme 4 *Reagent:* i, EtOH

It was also possible to open the isoxazole ring by using a primary amine. The amine which was used was the optically active (*R*)-(+)-α-aminoethylbenzene. The resulting diamino acid derivative **14** was obtained as a 1 : 1 mixture of two diastereoisomers which could be separated by chromatography.

Oxidative cleavage of the isoxazole ring by heating of the isoxazolone **8** in dimethyl sulfoxide (DMSO)[22] afforded the corresponding ketoamino acid derivative **15**. Removal of both of



Scheme 5 *Reagent:* i, DMSO

the benzoyl and the ester protecting groups was achieved by hydrochloric acid, and yielded the free hydrochloride **16**. Condensation of the keto ester **15** with 2,4-dinitrophenylhydrazine gave the hydrazone **17**.

Experimental

General methods

Mps were taken in a Thomas Hoover instrument. NMR Spectra were taken with Bruker WP-200 and Bruker AMX-300 spectrometers. *J* Values are given in Hz. Optical rotations were measured by a Perkin-Elmer 141 polarimeter, and $[\alpha]_D$-values are given in units of 10^{-1} deg cm^2 g^{-1}. Chromatographic separation was carried out with silica gel (230–400 mesh) in a 450 × 10 mm column. Light petroleum refers to the fraction with distillation range 40–60 °C.

Methyl *N*-benzoyl-3-(2-oxo-3-phenyl-2*H*-isoxazolo[2,3-*a*]-pyrimidin-5-yl)alaninate 8

Methyl L-*N*-benzoyl-3-(cyclohexa-1,4-dienyl)alaninate **6** (0.5 g) in 10 cm^3 of dichloromethane was added to a saturated solution of ozone in dichloromethane (15 cm^3) at −78 °C, buffered with 0.2 g of $NaHCO_3$. More ozone was added until the blue colour persisted. The mixture was purged with nitrogen, dimethyl sulfide (5 cm^3) was added, and the mixture was allowed to warm to room temperature overnight. The solution was filtered and the solvent was removed under reduced pressure. The residue was dissolved in ethanol (10 cm^3) and 3-amino-4-phenylisoxazol-5(2*H*)-one **7** (1.0 g) and 10 M ethanolic HCl (1 cm^3) were added. The solution was refluxed for 10 min under nitrogen with protection from light. The solvent was removed under reduced pressure and the crude product was extracted with ethyl acetate and chromatographed with a solvent gradient: ethyl acetate–light petroleum (1 : 3–2 : 1) to yield compound **8** as orange-yellow crystals (0.25 g, 52%), mp 146–147 °C (decomp.) (Found: C, 65.99; H, 4.32; N, 10.16. Calc. for $C_{23}H_{19}N_3O_5$: C, 66.18; H, 4.59; N, 10.07%); $[\alpha]_D^{25}$ −5 (*c* 1, EtOH); δ_H([$^2H_6$]DMSO) 9.08 (d, *J* 7.2, 1 H, pyr-CH), 8.94 (d, $J_{NH,\alpha}$ 7.7, 1 H, NH), 8.15 (d, *J* 8.4, 2 H, *o*-Ph), 7.83 (d, $J_{o,m}$ 7.2, 2 H, *o*-PhCO), 7.28–7.56 (m, 5 H, *m*-, *p*-PhCO + *m*-Ph), 7.13 (t, *J* 7.6, 1 H, *p*-Ph), 6.96 (d, *J* 7.2, 1 H, pyr-CH), 5.13 (dt, $J_{\alpha,NH}$ 7.6, $J_{\alpha,\beta}$ 6.3, 1 H, α-H), 3.68 (s, 3 H, ester) and 3.39 (m, 2 H, β-H_2).

Methyl *N*-benzoyl-3-(2-benzylpyrimidin-4-yl)alaninate 10

Methyl *N*-benzoyl-3-(2-oxo-3-phenyl-2*H*-isoxazolo[2,3-*a*]-pyrimidin-5-yl)alaninate **8** (0.4 g) was dissolved in acetic acid (50 cm^3), Zn powder (0.4 g) was added and the mixture was stirred for 5 h at room temperature before being filtered and the solvent was removed under reduced pressure. The residue was chromatographed with ethyl acetate–light petroleum (1 : 1) to give *compound* **10** (0.15 g, 42%) as an oil (Found: C, 70.87; H, 5.90; N, 11.07. $C_{22}H_{21}N_3O_3$ requires C, 70.38; H, 5.64; N, 11.19%); $[\alpha]_D^{25}$ +98 (*c* 1, dichloromethane); $\delta_H(CDCl_3)$ 8.57 (d, *J* 5.2, 1 H, pyr-CH), 7.63 (d, $J_{o,m}$ 8.5, 2 H, *o*-PhCO), 7.49 (d, $J_{NH,\alpha}$ 7.4, 1 H, NH), 7.40 (d, $J_{o,m}$ 7.7, 2 H, *o*-Ph), 7.20–7.34 (m, 6 H, *m*-, *p*-PhCO + *m*-, *p*-Ph), 7.02 (d, *J* 5.2, 1 H, pyr-CH), 5.21 (dt, $J_{\alpha,NH}$ 7.4, $J_{\alpha,\beta}$ 4.6, 1 H, α-H), 4.24 (s, 2 H, benzoyl), 3.68 (s, 3 H, ester) and 3.45 (dd, $J_{\beta,\alpha}$ 4.6, J_{gem} 16.1, 2 H, β-H_2).

3-(2-Benzylpyrimidin-4-yl)alanine hydrochloride 11

Methyl *N*-benzoyl-3-(2-benzylpyrimidin-4-yl)alaninate **10** (0.2 g) was dissolved in 5 M HCl (20 cm^3) and the solution was refluxed for 4 h. The reaction mixture was washed successively with diethyl ether (3 × 10 cm^3) and ethyl acetate (1 × 10 cm^3), mixed with decolorizing carbon (0.1 g) and filtered. The water was removed under reduced pressure to give the *salt* **11** (0.14 g, 90%) as a semisolid (Found: C, 57.87; H, 4.91; N, 13.99. $C_{14}H_{15}N_3O_2$·HCl requires C, 57.52; H, 5.18; N, 14.38%); $[\alpha]_D^{25}$ −7 (*c* 1, H_2O); $\delta_H(D_2O)$ 8.80 (d, *J* 5.1, 1 H, pyr-CH), 7.3–7.49 (m, 6 H, Ph + pyr-CH), 4.51 (m, 3 H, α-H + CH_2) and 3.66 (d, $J_{\beta,\alpha}$ 5.3, 2 H, β-H_2).

Methyl *N*-benzoyl-3-{2-[ethoxy(phenyl)methyl]pyrimidin-4-yl}alaninate 13

Methyl *N*-benzoyl-3-(2-oxo-3-phenyl-2*H*-isoxazolo[2,3-*a*]pyrimidin-5-yl)alaninate **8** (0.25 g) was dissolved in ethanol (40 cm^3). The mixture was refluxed under nitrogen with protection from light for 48 h. The solvent was evaporated off under reduced pressure. The residue was chromatographed, with ethyl acetate–light petroleum gradient (1 : 1–3 : 1) as eluent. The isolated *product* **13** was an oily 5 : 4 mixture of two diastereoisomers (0.11 g, 44%) (Found: C, 68.48; H, 5.71; N, 9.61. $C_{24}H_{25}N_3O_4$ requires C, 68.72; H, 6.01; N, 10.02%); $[\alpha]_D^{25}$ +52 (*c* 1, $CHCl_3$); $\delta_H(CDCl_3)$ major: 8.63 (d, *J* 5.1, 1 H, pyr-CH), 7.75 (d, $J_{o,m}$ 6.7, 2 H, *o*-PhCO), 7.25–7.55 (m, 9 H, *m*-, *p*-PhCO + Ph + NH), 7.05 (d, *J* 5.1, 1 H, pyr-CH), 5.55 (s, 1 H, CH), 5.27 (m, 1 H, α-H), 3.58 (s, 3 H, ester), 3.32–3.56 m, 4 H, ethyl + β-H_2) and 1.27 (t, *J* 7.0, 3 H, ethyl); minor: 8.62 (d, *J* 5.1, 1 H, pyr-CH), 7.74 (d, $J_{o,m}$ 6.7, 2 H, *o*-PhCO), 7.25–7.55 (m, 9 H, *m*-, *p*-PhCO + Ph + NH), 7.07 (d, *J* 5.1, 1 H, pyr-CH), 5.57 (s, 1 H, CH), 5.27 (m, 1 H, α-H), 3.69 (s, 3 H, ester), 3.32–3.56 (m, 4 H, ethyl + β-H_2) and 1.25 (t, *J* 7.0, 3 H, ethyl).

Ring opening of compound 8 by (*R*)-(+)-α-aminoethylbenzene

Methyl *N*-benzoyl-3-(2-oxo-3-phenyl-2*H*-isoxazolo[2,3-*a*]pyrimidin-5-yl)alaninate **8** (0.15 g) was dissolved in 1,4-dioxane (30 cm^3), and (*R*)-(+)-α-aminoethylbenzene (0.15 cm^3) was added. The mixture was refluxed under nitrogen with protection from light for 48 h. The solvent was evaporated off under reduced pressure. The residue was chromatographed with ethyl acetate–light petroleum gradient (1:3–1:1). The first isolated *diastereoisomer* of compound **14** was an oil (free base) (0.06 g, 34%) (Found: C, 72.57; H, 6.41; N, 11.33. $C_{30}H_{30}N_4O_3$ requires C, 72.85; H, 6.11; N, 11.47%); $[\alpha]_D^{25}$ +218 (*c* 1, $CHCl_3$); $\delta_H(CDCl_3)$ 8.51 (d, *J* 5.2, 1 H, pyr-CH), 7.74 (dd, $J_{o,m}$ 7.0, $J_{o,p}$ 1.6, 2 H, *o*-PhCO), 7.27–7.65 (m, 14 H, *m*-, *p*-PhCO + Ph + NH), 6.99 (d, *J* 5.2, 1 H, pyr-CH), 5.30 (dt, $J_{\alpha,NH}$ 7.9, $J_{\alpha,\beta}$ 4.5, 1 H, α-H), 4.79 (s, 1 H, CH), 3.73 (s, 3 H, ester), 3.36–3.68 (m, 3 H, CH + β-H_2) and 1.41 (d, *J* 6.5, 3 H, Me).

The second isolated *diastereoisomer* of *compound* **14** was an oil (free base) (0.05 g, 28%) (Found: C, 72.82; H, 6.23; N, 11.18%); $[\alpha]_D^{25}$ +96 (*c* 1, chloroform); $\delta_H(CDCl_3)$ 8.58 (d, *J* 5.1, 1 H, pyr-CH), 7.78 (d, $J_{o,m}$ 7.7, 2 H, *o*-PhCO), 7.25–7.66 (m, 14 H, *m*-, *p*-PhCO + Ph + NH), 7.02 (d, *J* 5.1, 1 H, pyr-CH), 5.21 (dt, $J_{\alpha,NH}$ 8.3, $J_{\alpha,\beta}$ 4.4, 1 H, α-CH), 5.01 (s, 1 H, CH), 3.37–3.66 (m, 3 H, CH + β-H_2), 3.48 (s, 3 H, ester) and 1.38 (d, *J* 6.4, 3 H, Me).

Methyl *N*-benzoyl-3-(2-benzoyl)pyrimidin-4-yl)alaninate 15

Methyl *N*-benzoyl-3-(2-oxo-3-phenyl-2*H*-isoxazolo[2,3-*a*]pyrimidin-5-yl)alaninate **8** (0.5 g) was dissolved in DMSO (5 cm^3) and heated for 5 min at 135 °C. The reaction mixture was loaded on a silica gel column and the products were eluted with ethyl acetate–light petroleum (1:1) to give *compound* **15** (0.4 g, 86%) as an oil (Found: C, 67.59; H, 5.09; N, 10.66. $C_{22}H_{19}N_3O_4$ requires C, 67.84; H, 4.92; N, 10.80%); $[\alpha]_D^{25}$ +113 (*c* 1, chloroform); $\delta_H(CDCl_3)$ 8.85 (d, *J* 5.1, 1 H, pyr-CH), 8.00 (dd, $J_{o,m}$ 7.0, $J_{o,p}$ 1.2, 2 H, *o*-PhCO), 7.80 (d, $J_{NH,\alpha}$ 7.7, 1 H, NH), 7.67 (dd, $J_{o,m}$ 7.1, $J_{o,p}$ 1.6, 2 H, *o*-Ph), 7.28–7.59 (m, 7 H, *m*-, *p*-PhCO + *m*-, *p*-Ph + pyr-CH), 5.28 (dt, $J_{\alpha,NH}$ 7.7, $J_{\alpha,\beta}$ 4.7, 1 H, α-H), 3.65 (s, 3 H, ester) and 3.56 (dd, $J_{\beta,\alpha}$ 4.7, J_{gem} 15.8, 2 H, β-H_2).

3-(2-Benzoylpyrimidin-4-yl)alanine hydrochloride 16

Methyl *N*-benzoyl-3-(2-benzoylpyrimidin-4-yl)alaninate **15** (0.05 g) was dissolved in 5 M HCl (20 cm^3) and the solution was refluxed for 4 h. The reaction mixture was washed successively with diethyl ether (3 × 10 cm^3) and ethyl acetate (1 × 10 cm^3) and the solvent was removed under reduced pressure to give the *salt* **16** (0.03 g, 76%), mp >300 °C (Found: C, 54.32; H, 4.43; N, 13.92. $C_{14}H_{13}N_3O_3 \cdot HCl$ requires C, 54.64; H, 4.59; N, 13.65%); $[\alpha]_D^{25}$ −27 (*c* 1, H_2O); $\delta_H(D_2O)$ 8.81 (d, *J* 5.3, 1 H, pyr-CH), 7.80 (d, $J_{o,m}$ 8.0, 2 H, *o*-PhCO), 7.69 (d, *J* 5.3, 1 H, pyr-CH), 7.66 (dd, $J_{p,m}$ 6.4, $J_{p,o}$ 1.1, 1 H, *p*-PhCO), 7.49 (dd, $J_{m,p}$ 6.4, $J_{m,o}$ 8.0, 2 H, *m*-PhCO), 4.61 (t, $J_{\alpha,\beta}$ 6.0, 1 H, α-H) and 3.61 (d, $J_{\beta,\alpha}$ 6.0, 2 H, β-H_2).

2,4-Dinitrophenylhydrazone derivative 17 of ester 15

Methyl *N*-benzoyl-3-(2-benzoylpyrimidin-4-yl)alanine **15** (0.05 g) was dissolved in ethanol (1 cm^3), 2,4-dinitrophenylhydrazine reagent[23] (2 cm^3) was added, and the mixture was boiled for 4 min. The *product* **17** precipitated on cooling (0.0.5 g, 68%), mp 154–155 °C (Found: C, 59.27; H, 4.19; N, 17.26. $C_{28}H_{23}N_7O_7$ requires C, 59.05; H, 4.07; N, 17.22%); $\delta_H(CDCl_3)$ 15.17 (s, 1 H, NH), 9.16 [d, *J* 2.43, 1 H, $C_6H_3(NO_2)_2$], 9.00 (d, *J* 5.1, 1 H, pyr-CH), 8.24–8.42 [m, 2 H, $C_6H_3(NO_2)_2$], 7.68 (dd, $J_{o,m}$ 6.3, $J_{o,p}$ 2.2, 4 H, *o*-PhCO + *o*-Ph), 7.27–7.48 (m, 8 H, *m*-, *p*-PhCO + *m*-, *p*-Ph + pyr-CH + NH), 5.31 (dt, $J_{\alpha,NH}$ 7.7, $J_{\alpha,\beta}$ 5.4, 1 H, α-H), 3.66 (s, 3 H, ester) and 3.56 (dd, $J_{\beta,\alpha}$ 5.4, J_{gem} 15.4, 2 H, β-H_2).

References

1 D. G. Martin, D. J. Duchamp and C. G. Chichester, *Tetrahedron Lett.*, 1973, 2549.
2 J. J. Hansen and P. Krogsgaard-Larsen, *J. Chem. Soc., Perkin Trans. 1*, 1980, 1826.
3 P. Krogsgaard-Larsen, E. Ø. Nielsen and D. R. Curtis, *J. Med. Chem.*, 1984, **27**, 585.
4 P. Krogsgaard-Larsen, T. Honoré, J. J. Hansen, D. R. Curtis and D. Lodge, *Nature* (*London*), 1980, **284**, 64.
5 P. Krogsgaard-Larsen, L. Brehm, J. S. Johansen, P. Vinzents, J. Lauridsen and D. R. Curtis, *J. Med. Chem.*, 1985, **28**, 668.
6 J. M. Martinez Martos, M. J. Ramirez Exposito, M. Arrazola Saniger and J. M. Ramirez Huerta, *Rev. Clin. Exp.*, 1996, **196**, 113.
7 M. Sugahara, S. Shibanoki, A. Matsumoto, T. Kubo and K. Ishikawa, *Neuroscience* (*Okayama, Jpn.*), 1995, **21**(Suppl), P127.
8 M. Morari, G. Calo, L. Ferraro, A. Fabrizi, N. Acciarri, G. Piazza, C. Bianchi and L. Beani, *Neurochem. Int.*, 1995, **26**, 77.
9 K. A. Trujillo and H. Akil, *Drug Alcohol Depend.*, 1995, **38**, 139.
10 J. E. Dolfini, H. E. Applegate, G. Bach, H. Basch, J. Bernstein, J. Schwartz and F. Weisenborn, *J. Med. Chem.*, 1971, **14**, 117.
11 G. R. Nagarajan, L. Diamond and S. Ressler, *J. Org. Chem.*, 1973, **38**, 621.
12 R. Johnson and J. F. Koerner, *J. Med. Chem.*, 1988, **31**, 2057.
13 R. Gmelin, *Z. Physiol. Chem.*, 1959, **316**, 164.
14 E. A. Bell, *Biochim. Biophys. Acta*, 1961, **47**, 602.
15 E. A. Bell and R. G. Foster, *Nature*, 1962, **194**, 91.
16 J. H. Dewar and G. Shaw, *J. Chem. Soc.*, 1962, 583.
17 W. J. Haggerty, Jr., R. H. Springer and C. C. Cheng, *J. Heterocycl. Chem.*, 1965, **2**, 1.
18 G. Zvilichovsky and V. Gurvich, *Tetrahedron*, 1995, **51**, 5479.
19 G. Zvilichovsky and V. Gurvich, *J. Chem. Soc., Perkin Trans. 1*, 1995, 2509.
20 G. Zvilichovsky and V. Gurvich, *Tetrahedron*, 1997, in the press.
21 G. Zvilichovsky, V. Gurvich and S. Segev, *J. Org. Chem.*, 1995, **60**, 5250.
22 G. Zvilichovsky and V. Gurvich, *J. Org. Chem.*, 1996, **61**, 3212.
23 A. I. Vogel, *Text-book of Practical Organic Chemistry*, Longman and Green, London–New York–Toronto, p. 923.

Paper 6/04879J
Received 11*th July* 1996
Accepted 22*nd November* 1996