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Synthesis of Optically Active α-(Imidazol-1-yl, Purin-9-yl or Uracil-1-yl) Propanoic and Succinic Acid Derivatives

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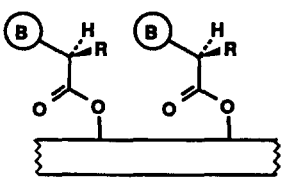
Abstract: An efficient route is reported to chiral propanoic and succinic acid derivatives substituted in the α-position with adenine, hypoxanthine and uracil groups. These nucleic acid base derivatives are obtained in optically pure form by a procedure which starts from an amino acid as a convenient chiral synthon.

Therapeutic intervention at the nucleic acid level is an attractive strategy and to this end much attention has been devoted to obtaining synthetic polynucleotides of conventional structure. An alternative approach involves the use of polynucleotide analogues in which the sugar-phosphate backbone of natural polynucleotides has been replaced with a synthetic polymer which is biologically stable. Thus some or all of the normal nucleic acid bases have been grafted on to polymers which have a carbon backbone with suitable pendant functional groups *eg* 1. Examples include poly(vinyl alcohol),[1] polyvinylamine,[2] copolymer of vinyl alcohol and vinylamine,[3] and polyacrylamides[4]. Polymers containing suitable reactive NH sites in the backbone have also been coupled to appropriately derivatised nucleic acid bases. These include polyethylenimine,[5] polytrimethylenimine,[6] polyurethane[7] and polylysine[8] and some of these polynucleotide analogues have been found to exhibit antiviral activity.[9]



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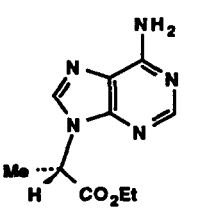

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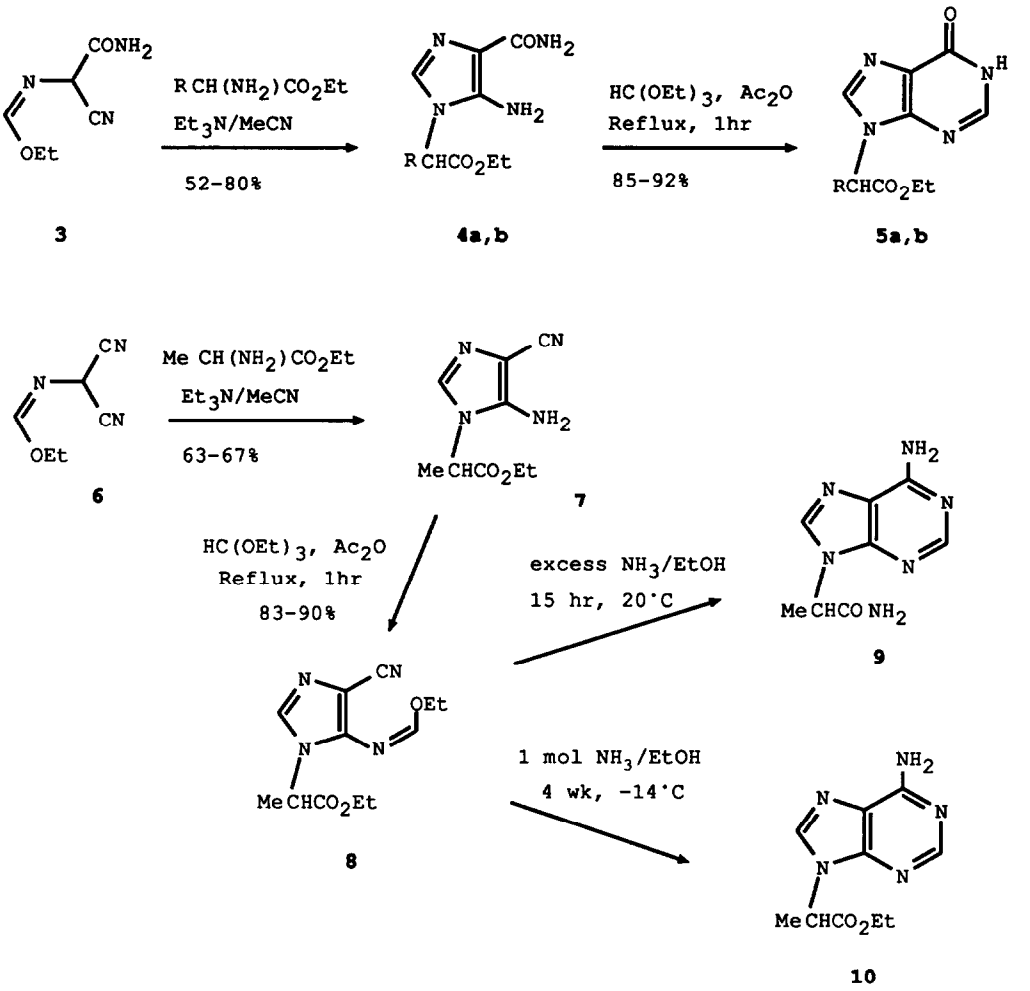
In all of these systems the nucleic acid base is usually coupled to the polymer through a chiral centre to simulate the stereochemical specificity of conventional nucleic acids. A simple but convenient approach to linking the polymer to the base involves the use of a carboxylic acid group (**1**). The nucleic acid base can be introduced as an α-substituent at the chiral centre and the acidic group provides a suitable means of linking to the polymer. Thus optically active α-substituted propanoic acids have been used in the synthesis of polynucleotide analogues, these derivatives having the advantage that they are suitable for the determination of enantiomeric excess with chiral shift reagents.

However the synthesis of suitable α-substituted propanoic acids, especially in enantiomerically pure form, has been difficult. Early work employed routes which were either nonstereospecific and hence required tedious resolution of enantiomers or gave low yields of products with variable levels of enantiomeric excess.[10] Recently the Mitsunobu reaction has been applied successfully[11] to the stereospecific synthesis of the *R* form of the adenine derivative **2** in modest yield (33%). However this methodology was unsuccessful when applied to the formation of the analogous pyrimidine derivatives which were only available in low yield by an alternative route which caused significant racemisation.

We report here a more general method for the synthesis of propanoic and succinic acids with imidazole, purine or uracil moieties as α-substituents, a method which gives these nucleic acid base derivatives in enantiomerically pure form. Our objective of the stereospecific attachment of a purine base (or related heterocyclic base) to the α-position of the carboxylic acid, has been achieved indirectly by starting with a chiral amino acid and building on an imidazole ring by a modification of the Shaw method[12,13] (**Scheme 1**). Thus the imidate **3** was condensed with the *R* and *S* forms of ethyl alanine or diethyl aspartate to afford the corresponding imidazoles **4a** and **4b** in good yield without compromising the chiral integrity of the starting amino acids. Similarly imidate **6** reacted with *R* and *S* ethyl alanine to give the *R* and *S* forms of imidazole **7**. The mild conditions for this amination/cyclisation reaction are crucial to the success of this strategy.

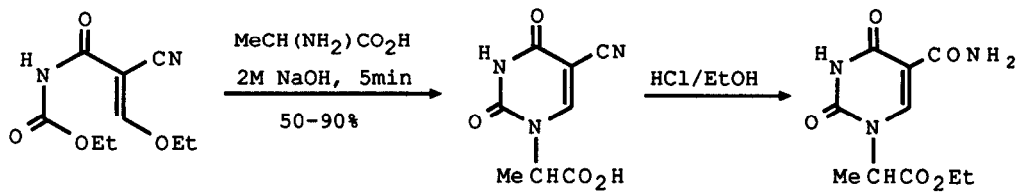
Cyclisation of the imidazoles **4a** and **4b** to the corresponding hypoxanthines **5a** and **5b** was easily achieved by the standard method[13] of refluxing for 12-15 hours with 0.5 equivalents of triethylorthoformate in dimethylformamide (DMF) but the product was totally racemised. If the amount of DMF was reduced to the minimum required to dissolve the reagents then this procedure led to only partial loss of chiral integrity (enantiomeric excess in the range 30-50%). Evidently the DMF solvent is sufficiently basic to promote proton exchange at the α-position of the side chain attached at the N-atom, either at the imidazoline stage and/or the hypoxanthine stage. To avoid this difficulty, excess triethylorthoformate was used as solvent with the addition of a few drops of acetic anhydride. This method[14] probably results in the transient formation of the reactive species $MeCO_2CH(OEt)_2$ which catalyses the condensation/cyclisation reaction since the reaction time was reduced to 1 hour. Using this modified procedure (*ie* in the absence of a base) the hypoxanthines **5** were obtained in optically pure form without seriously diminishing the yield (85-92%). Racemisation was also observed in the reaction of the cyanoimidazole **7** with triethylorthoformate in DMF. However using the conditions described above the imidate **8** was obtained in good yield without loss of chiral integrity. Cyclisation of imidates of type **8** to the corresponding adenine is usually carried out by exposure to excess ammonia for 12-15 hours at room temperature. Unfortunately such a procedure not only



Scheme 1

racemised the chiral centre but also converted the ester function to amide. Only the racemic amide **9** was obtained. Treatment of **8** with one equivalent of ammonia in ethanol for 6 hours at room temperature left the ester function unaltered but there was still substantial racemisation. However at −14 °C racemisation is totally suppressed although the cyclisation is now very slow and required four weeks to reach completion. Thus an optically pure form of the adenine derivative **10** was obtained in excellent yield.

Extension of this method of preparing optically active α-purinyl-propanoic acid derivatives to the formation of analogous uracilyl derivatives was also investigated (**Scheme 2**). Reaction of the precursor **11** with alanine in sodium hydroxide solution gave the corresponding uracil **12** without loss of chiral integrity. This reaction did not work as effectively with the amino ester, due to the tendency for the latter to dimerise



11 **12** **13**

Scheme 2

to give a diketopiperazine. Esterification of **12** with ethanolic HCl produced concomitant hydrolysis of the nitrile, presumably via the imidate, to give the 5-carbamoyluracil ester **13**.

The methodology described above for the formation of the enantiomeric forms of simple acids derivatised with nucleic acid bases offers a convenient route to optically active polynucleotide analogues. It has the advantage that the chirality is introduced via an amino acid which is reasonably inexpensive and readily available as either enantiomer in optically pure form.

EXPERIMENTAL

Optical rotations were measured at 589 nm in the concentration range 1-5 g dl^{-1} on an AA-10 automatic polarimeter. NMR spectra were obtained on FX90 or GX270 spectrometers. The spectra of enantiomers were generally identical to each other and to that of the corresponding racemate, and data are given only once. Determinations of enantiomeric excess (ee) were made using the enantioselective solvating agent (*S*)-(+)-2,2,2-trifluoro-1-(9-anthryl)ethanol (TAE) (from Aldrich). In our hands this reagent was more effective than chiral shift reagents based on lanthanide β-diketonates.[11] With TAE there is no broadening of signals and the TAE/substrate ratio (R_{TAE}) is easily determined from the spectrum obviating the need for weighing substrate or shift reagent.

Typically a substrate concentration of < 1 mg cm^{-3} was used and good enantiomeric discrimination obtained for a value of R_{TAE} in the range 5-10. In the case of purines and uracils with an N-substituent derived from alanine the methyl doublet signal at δ 1.6-1.8 was monitored. The analogous signal in the hypoxanthines derived from aspartic acid, the methylene doublet at δ 3.3, was less satisfactory. For an R_{TAE} value of *ca* 10 a very small splitting of this signal was observed but this was probably due to induced non-equivalence of these intrinsically prochiral protons. A more useful monitor of ee was the slightly broadened aromatic singlet initially at δ 8.05 (H-2) which shifts and splits to give two enantiomeric singlets at δ 7.85 and 7.87 for an R_{TAE} value of *ca* 10. The other aromatic singlet (H-8) shifts from δ 7.98 to 7.94 without splitting.

Ethyl 2-(5-amino-4-carbamoylimidazo-1-yl)propanoate (4a), diethyl 2-(5-amino-4-carbamoylimidazo-1-yl)succinate (4b) and ethyl 2-(5-amino-4-cyanoimidazo-1-yl)propanoate (7):

The synthesis of the racemic compounds, *RS*-**4a** and *RS*-**7**, have been described previously[13] and the following compounds were obtained by the same procedure starting from *R*- or *S*-alanine ethyl ester hydrochloride ($[\alpha]_D$, and $[\alpha]_D$ respectively), or *RS*-, *R*- or *S*-aspartic acid diethyl ester hydrochloride, ($[\alpha]_D$, and $[\alpha]_D$ respectively).

R-**4a**, 52%, mp 89-90 °C, λ_{max} (MeOH) 264 nm, $[\alpha]_D$ +31° (EtOH), ee = 100%; Anal. Calcd. for $C_9H_{14}N_4O_3$ (226.2): C, 47.03; H, 6.32; N, 24.38. Found C, 46.86; H, 6.12; N, 23.93. ^{1}H-NMR ($CDCl_3$): δ = 1.20 (t, 3H, CH_3), 1.65 (d, 3H, CH_3), 4.15 (q, 2H, CH_2), 5.02 (q, 1H, CH), 5.80 (s, 2H, NH_2), 6.80 (s, 2H, $CONH_2$), 7.20 (s, 1H, =CH).

S-**4a**, 61%, mp 87-89 °C, λ_{max} (MeOH) 263 nm, $[\alpha]_D$ −29° (EtOH), ee = 100%; Anal. Found C, 46.87; H, 6.32; N, 23.95.

RS-**4b**, 73%, mp 106 °C, λ_{max} (MeOH) 263 nm; Anal. Calcd. for $C_{12}H_{18}N_4O_5$ (298.3): C, 48.32; H, 6.08; N, 18.78. Found C, 48.55; H, 6.13; N, 18.62. ^{1}H-NMR (DMSO): δ = 1.20 (t, 6H, two CH_3), 3.20 (d, 2H, CH_2), 4.11 (q, 4H, two CH_2), 5.28 (t, 1H, CH), 5.85 (s, 2H, NH_2), 6.75 (s, 2H, $CONH_2$), 7.22 (s, 1H, =CH).

R-**4b**, 78%, mp 106 °C, λ_{max} (MeOH) 262 nm, $[\alpha]_D$ −18° (EtOH), ee = 100%; Anal. Found C, 48.26; H, 6.08; N, 18.62.

S-**4b**, 69%, mp 106 °C, λ_{max} (MeOH) 261 nm, $[\alpha]_D$ +19° (EtOH), ee = 100%; Anal. Found C, 48.31; H, 6.10; H, 18.50.

R-**7**, 63%, mp 155 °C, λ_{max} (MeOH) 245 nm, $[\alpha]_D$ +47° (EtOH), ee = 100%; Anal. Calcd. for $C_9H_{12}N_4O_2$ (208.2): C, 51.91; H, 5.81; N, 26.91: Found C, 51.89; H, 5.88; N, 26.96. ^{1}H-NMR (DMSO): δ = 1.31 (t, 3H, CH_3), 1.78 (d, 3H, CH_3), 4.26 (q, 2H, CH_2), 4.90 (m, 3H, CH and NH_2), 7.20 (s, 1H, =CH).

S-**7**, 67%, mp 154 °C, λ_{max} (MeOH) 245 nm, $[\alpha]_D$ −46° (EtOH), ee = 100%; Anal. Found C, 51.82; H, 5.83; N, 26.90.

Ethyl 2-(hypoxanthin-9-yl)propanoate (5a), diethyl 2-(hypoxanthin-9-yl)succinate (5b):

The imidazole **4a** or **4b** (2 mmol) in triethylorthoformate (20 g) containing two drops of acetic anhydride was refluxed for 3 hr, with exclusion of moisture. After cooling, the solvent was removed under vacuum, the residual oil azeotroped with dry ethanol (2 x 30 cm^3) and the resulting solid recrystallised (EtAc) to give *RS*-**5a**, 82%, mp 116 °C, (Lit.[13] 116 °C); ^{1}H-NMR ($CDCl_3$): δ = 1.28 (t, 3H, CH_3), 1.88 (d, 3H, CH_3), 4.26 (q, 2H, CH_2), 5.38 (q, 1H, CH), 8.00 (s, 1H, =CH), 8.16 (s, 1H, =CH), 12.82 (s, 1H, NH).

R-**5a**, 77%, mp 176-177 °C, $[\alpha]_D$ +2.3° (EtOH), ee = 100%; Anal. Calcd. for $C_{10}H_{12}N_4O_3$(236.2): C, 50.68; H, 5.05; N, 23.52. Found: C, 50.57; H, 5.07; N, 23.39.

S-**5a**, 69%, mp 173-174 °C, $[\alpha]_D$ −1.6° (EtOH), ee = 100%; Anal. Found C, 50.57; H, 5.07; N, 23.39.

RS-**5b**, 44%, mp 141-143 °C, λ_{max} (MeOH) 251 nm; Anal. Calcd. for $C_{13}H_{16}N_4O_5$ (308.3): C, 50.64; H, 5.23; N 18.18. Found C, 50.59; H, 5.30; N, 18.15. ^{1}H-NMR (DMSO): δ = 1.13 (t, 6H, two CH_3), 3.38 (d, 2H, CH_2), 4.10 (q, 4H, two CH_2), 5.79 (t, 1H, CH), 8.09 (s, 1H, =CH), 8.21 (s, 1H, =CH), 10.64 (s, 1H, NH).

R-**5b**, 57%, mp 151 °C, λ_{max} (MeOH) 247 nm, $[\alpha]_D$ +3.1 (EtOH), ee = 100%; Anal. Found C, 50.31; H, 5.85; N, 18.47.

S-**5b**, 49%, mp 151 °C, λ_{max} (MeOH) 247 nm, $[\alpha]_D$ −2.7° (EtOH), ee = 100%; Anal. Found C, 50.95; H, 5.42; N, 18.51.

Ethyl 2-(adenin-9-yl)propanoate (10):

The imidazole **7** was treated as above with two drops of acetic anhydride in excess triethylorthoformate for 1 hr. The oily product was taken up in toluene and precipitated with petroleum ether (40/60) and dried over P_2O_5 to afford **ethyl 2-(4-cyano-5-ethoxymethylene-aminoimidazol-1-yl)propanoate (8).**

R-**8**, 90%, mp 64.5-65 °C, $[\alpha]_D$ +2.6° (EtOH), ee = 100%; Anal. Calcd. for $C_{10}H_{12}N_4O_3$ (236.2): C, 54.55; H, 6.06; N 21.21. Found C, 54.83; H, 5.82; N, 21.53. ^{1}H-NMR (DMSO): δ = 1.26 (t, 3H, CH_3), 1.40 (t, 3H, CH_3), 1.75 (d, 3H, CH_3), 4.22 (q, 2H, OCH_2), 4.35 (q, 2H, OCH_2), 4.94 (q, 1H, CH), 7.45 (s, 1H, =CH), 8.38 (s, 1H, =CH).

S-**8**, 83%, mp 60.5-61.5 °C, $[\alpha]_D$ −1.6° (EtOH), ee = 100%; Anal. Found C, 54.80; H, 5.79; N, 21.52.

The ethoxymethylene derivative **8** (1.5 mmol) in EtOH (10 cm^3) and one equivalent of NH_3/EtOH were separately cooled to −20 °C and mixed slowly at that temperature. After 4 weeks at −14 °C the solvent was removed at reduced temperature, the residue azeotroped with ethanol (3 x 25 cm^3) and the resulting solid recrystallised (H_2O) to give *RS*-**10**, 71%, mp 165-166 °C, (lit.[13] 165 °C).

R-**10**, 54%, mp 178-179 °C, $[\alpha]_D$ +2.6° (EtOH), ee = 98%; Anal. Calcd. for $C_{10}H_{13}N_5O_2$ (235.3): C, 51.05; H, 5.57; N 29.77. Found C, 51.12; H, 5.61; N, 29.82. ^{1}H-NMR ($CDCl_3$): δ = 1.26 (t, 3H, CH_3), 1.88 (d, 3H, CH_3), 4.25 (q, 2H, OCH_2), 5.42 (q, 1H, CH), 6.00 (br s, 2H, NH_2), 8.05 (s, 1H, =CH), 8.35 (s, 1H, =CH).

S-**10**, 60%, mp 179-181 °C, $[\alpha]_D$, −2.7° (EtOH), ee = 99%; Anal. Found C, 50.98; H, 5.50; N, 29.95.

2-(Adenin-9-yl)propanamide (9):

The ethoxymethylene derivative **8** was treated with excess saturated ethanolic ammonia at 20 °C for 15 hr. The precipitate was the amide **9** (78%), mp >280 °C, λ_{max} (MeOH) 258 nm. Anal. Calcd. for $C_{10}H_{13}N_5O_2$ (235.2): C, 44.94; H, 5.11; N, 39.31. Found C, 44.96; H, 4.68; N, 39.63. ^{1}H-NMR ($CDCl_3$): δ = 1.72 (t, 3H, CH_3), 5.22 (s, 1H, CH), 7.22 (s, 2H, NH_2), 7.31 and 7.77 (two s, 2H, $CONH_2$), 8.13 (s, 1H, =CH), 8.24 (s, 1H, =CH).

2-(5-Cyanouracil-1-yl)propanoic acid (12):

α-Cyano-β-ethoxy-N-ethoxycarbonylacrylamide[15] (5.30 g, 25 mmol) was added to a solution of alanine (2.23 g, 25mmol) in sodium hydroxide (2M, 12.5 cm^3, 25 mmol). After dissolution was nearly complete, sodium hydroxide (10M, 2.5 cm^3, 25 mmol) was added, the solution stirred for 5 min and acidifed with conc HCl at 5 °C. The precipitated product was collected after 12 hr at 5 °C and recrystallised (EtOH) to give *RS*-**12**, 78%, mp >270 °C(dec.), λ_{max} (H_2O) 284 nm; Anal. Calcd. for $C_8H_7N_3O_4$ (209.2) C, 45.94; H, 3.37; N, 20.09. Found C, 45.46; H, 3.36; N 20.06. ^{1}H-NMR (DMSO): δ = 1.42 (d, 3H, CH_3), 4.76 (q, 1H, CH), 8.58 (s, 1H, =CH), 11.8 (br s, 1H, NH).

R-**12**, 66%, mp >270 °C(dec), λ_{max} (H_2O) 283 nm; Anal. Found C, 45.57; H, 3.41; N 19.99.

S-**12**, 48%, mp >270 °C(dec), λ_{max} (H_2O) 283 nm; Anal. Found C, 46.30; H, 3.43; N, 20.49.

Ethyl 2-(5-Carbamoyluracil-1-yl)propanoate (13):

The acid **12** (250 mg, 1.20 mmol) was suspended in ethanol (30 cm^3) and saturated with dry HCl gas at 5 °C. Solvents were removed under vacuum and the gummy residue crystallised from ethanol/ether and dried in vaccuo to *RS*-**13**, 23%, mp 204 °C; ^{1}H-NMR ($CDCl_3$): δ = 1.30 (t, 3H, CH_3), 1.72 (d, 3H, CH_3), 4.25 (q, 2H, CH_2), 5.25 (q, 1H, CH), 8.55 (s, 1H, =CH), 8.7 (br s, 1H, NH), 5.85 and 8.4 (two br s, 2H, $CONH_2$).

R-**13**, 26%, mp 204 °C, $[\alpha]_D$ +21.2 (dimethylformamide), ee = 100%; Anal. Calcd. for $C_{10}H_{13}N_3O_5$ (255.2) C, 47.06; H, 5.13; N, 16.46. Found C, 47.16; H, 5.06; N 16.53.

S-**13**, 16%, mp 206 °C, $[\alpha]_D$ −23.7 (dimethylformamide), ee = 100%; Anal. Found C, 46.66; H, 5.08; N 16.10.

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REFERENCES.

1. Overberger, C.G.; Chang, J.Y.; Gunn, V.E. *J. Polym. Sci., Polym. Chem. Ed.* **1985**, *23*, 1321.
   Overberger, C.G.; Chang, J.Y *J. Polym. Sci., Polym. Chem. Ed.* **1989**, *27*, 3589.
2. Overberger, C.G.; Chen, C.C. *J. Polym. Sci., Polym. Chem. Ed.* **1987**, *25*, 389.
3. Overberger, C.G.; Lu, C.X. *J. Polym. Sci., Polym. Chem. Ed.* **1987**, *25*, 1523.
4. Overberger, C.G.; Ferng, W. *J. Polym. Sci., Polym. Chem. Ed.* **1986**, *24*, 3349.
5. Overberger, C.G.; Morishima, Y. *J. Polym. Sci., Polym. Chem. Ed.* **1980**, *18*, 1247,1267.
   Wada, T.; Inaki, Y.; Takemoto, K. *Polym. J.* **1988**, *20*, 1059; **1989**, *21*, 11.
6. Overberger, C.G.; Chang, J.Y. *J. Polym. Sci., Polym. Chem. Ed.* **1989**, *27*, 4013.
   Overberger, C.G.; Chang, J.Y.; Gunn, V.E. *J. Polym. Sci., Polym. Chem. Ed.* **1989**, *27*, 99.
7. Lu, C.X.; Yang, Y.; Xiao, C.D.; Li, A.X. *J. Polym. Sci., Polym. Chem. Ed.* **1987**, *25*, 3267.
8. Wada, T.; Inaki, Y.; Takemoto, K. *J. Bioactive Compatible Polym.* **1992**, *7*, 25.
9. Pitha, J. *Adv. Polym. Sci.*, **1983**, *50*, 1.
10. Lan, M.J.; Overberger, C.G. *Org. Prep. Proc. Int.*, **1988**, *20*, 261.
11. Overberger, C.G.; Chang, J.Y. *Tetrahedron Lett.* **1989**, *30*, 51.
12. Shaw, G.; Warrener, R.N.; Ralph, R.K.; Butler, D.N. *J.Chem.Soc.* **1959**, 1648.
13. Birkett, P.R.; Chapleo, C.B.; Mackenzie, G. *Synthesis*, **1991**, 157.
14. Montgomery, J.A.; Temple, C. *J. Amer. Chem. Soc.*, **1957**, *79*, 5238.
15. Atkinson, M.R.; Shaw, G.; Warrener, R.N. *J. Chem. Soc.*, **1956**, 4118.