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Preparation of Anionic Active Ester Derivatives of Amino Acids

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Abstract

Benzyloxycarbonyl amino acids were condensed with the 2,4,6-trimethylbenzyl esters of 4-hydroxy-3-nitro-benzoic and -phenylacetic acids to give crystalline protected active esters, which could then be converted into anionic derivatives by either selective or complete removal of the protecting groups. Several anionic active esters of aliphatic dicarboxylic acids were also prepared similarly.

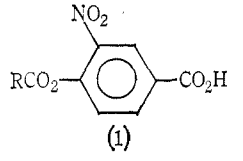
Anionic active esters (1) derived from straight- and branched-chain aliphatic carboxylic acids have been used extensively as substrates in studies on the catalytic activity of certain synthetic polymers as model enzyme systems.[1–4] Analogous ester derivatives of some amino acids were required for various purposes, and their preparation is described in the present communication.

Compounds (1) are readily obtained by the action of acid chlorides or anhydrides on 4-hydroxy-3-nitrobenzoic acid, although in one case a mixed anhydride [2; R = $CH_2CH(Me)Et$] was formed preferentially and subsequent base-catalysed rearrangement to (1) was necessary.[1] In order to avoid such complications, and also possible purification difficulties, it was considered that preparation of the proposed amino acid derivatives (3) would best proceed from phenolic components with the carboxyl group protected by the acid-labile 2,4,6-trimethylbenzyl group. Synthesis of the relevant precursors (4; $n = 0$ and 1) and related compounds from new 'active polyester' reagents has already been reported.[5,6] The homologues (4) with $n = 0$ and 1 were intended to provide two series of final anionic active esters (3) with substantially different pK_a values.

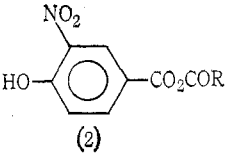
Condensation of the phenols (4) with various protected amino acids by a modification[6] of the standard *N,N'*-dicyclohexylcarbodiimide method recommended for *o*-nitrophenyl esters[7,8] gave the corresponding highly crystalline and easily purified protected active esters, which are listed in Table 1.

[1] Overberger, C. G., and Cho, I., *J. Polym. Sci., Polym. Chem. Ed.*, 1968, **6**, 2741.
[2] Overberger, C. G., Glowaky, R. C., and Vandewyer, P.-H., *J. Am. Chem. Soc.*, 1973, **95**, 6008.
[3] Overberger, C. G., and Pacansky, T. J., *J. Polym. Sci., Polym. Symp. No. 45*, 1974, 39.
[4] Ueoka, R., Shimamoto, K., Maezato, Y., and Ohkubo, K., *J. Org. Chem.*, 1978, **43**, 1815.
[5] Stewart, F. H. C., *Aust. J. Chem.*, 1978, **31**, 2523.
[6] Stewart, F. H. C., *Aust. J. Chem.*, 1979, **32**, 405.
[7] Bodanszky, M., Funk, K. W., and Fink, M. L., *J. Org. Chem.*, 1973, **38**, 3565.
[8] Bodanszky, M., Kondo, M., Yang Lin, C., and Sigler, G. F., *J. Org. Chem.*, 1974, **39**, 444.

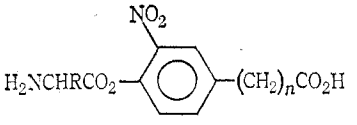
Some of these intermediates were converted into anionic active ester derivatives by either selective acidic cleavage of the 2,4,6-trimethylbenzyl group with trifluoroacetic acid[9] leaving benzyloxycarbonyl intact, or complete removal of both protecting groups with hydrogen bromide in acetic acid.[10] The benzyloxycarbonyl derivative of the glycine ester (3a) and the hydrobromides of anionic active esters of L-alanine (3b) and (3c), and L-phenylalanine (3d) were obtained by these procedures. The hydrobromide of (3c) was converted into the zwitterionic free ester which, however, was considerably less stable than the salt.



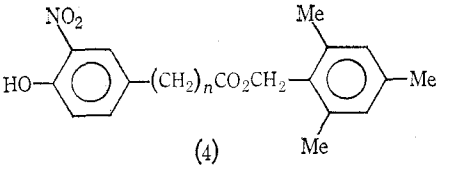
(1)



(2)



	n	R
(3a)	1	H
(3b)	0	Me
(3c)	1	Me
(3d)	1	CH_2Ph



(4)

The 2,4,6-trimethylbenzyl derivatives (4) were also used similarly for the preparation of several anionic active esters of adipic and sebacic acids, of interest in connection with certain cross-linking experiments.

Table 1. Protected amino acid active esters

Rotations are at concn 0·5 in dimethylformamide. Abbreviations used are: Z, benzyloxycarbonyl; Nps, *o*-nitrophenylthio

Protected amino acid	(4) *n*	Yield (%)	Recryst. solvent	M.p. (°C)	$[\alpha]_D$ (deg)	Found (%) C	H	N	Calc. (%) C	H	N
Z-Gly	0	95	EtOH	103–103·5	—	63·8	5·4	5·3	64·0	5·1	5·5
Z-Gly	1	87	EtOAc/pet	105–106	—	64·9	5·4	5·7	64·6	5·4	5·4
Nps-Ala	0	88	EtOH	104·5–106·5	−96·0	57·7	4·7	7·7	57·9	4·6	7·8
Z-Ala	0	78	EtOH	108·5–109·5	−35·0	65·0	5·5	5·2	64·6	5·4	5·4
Z-Ala	1	89	EtOAc/pet	132, 143	−34·4	65·4	5·8	5·4	65·2	5·6	5·2
Z-Val	0	79	EtOH	93·5–94·5	−21·2	65·9	5·7	5·3	65·7	5·8	5·1
Z-Val	1	89	Et_2O/pet	97–98	−16·6	66·3	6·0	5·0	66·2	6·0	5·0
Z-Leu	1	77	Et_2O/pet	70·5–73	−26·2	67·0	6·4	4·8	66·7	6·3	4·9
Z-Phe	1	88	EtOAc/pet	142·5–143	−42·0	69·1	5·8	4·4	68·9	5·6	4·6

Experimental

The microanalyses were carried out by the Australian Microanalytical Service, Melbourne. Melting points are uncorrected. Infrared spectra were obtained with KBr discs. Proton magnetic resonance spectra were run in [D_6]dimethyl sulfoxide with a Varian A-60D spectrometer. Satisfactory spectra were obtained for each product.

[9] Stewart, F. H. C., *Aust. J. Chem.*, 1966, **19**, 1511.
[10] Stewart, F. H. C., *Aust. J. Chem.*, 1966, **19**, 1067.

(*a*) *Protected Amino Acid 4-(2',4',6'-Trimethylbenzyloxycarbonyl)- and 4-(2',4',6'-Trimethylbenzyloxycarbonylmethyl)-2-nitrophenyl Esters*

The modified *N,N'*-dicyclohexylcarbodiimide procedure described previously for other 4-substituted 2-nitrophenols was used.[6] Details of the *protected active esters* are given in Table 1.

(*b*) *Preparation of Anionic Active Ester Derivatives of Amino Acids*

Selective or complete cleavage of the protecting groups in the 2,4,6-trimethylbenzyl precursors (Table 1) was effected by trifluoroacetic acid[9] or 2 M hydrogen bromide in acetic acid,[10] respectively.

(i) *(4-Benzyloxycarbonylglycyloxy-3-nitrophenyl)acetic acid* was obtained in quantitative yield and recrystallized from ethyl acetate/light petroleum, m.p. 133–134° (rapid heating). ν_{max} 1785, 1725, 1705 cm^{-1} (Found: C, 55·3; H, 4·3; N, 7·4. $C_{18}H_{16}N_2O_8$ requires C, 55·7; H, 4·1; N, 7·2%).

(ii) *4-(L-Alanyloxy)-3-nitrobenzoic acid hydrobromide* was obtained in 89% yield and recrystallized from ethanol/ether, m.p. 170° (dec.); ν_{max} 1780, 1710 cm^{-1}; $[\alpha]_D^{21}$ −17·2° (*c*, 0·5; $HCONMe_2$) (Found: C, 35·8; H, 3·3; N, 8·2. $C_{10}H_{11}BrN_2O_6$ requires C, 35·8; H, 3·3; N, 8·4%).

(iii) *(4-L-Phenylalanyloxy-3-nitrophenyl)acetic acid hydrobromide* (97% yield) was recrystallized from ethanol/ether, m.p. 180–182° (dec.); ν_{max} 1770, 1700 cm^{-1}; $[\alpha]_D^{22}$ −5·8° (*c*, 0·5; $HCONMe_2$) (Found: C, 48·4; H, 3·9; N, 6·4. $C_{17}H_{17}BrN_2O_6$ requires C, 48·0; H, 4·0; N, 6·6%).

(iv) *(4-L-Alanyloxy-3-nitrophenyl)acetic acid hydrobromide* (72% yield) was recrystallized from ethanol/ether, m.p. 178–180° (dec.); ν_{max} 1780, 1725 cm^{-1}; $[\alpha]_D^{22}$ −25·8° (*c*, 0·5; $HCONMe_2$) (Found: C, 37·8; H, 3·8; Br, 23·2; N, 8·1. $C_{11}H_{13}BrN_2O_6$ requires C, 37·8; H, 3·7; Br, 22·9; N, 8·0%).

A solution of the hydrobromide in ethanol was treated with a slight excess of pyridine. Zwitterionic *(4-L-alanyloxy-3-nitrophenyl)acetic acid* separated on chilling (46% yield); ν_{max} 1775, 1615 cm^{-1} (Found: C, 48·8; H, 4·7; N, 10·2. $C_{11}H_{12}N_2O_6$ requires C, 49·3; H, 4·5; N, 10·4%). Attempted recrystallization from aqueous ethanol resulted in extensive decomposition.

(*c*) *Anionic Active Ester Derivatives of Dicarboxylic Acids*

2,4,6-Trimethylbenzyl-protected intermediates were prepared as in (*a*), and the protecting group removed as in (*b*).

Bis[2-nitro-4-(2',4',6'-trimethylbenzyloxycarbonyl)phenyl] adipate (87% yield) was recrystallized from ethyl acetate/ethanol, m.p. 125–125·5°; ν_{max} 1764, 1721 cm^{-1} (Found: C, 64·9; H, 5·6; N, 3·9. $C_{40}H_{40}N_2O_{12}$ requires C, 64·9; H, 5·4; N, 3·8%).

4,4'-(Adipoyldioxy)bis(3-nitrobenzoic acid) was obtained in quantitative yield and recrystallized from dimethylformamide/ethanol, m.p. 205–206°; ν_{max} 1785, 1690 cm^{-1} (Found: C, 50·2; H, 3·4; N, 6·1. $C_{20}H_{16}N_2O_{12}$ requires C, 50·4; H, 3·4; N, 5·9%).

Bis[2-nitro-4-(2',4',6'-trimethylbenzyloxycarbonylmethyl)phenyl] adipate (65% yield) was recrystallized from dimethylformamide/ethanol, m.p. 188·5–189·5°; ν_{max} 1770, 1720 cm^{-1} (Found: C, 65·4; H, 5·7; N, 3·8. $C_{42}H_{44}N_2O_{12}$ requires C, 65·6; H, 5·7; N, 3·6%).

[4,4'-(Adipoyldioxy)bis(3-nitrophenyl)]bisacetic acid (92% yield) was recrystallized from dimethylformamide/ethanol, m.p. 175–176·5°; ν_{max} 1751, 1690 cm^{-1} (Found: C, 52·6; H, 3·8; N, 6·0. $C_{22}H_{20}N_2O_{12}$ requires C, 52·4; H, 4·0; N, 5·6%).

4,4'-(Sebacoyldioxy)bis(3-nitrobenzoic acid) was obtained directly from the washed 2,4,6-trimethylbenzyl precursor (66% yield) in 64% yield and recrystallized from ethanol/light petroleum, m.p. 189–190°; ν_{max} 1778, 1693 cm^{-1} (Found: C, 53·9; H, 4·7; N, 5·1. $C_{24}H_{24}N_2O_{12}$ requires C, 54·1; N, 4·5; N, 5·3%).

Acknowledgment

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