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Control of the Enantiomeric Purity and Correlation with the Absolute Configuration of N-Protected 2-Cyanoglycinates

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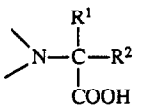
Abstract : The enantiomeric purity of substituted N-phthaloyl-2-cyanoglycine methyl esters obtained by resolution of covalent diastereoisomers was easily established by measuring the shift separation of signals using $Eu(hfc)_3$. A correlation with the absolute configuration of enantiomers was observed.

There has been a major interest shown in recent years in the synthesis and biological properties of non-proteinogenic aminoacids. In particular α,α-disubstituted α-aminoacids show an important activity as enzyme inhibitors, and could potentially be incorporated into peptides[1]. The presence of these unusual aminoacids induces specific conformations which are of interest in the structure stabilization of peptides and proteins. This observation may be correlated with the nature of the substituents R^1 and R^2 of aminoacids **1**, especially for α-methylated α-aminoacids[2].



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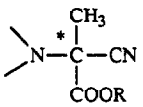

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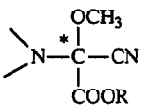
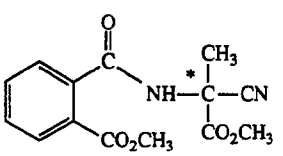


3

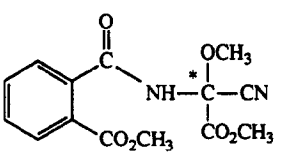
We are currently studying the synthesis of chiral α-disubstituted aminoesters **2** and **3** which contain several functional groups. These multifunctional carbon compounds[3] are chiral auxiliaries for the syntheses of optically active natural products. In a previous paper[4], we showed that these chiral cyanoglycinates are versatile building blocks for the enantioselective multistep synthesis of cephems, cephalosporins and cephamycins[5] (7-methoxycephalosporins), known antibiotics or enzyme inhibitors from the β-lactam series.

Moreover, at the present time the biological and biochemical properties of α-aminonitriles have been quite widely studied. Some important activities of these compounds, critically dependent on their configuration, have recently been reported[6].

Previously we have published the synthesis of chiral 2-cyanoglycinates **4** and **5** with an *o*-methoxycarbonylbenzoyl (OMCB) as the amino protecting group.



4 (*R* and *S* enantiomers)



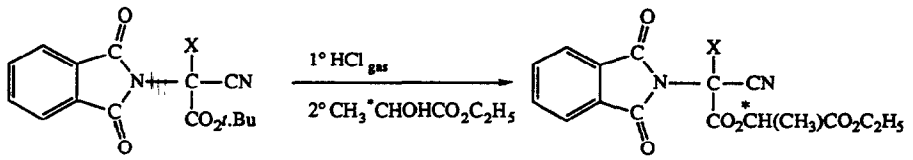
5 (*S* enantiomer)

In this paper we describe an original ring closure of this *o*-methoxycarbonylbenzoyl group leading to the phthaloyl group using 2-hydroxypyridine.

The enantiomeric purity was studied by complexation with a chiral NMR shift reagent. There was found to be an interesting linear correlation between the variations in the chemical shifts of the methyl esters signal and the signal of the methyl or methoxy substituent of the enantiomers. This different correlation corresponded with the *R* or *S* absolute configuration of the chiral centre of these α-disubstituted aminoesters.

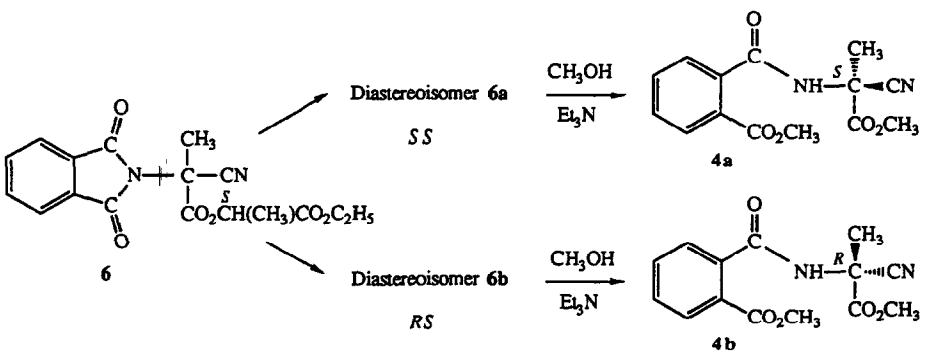
Results and discussion

The resolution of N-phthaloyl-2-cyanoglycinates by synthesis and separation of covalent diastereoisomers has already been described, using chiral ethyl lactate as a resolving reagent. The chiral auxiliary was introduced from the acid by esterification with a phenyldichlorophosphate[5a,7] (or phosphorus oxychloride)/N,N-dimethylformamide complex as coupling reagent.

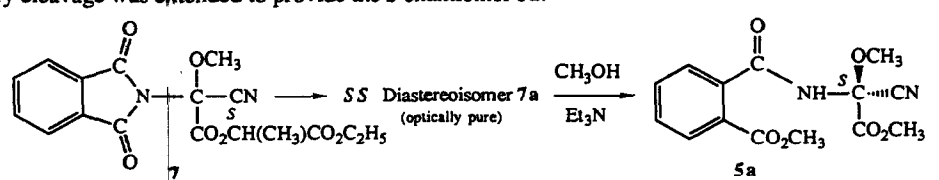


X = CH_3 or OCH_3

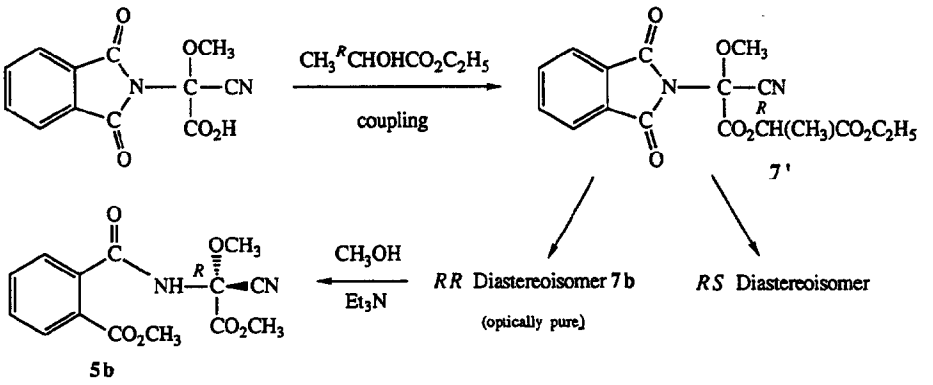
The resolution of the diastereoisomeric pair of 2-methyl-2-cyanoglycinate obtained from commercially available (*S*)-(-)-ethyl lactate was carried out by silica gel chromatography. The transesterification reaction with methanol in the presence of triethylamine to remove the chiral auxiliary was always accompanied by kinetically favoured ring opening of the phthaloyl protection[5a] leading to both *R* and *S* enantiomers **4a** and **4b** :



Successive crystallizations were necessary to resolve the 2-methoxy-2-cyanoglycinate synthons. Only the *SS* diastereoisomer **7a** crystallized in enantiomerically pure form. The same methodology for the chiral auxiliary cleavage was extended to provide the *S* enantiomer **5a**.



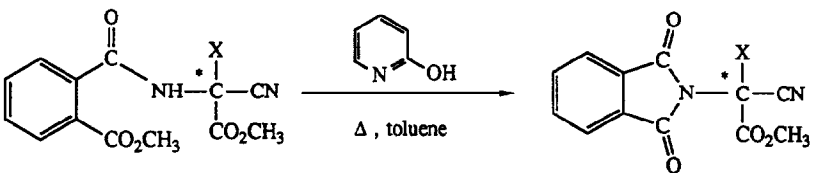
We investigated the synthesis of the corresponding pure *R* enantiomer **5b** starting from *(R)*-(+)-ethyl lactate. This chiral auxiliary was prepared by esterification of *(R)*-(-)-lactic acid* with ethanol in the presence of *p*-toluenesulfonic acid. Following our methodology to obtain covalent diastereoisomers, the *RR* diastereoisomer could be selectively crystallized in its pure form to give the pure *R* enantiomer **5b**, after similar transesterification.



We investigated intramolecular aminolysis for ring closure of the OMCB (*o*-methoxycarbonylbenzoyl) group into the phthaloyl group : products containing this protective group are more stable and easier to crystallize. Moreover, we thought that it would be easier to control the enantiomeric purity of compounds with the amino function** protected by the phthaloyl substituent.

2-Hydroxypyridine is known to be an effective catalyst for the aminolysis of esters[8]. The formation of the amide bond is considerably accelerated by this catalyst, possibly due to the participation of both the basic and the acidic groups of 2-hydroxypyridine to allow a concerted displacement of a pseudocyclic transition state[9].

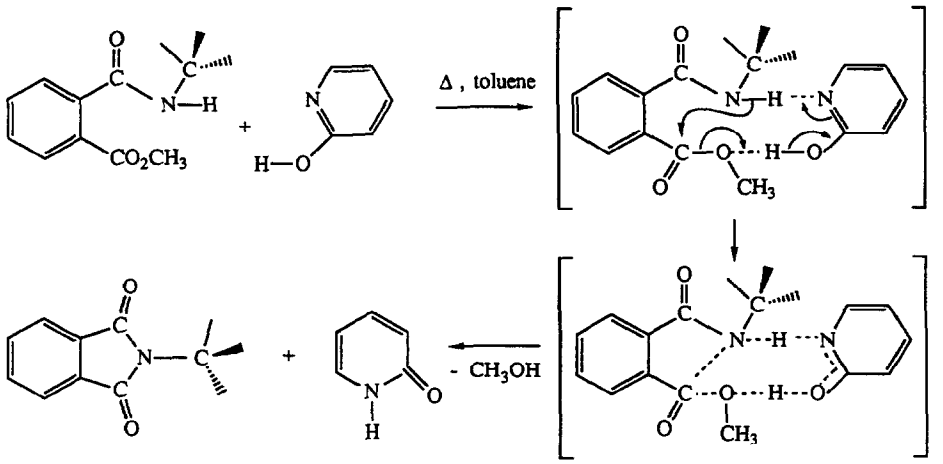
The reaction was successfully carried out by refluxing in toluene with this reagent to afford enantiomers **8** and **9** in high yields. Different spectra (^1H, ^{13}C and mass spectroscopy) are in accordance with the proposed structures of these compounds and are similar to those obtained before with the racemic synthons[10].



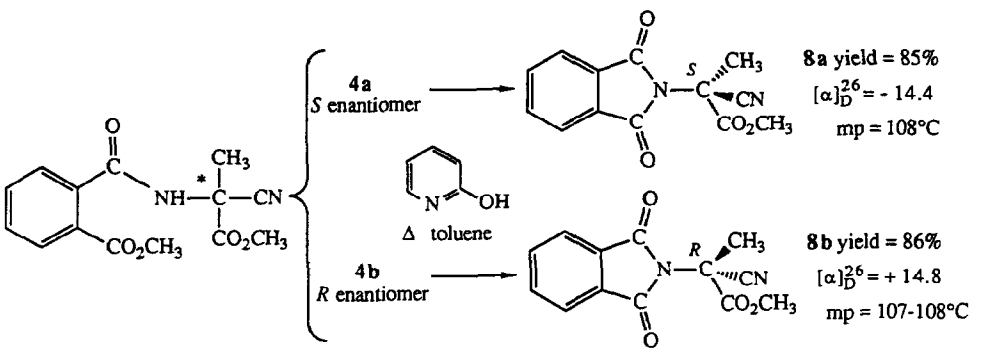
* We thank the Rhône-Poulenc Society for the gift of a sample.

** We thought that in the case of cyanoglycinates protected by a OMCB group, the carbamoyl function would be the stronger site of coordination with the chiral NMR shift reagent and one difficulty could be the possible superposition of the two methyl esters signals, thus preventing a correct measurement of enantiomeric purity.

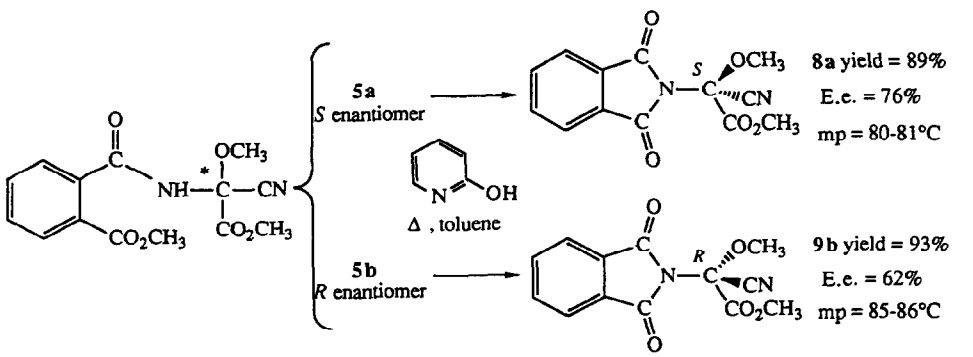
The suggested mechanism is described below :



This cyclisation reaction was applied to α-methyl and α-methoxycyanoglycinate enantiomers **4** and **5** of known absolute configuration :



(melting point of racemic mixture : 112-113°C)



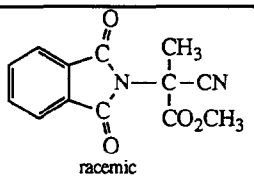
(melting point of racemic mixture : 109-110°C)

NMR determination of the enantiomeric purity[11] of α-aminoacid derivatives has previously been studied[12]. This NMR method requires the addition of a chiral lanthanide salt to convert the aminoacid derivative into a mixture of two diastereoisomers, which can usually be distinguished by their different chemical shifts.

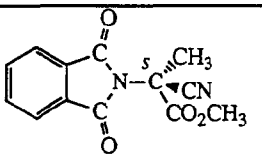
Firstly, in order to select the most suitable experimental parameters (nature of the chiral lanthanide salt, NMR solvent, substrate concentration), we recorded the NMR spectra of the racemic mixture of the N-phthaloyl α-methyl (**2**) and α-methoxy (**3**) synthons.

The NMR spectra of 0.2 molar $CDCl_3$ solutions of N-phthaloyl-α-methyl cyanoglycinates showed that following addition of increasing amounts of $Eu(hfc)_3$, only the methyl groups undergo a variation in chemical shift. The choice of this chiral NMR reagent was justified by the success previously obtained for aminoacid derivatives[12]. The large chemical shift variation of the methyl signal ($\Delta\delta$ CH_3) compared with that of the methyl ester signal ($\Delta\delta$ CO_2CH_3) could be explained by a preferential complexation of the amide function with the europium salt***.

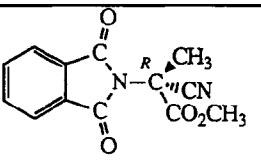
Furthermore, instead of considering the relation between the chemical shift variations and the amounts of $Eu(hfc)_3$ added, as is commonly used, we established a linear correlation between the recorded $\Delta\delta$ CH_3 and $\Delta\delta$ CO_2CH_3 values, giving two straight lines, each corresponding to one enantiomer (table 1). Taking these results into account, we applied the method to each *R* and *S* enantiomer using similar experimental conditions. Identical linear correlations were discovered, allowing us to establish a straightforward correlation with the absolute configuration of the stereogenic carbon atom.



racemic
δ_o CH_3 = 2.31
δ_o CO_2CH_3 = 3.93



S enantiomer
8a



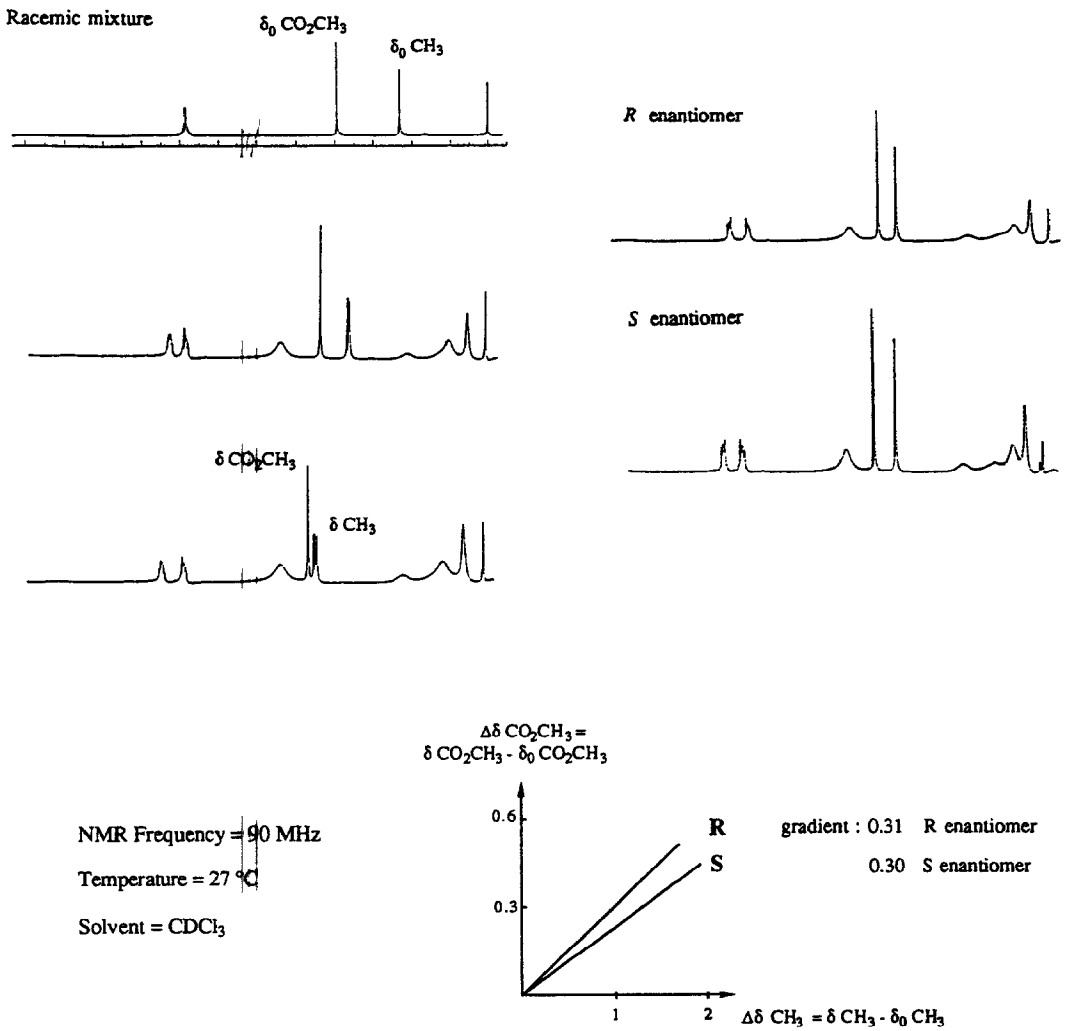
R enantiomer
8b

racemic		*S* enantiomer 8a		*R* enantiomer 8b	
$\Delta\delta$ CH_3	$\Delta\delta$ CO_2CH_3	$\Delta\delta$ CH_3	$\Delta\delta$ CO_2CH_3	$\Delta\delta$ CH_3	$\Delta\delta$ CO_2CH_3
0.14	0.05	0.41	0.13	0.42	0.14
0.38 and 0.39	0.13	0.82	0.26	0.74	0.24
0.94 and 0.98	0.31	1.14	0.35	1.00	0.31
1.28 and 1.31	0.41	1.44	0.44	1.29	0.41
1.52 and 1.57	0.48	1.65	0.50	1.54	0.48
2.05 and 2.11	0.65	1.91	0.58	1.90	0.60
2.25 and 2.32	0.70	2.23	0.67	2.09	0.66
2.47 and 2.55	0.78	2.50	0.75	2.38	0.75
a = 0.31	a = 0.30	a = 0.30		a = 0.31	
b = 0.01 and	b = 0.01	b = 0.01		b = 0.01	
r > 0.99	r > 0.99	r > 0.99		r > 0.99	

Table 1. $\Delta\delta$ values (in ppm) of the methyl and methyl ester signals of racemic and each enantiomer of N-phthalimido methyl cyanoglycinate with successive additions of $Eu(hfc)_3$.
Straight lines : $\Delta\delta$ CO_2CH_3 = a $\Delta\delta$ CH_3 + b (r : coefficient of linear regression)

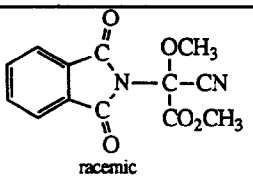
*** Thus only the splitting of the methyl group signal could be observed, the addition of $Eu(hfc)_3$ being without effect on the methyl ester group.

SCHEME 1

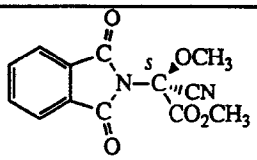


The excellent enantiomeric purity of this α-methylated synthon was confirmed by the presence of only one signal for the methyl group. Experimental determination of the enantiomeric purity of this chiral multifunctional compound proved the purity (>95%) of asymmetric molecules derived from this cyanoglycinate.

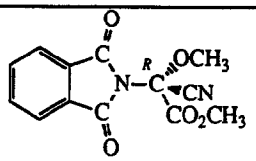
This method was then applied to N-phthaloyl α-methoxy cyanoglycinates using the previously determined experimental conditions. The splitting of both the methoxy and the methyl ester signals was established from the racemic mixture. As expected, a similar linear correlation was observed for the chemical shift variation $\Delta\delta$ of the methoxy signal ($\Delta\delta$ OCH_3) and the methyl ester signal ($\Delta\delta$ CO_2CH_3) for each enantiomer (table 2).



δ_o OCH_3 = 3.69
δ_o CO_2CH_3 = 3.97



S enantiomer
9a

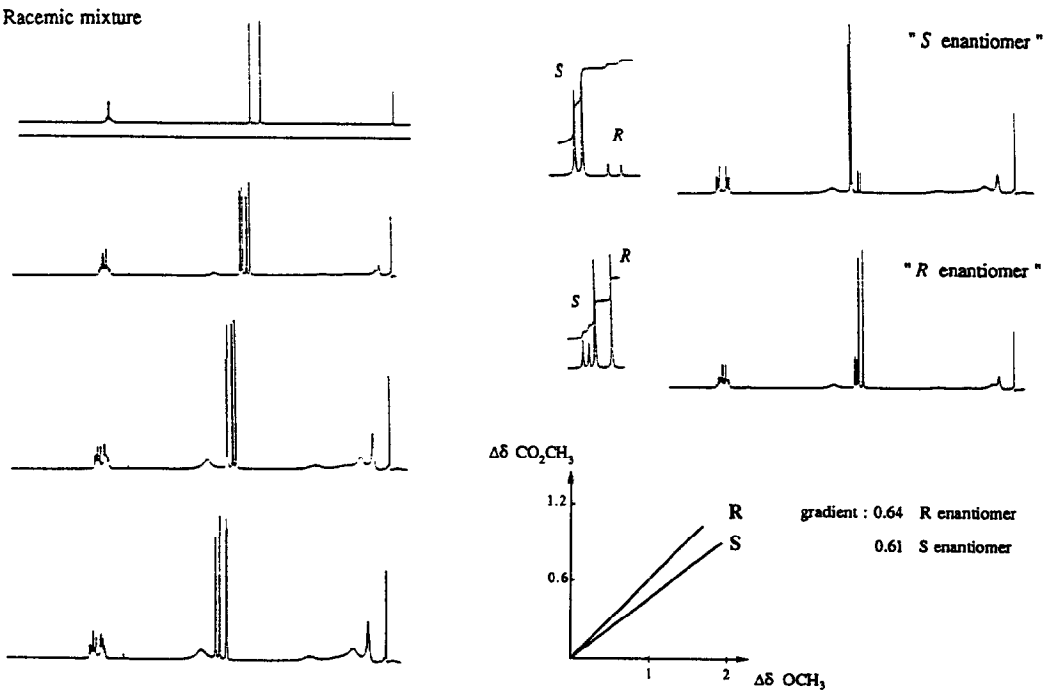


R enantiomer
9b

1st enantiomer		2nd enantiomer		*S* enantiomer **9a**		*R* enantiomer **9b**	
$\Delta\delta$ OCH_3	$\Delta\delta$ CO_2CH_3	$\Delta\delta$ OCH_3	$\Delta\delta$ CO_2CH_3	$\Delta\delta$ OCH_3	$\Delta\delta$ CO_2CH_3	$\Delta\delta$ OCH_3	$\Delta\delta$ CO_2CH_3
0.14	0.10	0.21	0.13	0.25	0.16	0.16	0.11
0.23	0.15	0.32	0.20	0.31	0.19	0.34	0.23
0.31	0.20	0.45	0.28	0.58	0.36	0.42	0.27
0.38	0.25	0.55	0.34	0.86	0.53	0.54	0.35
0.45	0.29	0.66	0.41			0.62	0.40
0.51	0.33	0.75	0.46				
0.58	0.37	0.84	0.51				
0.65	0.42	0.95	0.58				
0.71	0.45	1.03	0.63				
a = 0.64		a = 0.60		a = 0.61		a = 0.64	
b = 0.01		b = 0.01		b = 0.01		b = 0.01	
r > 0.99		r > 0.99		r > 0.99		r > 0.99	

Table 2. $\Delta\delta$ values (in ppm) of the methoxy and methyl ester signals of racemic and each enantiomer of N-phthalimido methoxy cyanoglycinate.

SCHEME 2



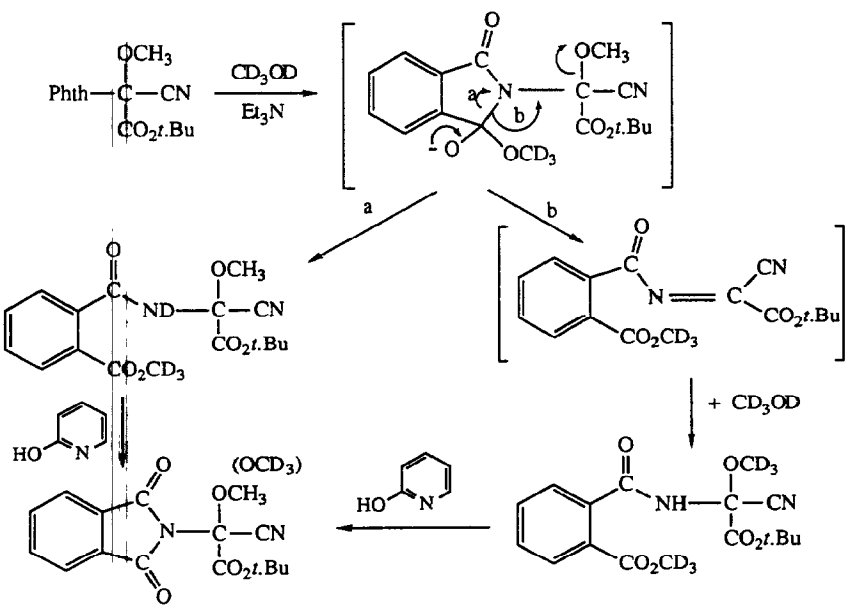
We observed that each isomer derived from methoxyglycinate was not enantiomerically pure, owing to the formation of two diastereoisomeric complexes. Resolution using *(S)*-(-)-ethyl lactate as chiral auxiliary led to enantiomers **9** in *S/R* = 88/12 ratio (e.e. = 76%). Under the same conditions the *RR* diastereoisomer **7b** purifed by successive crystallizations using *(R)*-(+)-ethyl lactate as chiral auxiliary afforded a mixture of enantiomers in *R/S* = 81/19 ratio (e.e. = 62%).

The loss of the enantiomeric purity of compound **9** could be explained during the access to the enantiomers from the corresponding pure diastereoisomer. The mechanism of the transesterification reaction on the racemic synthon **3** was established using CD_3OD and triethylamine, then the ring closure of the OMCB protection was carried out using 2-hydroxypyridine. The cyanoglycinate *ter*-butyl ester was used to avoid the possible transesterification of primary (methyl) and secondary (derived from ethyl lactate) alcohols.

First, we noted that the ratio of intensities between the ^1H NMR signals of the methoxy and the *ter*-butyl groups was not 1/3 as expected but 1/3.84, corresponding to a deutereted methoxylation of 22%. An identical relationship was observed using mass spectrometry by comparing the spectra of the starting material with the same product after the two afore-mentioned reactions.

Phth—C(OCH$_3$/OCD$_3$)(CN)—CO$_2$t.Bu	215/218 ($M^{+\cdot}$ - CO_2tBu)	290/293 ($M^{+\cdot}$ - 26)	334/337 *CI* $(M+NH_3)^+$
Initial substrate	100/2.1	100/3.1	100/0.7
After the two reactions	100/18.5	100/15.8	100/16.7

The incorporation of OCD_3 could result from the previously described[13] methoxylation mechanism.



Addition of deuterated methanol to the acylimine intermediate could induce the observed loss of optical purity. This competition is not possible when the tetrafunctional carbon is substituted by the methyl group, which is not a leaving group, as the methoxy substituent.

In conclusion, the access to N-phthaloyl methyl ester cyanoglycinate enantiomers was shown to be effective after the resolution of covalent diastereoisomers by transesterification with methanol, followed by a powerful method of ring closure with 2-hydroxypyridine to regenerate the desired phthaloyl protection. The enantiomeric purity of these chirons could easily be established by ^1H NMR shift separation of the different enantiomers signals by addition of $Eu(hfc)_3$. A straightforward correlation with the absolute configuration was described.

Experimental:

^1H NMR spectra were recorded on a JEOL instrument J.N.M. FX 90-MHz. Chemical shifts are reported as δ values in ppm down field from internal standard (Me_4Si). Mass spectra were recorded on a HP 5889 A spectrometer at 70 eV. Optical rotatory powers were measured using a AA.10 OPTICAL ACTIVITY polarimeter. Melting points were determined using a microscope with a Kofler hot stage and are uncorrected.

2-cyano-2-methoxy-2-*o*-methoxycarbonylbenzamido methylethanoate : 2.2 mmol (0.3 ml) of triethylamine were added to 1 mmol (0.36 g) of pure 2*R*-2*R*' diastereoisomer in 10 ml of anhydrous methanol. The solution was stirred for 2 hr at room temperature, then concentrated under reduced pressure. The residue was dissolved in 100 ml of EtOAc and washed with 2x50 ml of brine. The solution was then dried ($MgSO_4$) and concentrated to white crystals. M.p. = 68-70°C (petroleum ether/EtOAc).

General procedure for ring closing to the phthaloyl group :
1.2 mmol of 2-hydroxypyridine were added to 1 mmol of 2-cyano-2-*o*-methoxycarbonylbenzamido substituted ethanoate in 15 ml of anhydrous toluene. The advancement of the reaction, brought to reflux under nitrogen atmosphere, was monitored by TLC. The solution was concentrated and purified by silica gel chromatography (petroleum ether/EtOAc : 3/2).

2-cyano-2-phthalimido methylpropanoate : *R* enantiomer : 86% Yield, m.p. = 107-108°C (petroleum ether/EtOAc). $[\alpha]_D^{26}$ = + 14.8 (c = 0.4, $CHCl_3$). *S* enantiomer : 85% Yield, m.p. = 108°C (petroleum ether/EtOAc). $[\alpha]_D^{26}$ = - 14.4 (c = 0.4, $CHCl_3$). Racemic : 84% Yield, m.p. = 112-113°C (petroleum ether/EtOAc).

2-cyano-2-methoxy-2-phthalimido methylethanoate : *S* enantiomer : 93% Yield, m.p. = 77-78°C (petroleum ether/EtOAc). *R* enantiomer : 89% Yield, m.p. = 80-81°C (petroleum ether/EtOAc). Racemic : m.p. = 109-110°C (petroleum ether/EtOAc).

References :

1 a) Barrett, G. C. *Chemistry and Biochemistry of the Amino Acids*, Ed. Chapman and Hal, London, **1985**. b) O'Donnell, M. J. *α-Aminoacid Synthesis, Tetrahedron Symposia-in-Print number 33, Tetrahedron*, **1988**, *44*, [17], 5253-5614. c) *Synthesis of Optically Active α-Aminoacids*, Williams, R. M. Pergamon, Oxford, **1989**.

2 a) Altmann, E.; Nebel, K.; Mutter, M. *Helv. Chim. Acta*, **1991**, *74*, 800-806. b) Obrecht, D.; Spiegler, C.; Schönholzer, P.; Müller, K.; Heimgartner, H.; Stierli, F. *Helv. Chim. Acta*, **1992**, *75*, 1666-1696.

3 a) Takeuchi, Y.; Asahina, M.; Murayama, A.; Hori, K.; Koizumi, T. *J. Org. Chem.*, **1986**, *51*, 955-956. b) Takeuchi, Y.; Asahina, M.; Nagata, K.; Koizumi, T. *J. Chem. Soc. Perkin Trans. I*, **1987**, 2203-2207. c) Takeuchi, Y.; Nagata, K.; Koizumi, T. *J. Org. Chem.*, **1987**, *52*, 5061-5063. d) Takeuchi, Y.; Asahina, M.; Hori, K.; Koizumi, T. *J. Chem. Soc. Perkin Trans. I*, **1988**, 1149-1153. e) Takeuchi, Y.; Ogura, H.; Ishii, Y.; Koizumi, T. *J. Chem. Soc. Perkin Trans. I*, **1989**, 1721-1725. f) Takeuchi, Y.; Nabetani, M.; Tagaki, K.; Hagi, T.; Koizumi, T. *J. Chem. Soc. Perkin Trans I*, **1991**, 49-53. g) Takeuchi, Y. *Reviews on Heteroatom Chemistry*, **1991**, *4*, 64-77. h) Takeuchi, Y.; Ogura, H.; Kanada, A.; Koizumi, T. *J. Org. Chem.*, **1992**, *57*, 2196-2199. i) Takeuchi, Y.; Itoh, N.; Kawahara, S.; Koizumi, T. *Tetrahedron*, **1993**, *49*, [9], 1861-1870.

4 Hudhomme, P.; Duguay, G. *Bull. Soc. Chim. Fr.*, **1991**, *128*, 760-772.

5 a) Hudhomme, P.; Toupet, L.; Duguay, G. *Tetrahedron: Asymmetry*, **1990**, *1*, [9], 611-620. b) Boussoufi, A.; Hudhomme, P.; Hitchcock, P.; Duguay, G. *Tetrahedron: Asymmetry*, **1991**, *2*, [2], 157-164. c) Bakasse, M.; Reliquet, A.; Reliquet, F.; Duguay, G.; Quiniou, H. *J. Org. Chem.*, **1989**, *54*, [12], 2889-2893. d) Bakasse, M.; Duguay, G. *Sulfur Lett.*, **1990**, *11*, [3], 123-128.

6 Shafran, Y.M.; Bakulev, V.A.; Mokrushin, V.S. *Russian Chem. Reviews*, **1989**, *58*, 250-274.

7 a) Liu, Hsing-Jang; Chan, Wing Hong; Ping Lee, Sing. *Tetrahedron Lett.*, **1978**, *46*, 4461-4464. b) Garcia, T.; Arrieta, A.; Palomo, C. *Synth. Commun.*, **1982**, *12* , [9], 681-690.

8 a) Helmchen, G.; Nill, G.; Flockerzi, D.; Schühle, W.; Youssef, M. S. K. *Angew. Chem. Int. Ed. Engl.*, **1979**, *18*, [1], 62-65 and Helmchen, G.; Nill, G. *Angew. Chem. Int. Ed. Engl.*, **1979**, *18*, [1], 65-66. b) Beyerman, H. C.; Maassen van den Brink, W. *Proceedings*, **1963**, 266.

9 a) Openshaw, H. T.; Whittaker, N. *J. Chem. Soc. (C)*, **1969**, 89-91. b) Gardner Swain, C.; Brown Jr, J. F. *J. Amer. Chem. Soc.*, **1952**, *74*, 2538-2543.

10 Hudhomme, P.; Duguay, G. *Tetrahedron*, **1990**, *46*, [15], 5263-5272.

11 Parker, D. *Chem. Rev.*, **1991**, *91*, 1441-1457.

12 a) Calmes, M.; Daunis, J.; Jacquier, R.; Verducci, J. *Tetrahedron*, **1987**, *43*, [10], 2285-2292. b) Calmes, M.; Daunis, J.; Elyacoubi, R.; Jacquier, R. *Tetrahedron: Asymmetry*, **1990**, *1*, [5], 329-334.

13 Chehna, M.; Lees, M.; Ali Riahi, M.; Bujoli, B.; Duguay, G.; Quiniou, H. *C. R. Acad. Sc. Fr.*, **1983**, *297, Série II*, 663-666.