ORGANIC LETTERS

2000
Vol. 2, No. 16
2431−2434

Determination of Absolute Configurations of Amines and Amino Acids Using Nonchiral Derivatizing Agents (NCDA) and Deuterium NMR

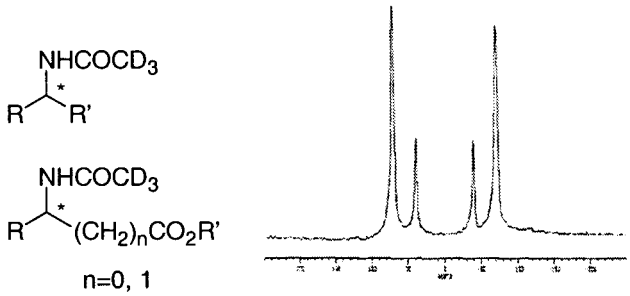
Pierre Chalard, Magali Bertrand, Isabelle Canet,* Vincent Théry, Roland Remuson, and Georges Jeminet

Laboratoire SEESIB, UMR CNRS 6504, Université Blaise Pascal, 63177 Aubière Cedex, France

icanet@chimtp.univ-bpclermont.fr

Received May 16, 2000

ABSTRACT



Enantiomeric analysis and empirical determination of the absolute configuration of amines and amino acids can be easily performed using acetyl-d_3 chloride as a nonchiral derivatizing agent (deuterium probe) and deuterium NMR in a chiral solvent (Courtieu's method). In the case of amino acids, derivatization to amido esters, performed with methanol-d_4 and acetyl-d_3 chloride, gives a double opportunity for enantiomeric analysis.

One of the most useful methods for the determination of the absolute stereochemistry of organic compounds in solution is based on NMR techniques, using chiral derivatizing agents (CDA).[1] In the specific cases of amines and amino acids, some new reagents[2] have been developed since the introduction of MTPA (Mosher)[3a] and MPA (Trost),[3b] although MTPA remains the most efficient agent.[3c] This method is limited however since both derivatives (from (*R*)- and (*S*)-MTPA) must be prepared, and assignment of configuration is based on small $\Delta\delta^{RS}$ shifts from complex mixtures of amide conformers.[1]

On the other hand, we have previously reported a convenient and general method for enantiomeric analysis through deuterium NMR in a chiral liquid crystal solvent (composed of poly-γ-benzyl-L-glutamate PBLG and dichloromethane in a 12% w/w ratio).[4] In this anisotropic medium, enantiomeric differentiation is achieved through the appear-

(1) For reviews, see: Uray, G. In *Houben-Weyl: Methods in Organic Chemistry*; Helmchen, G., Hoffmann, R. W., Mulzer, J., Schaumann, E., Eds.; Thieme: Stuttgart, 1996; Vol. 1, p 253. Eliel, E. L. In *Stereochemistry of Organic Compounds*; Wilen, S. H., Mander, L. N., Eds.; Wiley-Interscience: New York, 1994.

(2) (a) Lopez, B.; Quiñoa, E.; Riguera, R. *J. Am. Chem. Soc.* **1999**, *121*, 9724−9725. (b) Seco, J. M.; Quiñoa, E.; Riguera, R. *J. Org. Chem.* **1999**, *64*, 4669−4675. (c) Chinchilla, R.; Falvello, L. R.; Najera, C. *J. Org. Chem.* **1996**, *61*, 7285−7290. (d) Hoye, T. R.; Rennes, M. K. *J. Org. Chem.* **1996**, *61*, 8489−8495.

(3) (a) Sullivan, G. R.; Dale, J. A.; Mosher, H. S. *J. Org. Chem.* **1973**, *38*, 2143−2147. (b) Trost, B. M.; Bunt, R. C.; Pulley, S. R. *J. Org. Chem.* **1994**, *59*, 4202−4205. (c) Seco, J. M.; Latypov, S. K.; Quiñoa, E.; Riguera, R. *J. Org. Chem.* **1997**, *62*, 7569−7574.

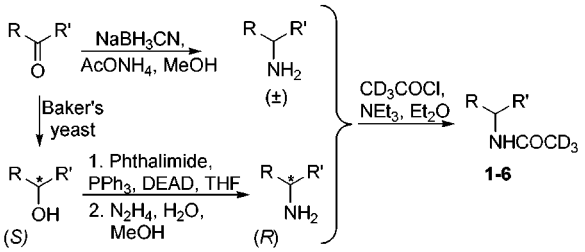
(4) (a) Meddour, A.; Canet, I.; Loewenstein, A.; Péchiné, J. M.; Courtieu, J. *J. Am. Chem. Soc.* **1994**, *116*, 9652−9656. (b) Canet, I.; Courtieu, J.; Loewenstein, A.; Meddour, A.; Péchiné, J. M. *J. Am. Chem. Soc.* **1995**, *117*, 6520−6526.

10.1021/ol006066g CCC: $19.00
Published on Web 07/06/2000

ance of two doublets centered at the same frequency, one for each enantiomer, in the ^2H NMR spectrum. As the enantiomeric discrimination depends on sole interactions with the chiral solvent but requires isotopic labeling, we subsequently proposed an improved method for enantiomeric analysis, using deuterated nonchiral derivatizing agents as a simple way of incorporating deuterium in the substrates.[5] The first application concerned amino acids, on which esterification by methanol-d_4 allowed enantiomeric analysis. However, this procedure required the stabilization of the amino ester derivative as an iminoester by a second reaction step with benzophenone imine.[5a] More recently, we demonstrated the enantiomeric analysis of primary and secondary amines, using acetyl-d_3 chloride as NCDA.[5b] At the same time, Courtieu et al. used perdeuterated benzoyl chloride as the derivatizing agent for alcohols, amines, and amino acids.[5c] As spectral discrimination between enantiomers of acetamides-d_3 was particularly pronounced and therefore suitable for enantiomeric analysis, we decided to extend the use of acetyl chloride to amino acids and to evaluate the ability of this technique to establish the absolute configuration of these substrates, together with amines.

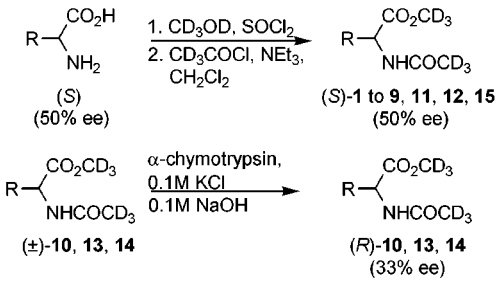
Racemic primary amines, if not commercially available, were synthesized from the corresponding ketones by reductive amination using sodium cyanoborohydride and ammonium acetate[6] (Scheme 1). Optically active amines were

Scheme 1



synthesized in three steps from the ketones by subsequent bakers' yeast reduction,[7] Mitsunobu reaction with phthalimide, and then hydrazinolysis.[8] Derivatization of racemic and optically active amines (ee created by weight of both components) was achieved using acetyl-d_3 chloride in the presence of triethylamine in ether.[5b] In the case of amino acids, we decided to perform a double derivatization, esterification and *N*-acetylation, to compare the ability of methanol-d_4 and acetyl-d_3 chloride to act as NCDA for enantiomeric analysis. Therefore, amino acids were quantitatively doubly derivatized in two steps: esterification using methanol-d_4 in the presence of thionyl chloride (1.2 equiv) followed by reaction with acetyl-d_3 chloride (1.1 equiv) and triethylamine (2.2 equiv) in dichloromethane (Scheme 2).

Scheme 2



For natural amino acids, optically active mixtures were prepared by weighing racemic and L-compounds. For nonnatural ones, enrichment in the D-isomer was performed from the racemic amido esters using α-chymotrypsin:[9] hydrolysis, monitored by 0.1 M NaOH addition, was stopped after half-consumption of the L-ester, to obtain 33% ee. Synthesis of β-amino acids is described elsewhere.[10]

We recorded the proton-decoupled deuterium NMR spectra of amides **1**−**6** and of amido esters **7**−**19**, dissolved in PBLG−CH_2Cl_2 solvent.[11]

The results obtained for amides are listed in Table 1. As amides contain one type of deuterium, NMR spectra are composed of two doublets, corresponding to the differentiation of enantiomers (Figure 1). For all the amides, enanti-

Table 1. Values of Quadrupolar Splittings for Enantiomers of Amides **1**−**6**

NHCOCD3 R1 R2			ee (%)	abs. conf.[a]	vol. abs. conf.[b]	$\Delta\nu_Q^R$ (Hz)	$\Delta\nu_Q^S$ (Hz)	$\Delta\nu_Q^R-\Delta\nu_Q^S$ (Hz)
R_1	R_2							
Ph	CH_3	**1**	0	-	-	82	8	74
			96	(*R*)	(S_v)	71	9	62
CH_2Ph	CH_3	**2**	48	(*R*)	(S_v)	59	14	45
Ph	C_3H_7	**3**	35	(*R*)	(R_v)	159	93	66
C_6H_{11}	CH_3	**4**	52	(*R*)	(S_v)	131	68	63
C_5H_{11}	CH_3	**5**	50	(*R*)	(S_v)	142	79	63
$(CH_2)_2$-$OSiMe_3$	CH_3	**6**	0	-	-	139	109	30
			100	(*R*)	(S_v)	150	-	-

[a] Absolute configurations were determined by comparison of the optical rotation of alcohols and amines with those reported in the litterature. [b] Volumic absolute configurations were defined following Cahn−Ingold−Prelog rules, using substituent hierarchy based on the van der Waals volumes (ref 13) instead of the atomic numbers.

(5) (a) Canet, I.; Meddour, A.; Courtieu, J.; Canet, J. L.; Salaün, J. *J. Am. Chem. Soc.* **1994**, *116*, 2155−2156. (b) Canet, J. L.; Canet, I.; Courtieu, J.; Da Silva, S.; Gelas, J.; Troin, Y. *J. Org. Chem.* **1996**, *61*, 9035−9037. (c) Meddour, A.; Loewenstein, A.; Péchiné, J. M.; Courtieu, J. *Tetrahedron: Asymmetry* **1997**, *8*, 485−494.

(6) Borch, R. F.; Bernstein, M. D.; Dupont Durst, H. *J. Am. Chem. Soc.* **1971**, *93*, 2897.

(7) Besse, P.; Bolte, J.; Fauve, A.; Veschambre, H. *Bioorg. Chem.* **1993**, *21*, 342−353.

(8) Roush, W. R.; Straub, J. A.; Brown, R. J. *J. Org. Chem.* **1987**, *52*, 5127−5136. Heatcock, C. H.; Davidsen, S. K.; Mills, S. G.; Sanner, M. A. *J. Org. Chem.* **1992**, *57*, 2531−2544.

(9) Bernasconi, S.; Corbella, A.; Gariboldi, P.; Jommi, G. *Gazz. Chim. Ital.* **1977**, *107*, 95−99.

(10) Chalard, P.; Remuson, R.; Gelas-Mialhe, Y.; Gramain, J. C. *Tetrahedron: Asymmetry* **1998**, *24*, 4361−4368. Chalard, P.; Remuson, R.; Gelas-Mialhe, Y.; Gramain, J. C.; Canet, I. *Tetrahedron Lett.* **1999**, *40*, 1661−1664.

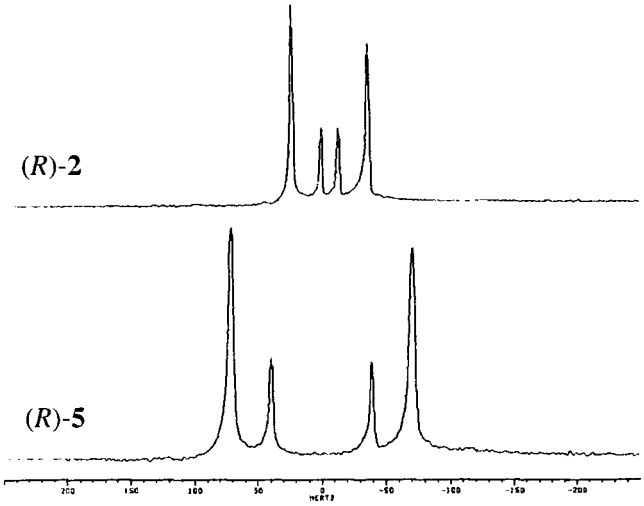


Figure 1. ^{2}H NMR spectra of amides **2** and **5**.

omers are well distinguished in their deuterium NMR spectra, with good resolution,[12] and differences between the quadrupolar splittings are large enough to allow an accurate measurement of the enantiomeric excess. Furthermore, the splittings of the (*R*)-amides are always larger than those of the (*S*) ones (see ($\Delta\nu^R - \Delta\nu^S$) in Table 1).

The same relation between splittings can be made using volumic absolute configurations[13] ((S_V) larger than (R_V)), except for amide **3** for which the larger splitting is observed for the (R_V) isomer. However, volumes obtained from Bondi's increments [$V(C_3H_7)$ = 56.7 A^3/V(NHCOCH$_3$) = 55.2 A^3; ^{2}H is not available in ref 13] or from molecular modeling[14] are rather close, and consequently, the attribution of the volumic absolute configuration is uncertain.

Results of the proton-decoupled deuterium NMR spectra of amido esters **7**−**19** are listed in Table 2. For doubly derivatized amido esters, NMR spectra are expected to present two sets of two doublets, corresponding to the differentiation of enantiomers via the ester and the amide functions. In these cases, the difference in chemical shifts is large enough to separate the outside signals (Figure 2).

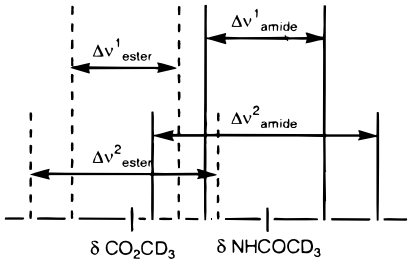
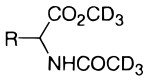
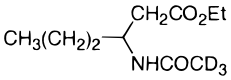
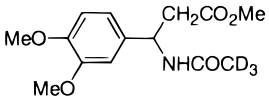


Figure 2. Theoretical ^{2}H NMR spectrum of a doubly derivatized amido ester (33% ee).

For all amido esters, enantiomers are well distinguished on the amido part of the spectra (Figure 3). For the ester moiety, the differentiation of enantiomers is obtained in most cases but it has to be noted that the separation between these doublets is generally smaller. As the discrimination originates from a difference of ordering between enantiomers, an amido group, more rigid than an ester, seems more suitable for an

Table 2. Values of Quadrupolar Splittings for Enantiomers of Amido Esters **7**−**19**

amido ester			ee	abs. conf.	vol. abs. conf.	ester			amide		
			(%)			$\Delta\nu_Q^R$ (Hz)	$\Delta\nu_Q^S$ (Hz)	$\Delta\nu_Q^R$-$\Delta\nu_Q^S$ (Hz)	$\Delta\nu_Q^R$ (Hz)	$\Delta\nu_Q^S$ (Hz)	$\Delta\nu_Q^R$-$\Delta\nu_Q^S$ (Hz)
R=	CH_3	**7**	50	(*S*)	(S_V)	146	114	32	121	28	93
	Ph	**8**	50	(*S*)	(S_V)	99	45	54	90	62	28
	CH_2Ph	**9**	50	(*S*)	(S_V)	101	76	25	150	86	64
	CH_2CH_3	**10**	33	(*R*)	(R_V)	110	88	22	118	14	104
	$(CH_2)_3CH_3$	**11**	50	(*S*)	(S_V)	67	67	0	170	113	57
	$CH_2CH(CH_3)_2$	**12**	50	(*S*)	(S_V)	70	52	18	158	86	72
	$(CH_2)_5CH_3$	**13**	33	(*R*)	(R_V)	60	83	- 23	240	193	47
	CH_2OCH_3	**14**	33	(*R*)	(R_V)	42	52	- 10	119	55	64
	$(CH_2)_2SCH_3$	**15**	50	(*S*)	(S_V)	87	87	0	154	86	68
	$CH_2CO_2CH_3$	**16**	50	(*S*)	(S_V)	103	71	32	135	95	40
	$(CH_2)_2CO_2CH_3$	**17**	50	(*S*)	(S_V)	-	-	-	153	110	43
		**18**	-	(±)		-	-	-	277	178	99
				(*R*)	(R_V)	-	-	-	279	174	105
		**19**	-	(±)		-	-	-	218	194	24
				(*S*)	(S_V)	-	-	-	234	205	29

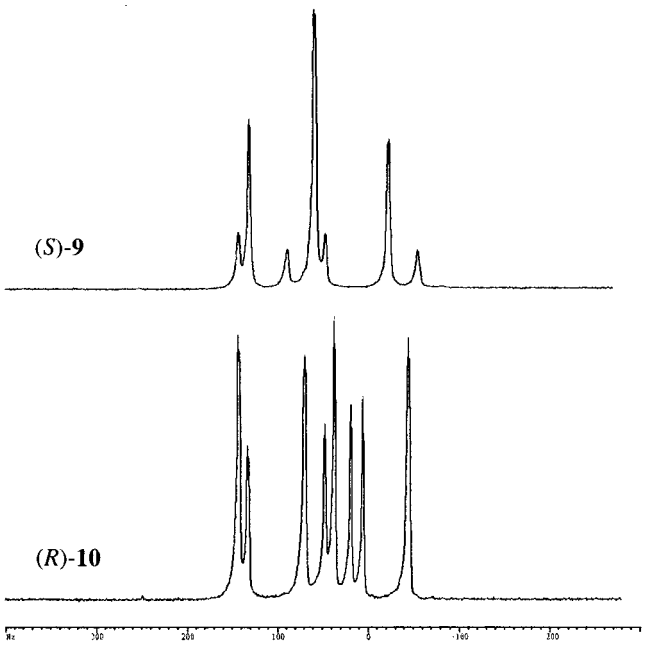


Figure 3. ^{2}H NMR spectra of amido esters **9** and **10**.

optimal difference of quadrupolar splittings. Concerning the determination of absolute configurations, as for amides **1**−**6**, the splittings of the amido group of the (*R*)-amido esters are always larger than those for the (*S*) compounds. For the ester group, this relation does not always obtain (e.g., see **13** and **14** in Table 2).

Concerning volumic absolute configurations, some comments can be made: (R_V)-amido splittings are greater than (S_V) ones. Splittings of the ester part are not related to volumic absolute configurations. Furthermore, comparing these results with those obtained for amides, we note an inversion in the position of the doublets, as outside doublets correspond to (S_V)-amides and to (R_V)-amido esters. From these observations, some questions arise: (1) For amido esters, calculated van der Waals volumes of CO_2CH_3 and $NHCOCH_3$ are rather close.[13] From molecular modeling, differences in the volumes of ester and amide are small (Table 3), within confidence limits, and invert depending on the conformation. It is consequently difficult to assume that (*R*)-amido esters are (R_V) compounds. (2) Using a definition of absolute configurations based on van der Waals volumes, we assume that the main factor for enantiomeric discrimination is of a geometrical nature. However, this definition does not take into account the solute conformations in the liquid crystal solvent. We therefore calculated Connolly areas, which represent the accessible areas of the substituents to the solvent. As shown in Table 3, we obtained areas for which priority orders were sometimes inverted compared to the volumes. This first approach, obtained by a model in the gas phase, illustrates the complexity of the problem of finding a general method for attribution of absolute configuration by NMR.

Table 3. van der Waals Volumes and Connolly Areas for Some Amido Esters[13]

amino ester	van der Waals volumes (A^3)			vol. abs. conf.	Connolly areas (A^2)			area abs. conf.	ΔE
	ester	amide	C_4H_9		ester	amide	C_4H_9		
(*S*)-**11**	48.0	63.4	81.8	S_V	54.4	64.5	90.3	S_A	0.0
	56.2	55.9	82.2	R_V	61.3	60.2	89.5	R_A	0.2
	55.2	54.7	80.4	R_V	56.6	59.4	82.1	S_A	0.5
	ester	amide	C_3H_7		ester	amide	C_3H_7		
(*R*)-**18**	98.8	47.5	84.1	R_V	89.7	58.2	84.1	R_A	0.0
	92.6	37.2	90.1	R_V	94.9	62.8	62.6	S_A	0.5
	96.0	59.1	61.0	R_V	92.0	56.4	65.4	R_A	0.7

In summary, this study has shown that acetyl-d_3 chloride is a convenient nonchiral derivatizing agent for the enantiomeric analysis of amines and amino acids through ^{2}H NMR. In the case of amino acids, a double derivatization, adding methanol-d_4 as a second NCDA, gives the organic chemist two opportunities for an accurate measurement of the enantiomeric excess of these compounds. For the attribution of the absolute configuration, ^{2}H NMR spectra of acetyl-d_3 amides allow an empirical determination, as splittings of (*R*)-derivatives of amines and amino acids are larger than those obtained for (*S*)-ones. To understand the relation between molecular structure and NMR splitting effects, we are currently modeling the behavior of the solutes in interaction with the PBLG−CH_2Cl_2 solvent. This study could allow a generalization of the empirical rule for determination of absolute configurations by NMR.

Supporting Information Available: Experimental procedures for derivatization of amines and amino acids and for NMR procedures. This material is available free of charge via the Internet at http://pubs.acs.org.

OL006066G

(11) Experimental details are available as Supporting Information.
(12) Using 5 mm tubes in a 10 mm probe leads to 5−10 Hz line widths. Better resolution (1−3 Hz) is obtained using a selective 5 mm deuterium probe (see ref 4).
(13) Bondi, A. *J. Phys. Chem.* **1964**, *68*, 441−451.
(14) Obtained from Sybil 6.3, TRIPOS INC., Molcad software.