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Determination of the enantiomeric composition of chiral delta-2-thiazolines-1,3 by ^{1}H and ^{19}F NMR spectroscopy using chiral solvating agents

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Abstract. Studies of the perturbing effect of chiral solvating agents (CSAs) namely the fluoroalcohols **5a** and **5b** upon the NMR spectra of chiral Δ^2-thiazolines **1** presenting interesting insecticidal properties demonstrated the ability of these CSAs to afford diastereomeric solvates from these substrates providing their enantiomeric discrimination. Thus, for five of the six tested Δ^2-thiazolines **1A** and **1B** there is at least one possibility to proceed to their enantiomeric discrimination either by ^{1}H or ^{19}F NMR using mostly **5b** as CSA.

Keywords: Δ^2-thiazolines-1,3, stereomer, enantiomer, chiral solvating agent, optical purity analysis

1. Introduction

For several years, in a proinsecticide perspective, we have been working to develop molecules susceptible to afford a reversible masking of carboxylic acids and/or β-ethanolamines selected as active principles. Among several candidate structures subjected to a first biological screening: enols esters [1], N-acyl aziridines [2–4], Δ^2-oxazolines-1,3 [5,6] and Δ^2-thiazolines 1-3 [7,8], the two last series appeared as the most promising ones and were therefore more thoroughly investigated. Due to the efficient unmasking of the carboxylic acids evidenced during *in vitro* monitoring in insect tissues [5–8] these heterocycles were confirmed as proactive compounds. Moreover, the mechanism of the enzymatic hydrolysis of oxa- [5,6] and thiazolines [7] triggered in such conditions is now well-documented (see Fig. 1A) and the hypothesis of an intermediate **I** (X = O) of the β-ethanolamine ester type has clearly been established for oxazolines [2,5,7]. Taking into accounts the involvement of this intermediate, one could expect that insects strongly provided in esterases should be more sensitive to oxazolines as previously shown for a series of esters [9]. Therefore two strains of aphid *M. persicae* selected for their difference in sensitivity towards classical insecticides – the resistance of the mutant strain being known as the result of a

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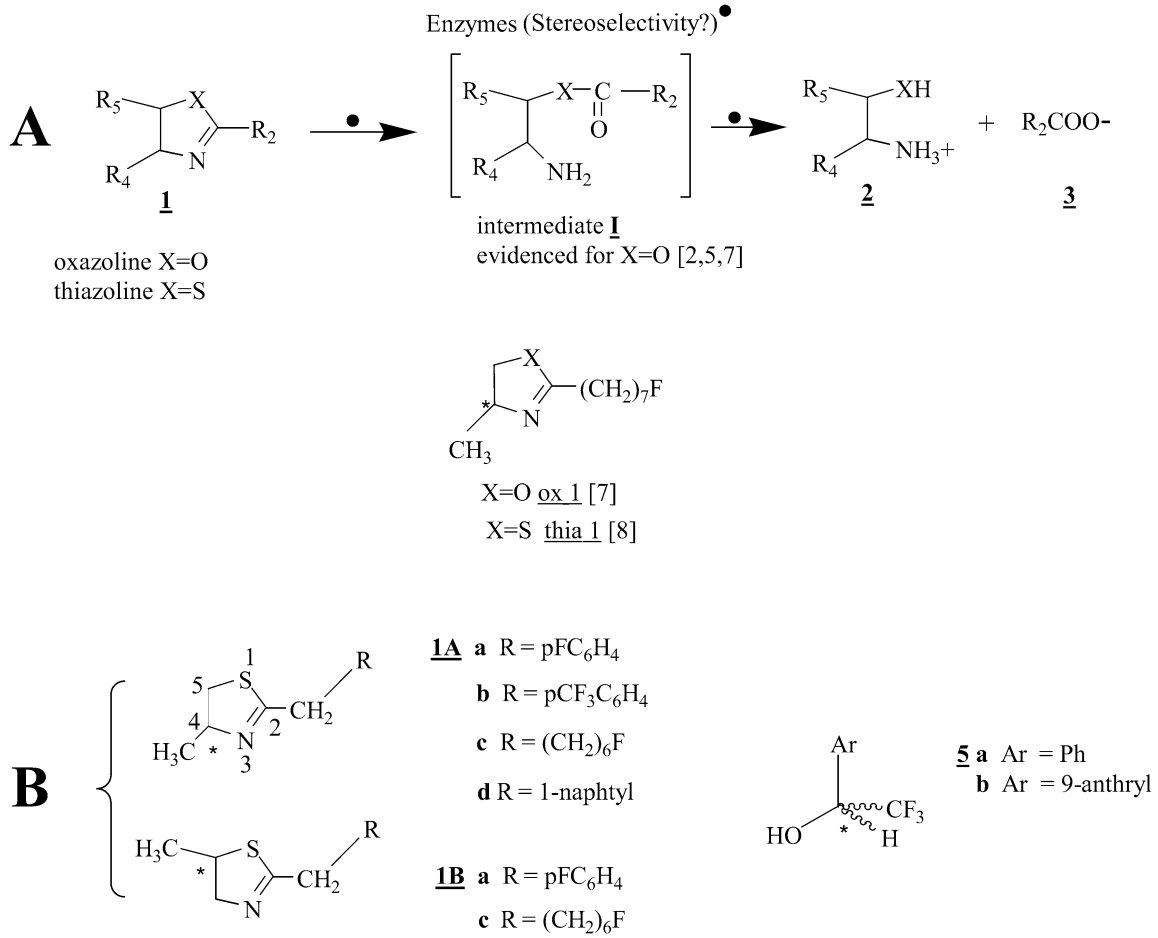


Fig. 1. **A**: The enzymatic unmasking of carboxylate from oxazolines (X = O) [2,5,7] and thiazolines (X = S) [8] in locust tissues. **B**: Structure of chiral thiazolines **1** and chiral solvating agent CSA **5** used in this work. The asterisk denotes the stereogenic center.

reinforcement in esterases [10,11] – were subjected to assays against the fluorinated oxazoline **ox 1** (the most active of the series, see Fig. 1A) [12]. As expected, it appeared that **ox 1** presents a lesser LD_{50} against the resistant strain (about 3.5 folds lesser) comparatively to that observed for the sensitive strain [7,12]. Similar assays conducted with the fluorinated thiazoline **thia 1** (see Fig. 1A) resulted in the same observation of a lesser LD_{50} of **thia 1** against the resistant strain (about 3.7 folds lesser) [8,12], which indirectly confirms the hypothesis [8] of an intermediate **I** (X = S) of the β-thio-ethanolamine ester type during the hydrolysis of thiazolines (see Fig. 1A).

These two results not only constitute a good example of crossed negative resistance (CNR) [13,14] but also they valide our proinsecticide approach that can result in a significant diminishing of LD_{50} through successive and fruitful inter-relations between biological testings and metabolism.

In an effort to pursue the improvement of these series we are presently investigating possible impact of the chirality of our oxazolines and thiazolines from both the standpoints of their biological activities – by independent testing of the enantiomers – and of their metabolism.

The present paper deals with the last standpoint in the case of the thiazoline series. In fact, the enzymatic character of their hydrolysis raises the question of a possible chiral recognition during the unmasking of these chiral proinsecticides triggered by biological tissues of locust. As a matter of fact, enantiomers can interact differently with enzymes that are themselves chiral. In our group, a remarkable enantioselectivity has previously been observed in such tissues [15] during the unmasking of chiral esters studied independently with pure enantiomers.

Thus, we wanted to develop an analytical method allowing a determination of the enantiomeric composition of chiral thiazolines **1Aa–d**, **1Ba** and **1Bc** presenting a chiral center in their masked β-thioethanolamine moiety **2** (see Fig. 1B) during the enzymatic hydrolysis of their racemate. In fact with these compounds we had the preoccupation to spare the stocks of enantiomeres in view of their biological testings.

Recently, working on racemates, we succeeded in the enantiomeric discrimination of chiral oxazolines by ^{1}H or ^{19}F NMR analysis using chiral lanthanide shift reagents (CLSRs) [16] or chiral solvating agents (CSAs) of the trifluoromethyl carbinol type [17] **5** (see Fig. 1B) that have provide easier NMR analysis [18].

If the basic principles for use of CSAs have previously been intensively discussed in reviews compiling also their numerous applications in enantiomeric purity determination [19–22], considerable effort is still dedicated to the study of labile diastereomers and the preparation of more general and/or stronger CSAs [23–27].

Therefore, taking into accounts the presence of two basic centers in thiazolines as in oxazolines, we developed a similar analytical NMR method as for oxazolines [18], expecting the ability of thiazolines to also interact with acidic CSAs **5**.

2. Material and methods

2.1. Chemicals

Chiral thiazolines α-N-substituted **1Aa–d** and α-S-substituted **1Ba** and **1Bc** were prepared, either as racemates or as pure enantiomers, starting from the corresponding β-hydroxylamides **6A** and **6B**, respectively, using Lawesson reagent [28] in a one-pot reaction that entails both the substitution and the cyclisation of the starting material [29] (see Fig. 2). Structural characterization (^{1}H and ^{13}C NMR, IR and MS) of racemates and enantiomers of thiazolines **1A** and **1B** and of their corresponding precursors **6A** and **6B** that agree well with the proposed structures will be published elsewhere [30] (see also Table 1

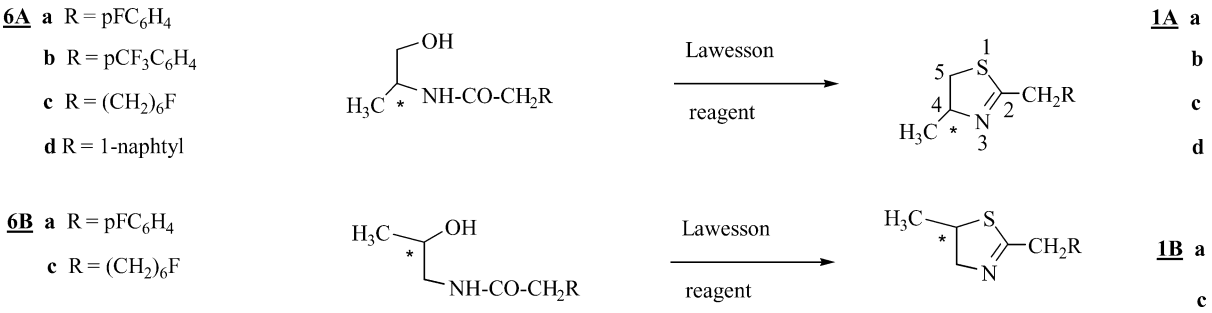
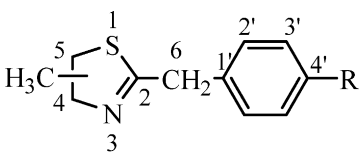
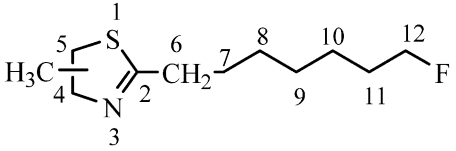


Fig. 2. Preparation of thiazolines **1A** and **1B** starting from the corresponding hydroxylamides **6A** and **6B**.

Table 1
Achiral solvent effects on the 300 MHz ^{1}H NMR spectra of Δ^2-thiazolines-1,3 **1**





δ ppm (multiplicity)		CH_3–C_4	CH_3–C_5	H_4	$H_{4'}$	H_5	$H_{5'}$	H_6	H_7/H_{11}	$H_8H_9H_{10}$	H_{12}	$H_{2'}$[d]	$H_{3'}$[d]
1Aa	$CDCl_3$[a]	1.37 (d, 6.5)	–	4.57 (m = ddqt)	–	3.41[e] (dd, 10.8, 8.4)	2.91[e] (dd, 10.8, 7.5)	3.79 (bs)	–	–	–	7.26 (m)	7.02 (pt, 8.8)
	C_6D_6[a]	1.09 (d, 6.6)	–	4.23 (m = ddqt)	–	2.80 (dd, 10.8, 8.3)	2.37 (dd, 10.8, 7.3)	3.53 (bs)	–	–	–	6 96 (m)	6.73 (pt, 8.8)
1Ba	$CDCl_3$[b]	–	1.29 (d, 6.7)	4.19 (m = 4 ddt, 14.9, 7.9, 1.6)	3.96 (m = 4 ddt, 14.9, 4.4, 1.2)	3.89 (m)	–	3.79 (bs)	–	–	–	7.24 (m)	7.00 (pt, 8.8)
	C_6D_6[b]	–	0.86 (d, 6.8)	∼3.88 (m = ddt, 15.2, 7.9, 1.5)	∼3.68 (m = ddt, 15.2, 4.3, 1.1)	3.30 (m)	–	3.55 (bs)	–	–	–	6.96 (m)	6.74 (pt, 8.8)
1Ab	$CDCl_3$[a]	1.37 (d, 6.7)	–	4.59 (m = ddqt)	–	3.43 (dd, 10.9, 8.5)	2.93 (dd, 10.9, 7.5)	3.87 (bs)	–	–	–	7.41 (m)	7.59 (m)
	C_6D_6[a]	1.07 (d, 6.7)	–	4.21 (m = ddqt)	–	2.79 (dd, 10.8, 8.4)	2.36 (dd, 10.8, 7.4)	3.48 (bs)	–	–	–	6.98 (m)	7.26 (m)
1Ac	$CDCl_3$[a]	1.35 (d, 6.7)	–	4.55 (m = ddqt)	–	3.41 (dd, 10.8, 8.3)	2.90 (dd, 10.8, 7.6)	2.50 (pt, 7.6)	1.60–1.80 (m, 4H)	1.20–1.45 (m, 6H)	4.43 (dt, 47.3, 6.1)	–	–
	C_6D_6[a]	1.15 (d, 6.6)	–	4.33 (m = ddqt)	–	2.86 (dd, 10.8, 8.4)	2.42 (m)	2.42 (m)	1.23–1.45 (m, 2H) 1.50–1.63 (m, 2H)	1.00–1.20 (m, 6H)	4.07 (dt, 47.3, 6.1)	–	–

Table 1
(Continued)

	δ ppm (multiplicity)	CH_3–C_4	CH_3–C_5	H_4	$H_{4'}$	H_5	$H_{5'}$	H_6	H_7/H_{11}	$H_8H_9H_{10}$	H_{12}	$H_{2'}$[d]	$H_{3'}$[d]
1Bc	$CDCl_3$[c]	–	1.35 (d, 6.5)	4.17 (m = ddt)	∼3.92 (m)	∼3.92 (m)	–	2.57 (pt, 7.6)	1.60–1.80 (m, 4H)	1.20–1.45 (m, 6H)	4.43 (dt, 47.3, 6.1)	–	–
	C_6D_6[b]	–	0.92 (d, 6.9)	∼3.89 (m = ddt)	∼3.70 (m = ddt)	3.31 (m)	–	2.47 (pt, 6.7)	1.20–1.40 (m, 2H) (1.50–1.70) (m, 2H)	1.00–1.20 (m, 6H)	4.05 (dt, 47.3, 6.2)	–	–
1Ad	$CDCl_3$[a]	1.36 (d, 6.6)	–	4.61 (m = ddqt)	–	3.35 (dd, 10.8, 8.3)	2.86 (dd, 10.8, 7.3)	4.29 (bs)	–	–	–	H arom. (m, 7H)	7.40–8.20
	C_6D_6[a]	1.07 (d, 6.7)	–	4.24 (m = ddqt)	–	2.72 (dd, 10.8, 8.6)	2.29 (dd, 10.8, 6.8)	4.15 (bs)	–	–	–	H arom. (m, 7H)	7.20–8.30

[a] H_4, H_5 and $H_{5'}$ protons constitute an AMX system which is complicated for the X part (H_4) by a ^{3}J coupling with CH_3–C_4 and a ^{5}J coupling of the homoallylic type with H_6 protons.

[b] H_4, $H_{4'}$ and H_5 protons constitute an ABX system also complicated for H_4 proton (by a ^{5}J coupling with H_6 protons) and for H_5 proton (by a ^{3}J coupling with CH_3–C_5). This ABX system was not studied neither by the sub-spectra method nor by simulation. However, the chemical shifts were approximated as the multiplets centers. As a matter of fact, this approximation was evidenced in the case of corresponding oxazolines[9] as entailing only ∼0.1 Hz error on the frequencies. The couplings were assigned by reference to the values obtained from calculated spectra of oxazolines and from observed values for thiazolines of the **1Ax** type.

[c] H_4, $H_{4'}$ and H_5 protons constitute an A_2X system.

[d] $H_{2'}$ and $H_{3'}$ protons constitute an $AA'XX'$ system which is complicated in the cases of **1Aa** and **1Ba** by ^{3}J and ^{2}J couplings with the ^{19}F nucleus, respectively. Therefore $H_{3'}$ appears as a pseudotriplet (pt).

[e] The pre-irradiation of the CH_3–C_4 entailed a significant NOE of the most shielded signal allowing its attribution to $H_{5'}$ i.e. to the proton in configuration cis relative to CH_3.

for ^{1}H NMR data). Thiazolines were stored under argon at 255 K to minimize their slow hydrolysis into the corresponding thio-hydroxy analogous of hydroxylamides **6A** and **6B**.

Chiral CSAs: (R)–2,2,2-trifluoro-1-phenylethanol **5a** and (R)–2,2,2-trifluoro-1-(9-anthryl)ethanol **5b** were obtained from Fluka (Saint-Quentin-Favallier, France). Deuterated solvents C_6D_6 and $CDCl_3$ were obtained from Euriso-Top (Gif-Sur-Yvette, France). The solvents were stored on 3 Å molecular sieves. CSA **5a** was stored in a desiccator over P_2O_5 and CSA **5b** under argon at 277 K.

2.2. Spectroscopic studies

Spectra were recorded on a 300 MHz (^{1}H) or 282 MHz (^{19}F) AC 300 Bruker equipped with a 5 mm probe and a Bruker Aspect 3000 computer.

For ^{1}H NMR spectra the following abbreviations were used for the spectra description: s = singlet; bs = broad singlet; d = doublet; dd = doublet of doublet; t = triplet; q = quadruplet; m = multiplet, and p for pseudo i.e., pt = pseudo triplet.

Typical conditions for recording one-dimensional spectra were as follows: spectral width 5376 Hz (^{1}H) or 5917 Hz (^{19}F), data points 32 K (^{1}H) or 64 K (^{19}F), flip angle 30° (^{1}H) or 60° (^{19}F), pulse repetition 1 s (^{1}H) and (^{19}F), acquisition time 3.047 s (^{1}H) or 5.832 s (^{19}F). Resolution enhancement was performed by using a gaussian window. The Fourier transform was carried out with 65 K (^{1}H) or 130 K (^{19}F), after zero filling so that the digital resolution was 0.17 Hz per point (^{1}H) or 0.08 Hz per point (^{19}F).

The different thiazolines **1Aa–d**, **1Ba** and **1Bc** were dissolved either in a mixture of deuterated chloroform/carbon tetrachloride (30/70, v/v), or in deuterated benzene to give concentrations ranging from 1.1 to $1.4 \cdot 10^{-2}$ mole·l^{-1}. Measurements were mostly carried out at a probe temperature of 296 ±1 K and the proton chemical shifts were initially referenced to the solvent values of 7.16 and 7.27 ppm (RMN ^{1}H in C_6D_6 and $CDCl_3$, respectively). For ^{19}F[^{1}H] spectra, the fluorine chemical shifts are reported in ppm relative to external reference CCl_3F (5% in C_6D_6 v/v).

In runs with CSAs **5a** and **5b**, the solid reagent was accurately weighed and rapidly introduced into a vial containing an aliquot of standard solution of thiazoline **1**.

When enantiomeric shift differences were observed for selected resonances in the presence of CSAs, the following abbreviations $\Delta\nu$, $\Delta\delta$ and $h\%$ represent, respectively: $|\nu_R - \nu_S|$ expressed in Hz, the average value of the chemical shifts $|(\Delta\delta_S + \Delta\delta_R)/2|$ and (valley height/average peak height) × 100 of a signal obtained with CSA, see Table 1 for initial chemical shifts without CSA.

2.3. Precision on e.e. determination

A solution of racemate **1Ab** (1.05 mg) was enriched with S enantiomer (1.54 mg), the samples being weighted on a precision balance (±0.01 mg). The theoretical e.e. for **1Ab** was 59.46%. Using typical conditions for recording one-dimension spectra and a Gaussian window for resolution enhancement, the e.e. measured by ^{1}H NMR was 60.45 and 60.12% for H_5 ($\Delta\nu$ = 3.2 Hz) and CH_3–C_4 ($\Delta\nu$ = 0.6 Hz), respectively, indicating a precision of about ±1% on the e.e. determination. Similar results were obtained with **1Ac** for H_5 ($\Delta\nu$ = 3.0 Hz), see Table 3. It is noticeable that the precision on the e.e. determination can remain good, despite small values of $\Delta\nu$.

Table 2
Enantiomeric discrimination of ^{1}H NMR and ^{19}F NMR signals of Δ^{2}-thiazolines-1,3 by solvatation with chiral solvating agent CSA **5b** at 296 K

	Achiral solvent	[(**R**)-**5c**] /[**1**]	NMR ^{1}H				^{19}F[^{1}H] NMR			
			Proton of **1**	$\Delta\delta$[a] (ppm)	$\Delta\nu$[b] (Hz)	h%[c]	δ (ppm) without CSA	$\Delta\delta$[a] (ppm)	$\Delta\nu$[b] (Hz)	h%[c]
1Aa	CCl_4/ $CDCl_3$	1/1	**H_5**	**−0.05**	**2.46**	**0**	−116.16	0.11	0	100
			$H_{5'}$	**−0.05**	**2.83**	**0**				
			$H_{3'}$	−0.04	1.80	20.5				
			CH_3–C_4	−0.05	1.15	44.0				
	C_6D_6	1/1	**$H_{5'}$**	**−0.06**	**1.83**	**6.8**	−115.85	0.10	0	100
			$H_{3'}$	−0.03	1.02	50.0				
			CH_3–C_4	−0.06	1.11	10.9				
		2/1	**$H_{5'}$**	**−0.12**	**3.45**	**0**		0.18	0.96	63.4
			$H_{3'}$	**−0.05**	**1.80**	**0**				
			CH_3–C_4	**−0.11**	**1.95**	**0**				
		6/1	**H_5**	**−0.22**	**2.14**	**0**		0.31	0.96	41.7
			$H_{5'}$	**−0.21**	**5.41**	**0**				
			$H_{3'}$	**−0.08**	**2.84**	**0**				
			CH_3–C_4	**−0.18**	**4.02**	**0**				
1Ba	C_6D_6	3/1	H_4	−0.21	0.80	35.0	−115.92	0.23	1.38	78.5
		5/1	H_4	−0.27	n.u.[d]	–		0.31	1.89	38.3
		6/1	H_4	−0.29	n.u.	–		**0.33**	**2.07**	**0**
1Ab	C_6D_6	2/1	H_5	−0.07	1.80	23.0	−62.13	−0.02	0.36	81.7
			CH_3–C_4	−0.01	1.00	58.0				
		5/1	**H_5**	**−0.15**	**4.30**	**0**		−0.03	0.75	55.4
			CH_3–C_4	**−0.12**	**2.70**	**6.0**				
		8/1	H_5	−0.15	n.u.	–		−0.03	0.84	78.9
			CH_3–C_4	**−0.13**	**2.7**	**7.0**				
1Ac	CCl_4/ $CDCl_3$	2/1	**H_5**	**−0.06**	**3.10**	**0**	−218.25	0.05	0	100
			H_{12}	**−0.02**	**1.70**	**6.0**				
		5/1	**H_5**	**−0.09**	**4.70**	**3.0**		0.11	0	100
			H_{12}	−0.04	2.30	22.0				
1Bc	C_6D_6	2/1, 5/1, 8/1		n.d.[e]				n.d.		
1Ad	CCl_4/ $CDCl_3$	2/1	**H_5**	**−0.06**	**3.50**	**0**		n.d.		
			$H_{5'}$	**−0.02**	**4.70**	**0**				
			H_6	−0.10	4.60	10.0				
		5/1	**H_5**	**−0.10**	**6.70**	**0**		n.d.		
			$H_{5'}$	−0.08	n.u.	–				
			H_6[f]	**−0.19**	**8.50**	**9.0**				
		8/1	H_5	−0.14	6.90	11.0		n.d.		
			H_6	−0.26	11.60	12.0				
			H_5	−0.07	2.00	8.0				

Table 2
(Continued)

Achiral solvent	[(**R**)-**5c**] /[**1**]	NMR ^{1}H				^{19}F[^{1}H] NMR			
		Proton of **1**	$\Delta\delta^a$ (ppm)	$\Delta\nu^b$ (Hz)	$h\%^c$	δ (ppm) without CSA	$\Delta\delta^a$ (ppm)	$\Delta\nu^b$ (Hz)	$h\%^c$
C_6D_6	2/1	$H_{5'}$	−0.07	1.30	66.0		n.d.		
		H_5	−0.16	n.u.	–				
	5/1	**$H_{5'}$**	**−0.17**	**2.80**	**3.0**		n.d.		
		H_6	−0.16	n.u.	–				
		H_5	−0.20	n.u.	–				
	8/1	$H_{5'}$	−0.22	n.u.	–		n.d.		
		H_6	−0.21	7.70	21.0				

In bold characters are indicated the usable signals for enantiomeric NMR discrimination.
[a] $\Delta\delta$ = mean chemical shift $(\Delta\delta_R + \Delta\delta_S)/2$ induced by CSA. See Table 1 for initial chemical shifts without CSA.
[b] $\Delta\nu = |\nu_R - \nu_S|$.
[c] $h\%$ = (valley height/average peak height) × 100.
[d] n.u.: discrimination not usable due to overlapping either with other signals of thiazoline or with the **5b** hydroxyl group.
[e] n.d.: no discrimination.
[f] The most deshielded signal is splitted into AB system while the shielded signal appears as a quasi A_2 system.

Table 3
Inequivalence sense for the NMR enantiomeric discrimination of thiazolines **1** induced by solvatation with CSA (**R**)-**5b**

	Achiral solvent	[(**R**)-**1**]/ [(**S**)-**1**]	[(**R**)-**5b**]/**1**	Temperature (K)	^{1}H NMR		^{19}F NMR
					Proton	ν_R–ν_S (Hz)	ν_R–ν_S (Hz)
1Aa	C_6D_6	4/1	1/1	296	H_5	+1.97	–
					CH_3	+1.15	
1Ba	C_6D_6	4/1	6/1	296	–	–	+1.78
1Ab	C_6D_6	1/4	5/1	286	H_5	+3.20	−0.54
					CH_3	+0.60	
1Ac	CCl_4/	4/1	2/1	296	H_5	+3.00	–
	$CDCl_3$				H_{12}	−1.60	
1Ad	CCl_4/	4/1	2/1	296	$H_{5'}$	+3.93	–
	$CDCl_3$				H_{12}	+8.70	

The determination is obtained by overloading with one enantiomer of **1**.

3. Results and discussion

3.1. NMR data for thiazolines 1 without CSA

We have been using $CDCl_3$ and C_6D_6 as achiral solvents for recording the 300 MHz ^{1}H NMR spectra of α-N-substituted thiazolines **1Aa–d** and α-S-substituted thiazolines **1Ba** and **1Bc** in order to choose the most simplest spectra as possible. As expected C_6D_6 entails appreciable aromatic solvent induced shifts, ASIS [31], all the signals being significantly shielded compared to their values in $CDCl_3$ solution [30] (see Table 1). With both of these solvents, protons H_5 and $H_{5'}$ of thiazolines **1A** and **1B** are more shielded compared to H_4 and $H_{4'}$, which represents a situation that is exactly reversed relatively to corresponding oxazolines [5,7]. In most cases these three protons constitute an AMX pattern except for **1Ba** (ABX pattern in $CDCl_3$ and C_6D_6 solutions) and **1Bc** (A_2X pattern in $CDCl_3$ solution and ABX pattern in C_6D_6 solution).

A remarkable difference in the geminal coupling can be seen between **1Ba** ($^2J = -14.9$ Hz) and **1Aa** ($^2J = -10.8$ Hz) corresponding to the known increase in J_{gem} resulting from the substitution of the methylene group by electronegative groups [32]. The same trend has been previously observed for corresponding series of oxazolines, though more pronounced according to the stronger withdrawing electronic effect of oxygen compared to sulphur atom [5,7]. To notice is also the ^5J coupling of the homoallylic type between H_4 and H_6 protons previously mentioned for oxazolines [2,5,7,33]. This coupling is sometimes measurable in the $H_{4,4'}$ part of thiazolines spectra but entails, more often a simple broadening of the corresponding signals particularly for H_6. The values observed for this coupling are in the range $\sim$0 to 1.6 Hz and can be different for the geminal H_4 and $H_{4'}$ protons (see Table 1).

3.2. Experiments with trifluorinated carbinols as CSAs

3.2.1. First assays

A first series of assays was performed at 296 K with solutions of thiazolines **1Aa–d**, **1Ba** and **1Bc** in C_6D_6 or $CCl_4/CDCl_3(70/30$, v/v) as achiral and apolar solvent [18], using CSA **5b** with [**5**]/[**1**] ratios ranging from 1 to 8. From ^1H and ^{19}F NMR data presented in Table 2 and with some illustrations in Fig. 3, it appears that:

1. CSA **5b** provides for five of the six thiazolines tested a good enantiomeric anisochrony at least for one of the ^1H or ^{19}F NMR signals, with $\Delta\nu = |\nu_R - \nu_S|$ up to 12 Hz (at 300 MHz) and 2 Hz (at 282 MHz), respectively. Thus, even for the ratio [**CSA**]/[**1**] as low as 1 or 2, the criterion relative height reaches magnitudes in the range: $0 \leqslant h\% \leqslant 10\%$. Such values made the method usable for quantitation without deconvolution of the ^1H NMR signals, for H_5, $H_{5'}$, $H_{3'}$, $\underline{CH_3}$–C_4 of **1Aa**; H_5 and H_{12} of **1Ac**; and H_5, $H_{5'}$, H_6 of **1Ad**. For **1Ab** and **1Ba** it was necessary to increase the ratio [**CSA**]/[**1**] up to five or six folds to obtain satisfactory enantiomeric discrimination either by ^1H or ^{19}F NMR. But even with a ratio as high as 8/1 no discrimination was observed for the ^1H and ^{19}F NMR signals of the thiazoline **1Bc**.
2. Therefore enantiomeric discrimination is more difficult for α-S substituted thiazolines **1Ba** and **1Bc** than for the corresponding α-N regiomer **1Aa** and **1Ac**. For the corresponding regiomers of oxazolines the same trend was also observed with CSAs [18] or with CLSRs [16]. The difference in the behaviour of the regiomers was interpreted, in the last case, considering the greater distance of the preponderant interacting site N_3 from the stereocenter for α-O substituted oxazolines. But this argument seems inadequate with the thiazoline series since:
   - $\underline{CH_3}$–C_4^* and $\underline{CH_3}$–C_5^* signals are not at all the best NMR markers for chiral discrimination despite their proximity to the stereocenter;
   - the methylene signal H_{12} of **1Ac** is almost enantiomerically resolved to baseline despite its appearent greater distance to the stereocenter (see Table 2).

3.2.2. Enantiomeric composition

3.2.2.1. Inequivalence sense The inequivalence sense [19] was determined using enantiomerically enriched mixtures for each thiazoline and CSA **5b** (see Table 3). It appeared that generally, in ^1H NMR spectra, the most shielded signals (for assigned signals) correspond to (S) enantiomer for thiazolines **1Aa–d** and **1Ba**. Noticeable is the fact that the same sense prevails also for the corresponding oxazolines [18]. The opposite sense occurs only for H(12) of **1Ac** and for the ^{19}F signals of **1Ab**.

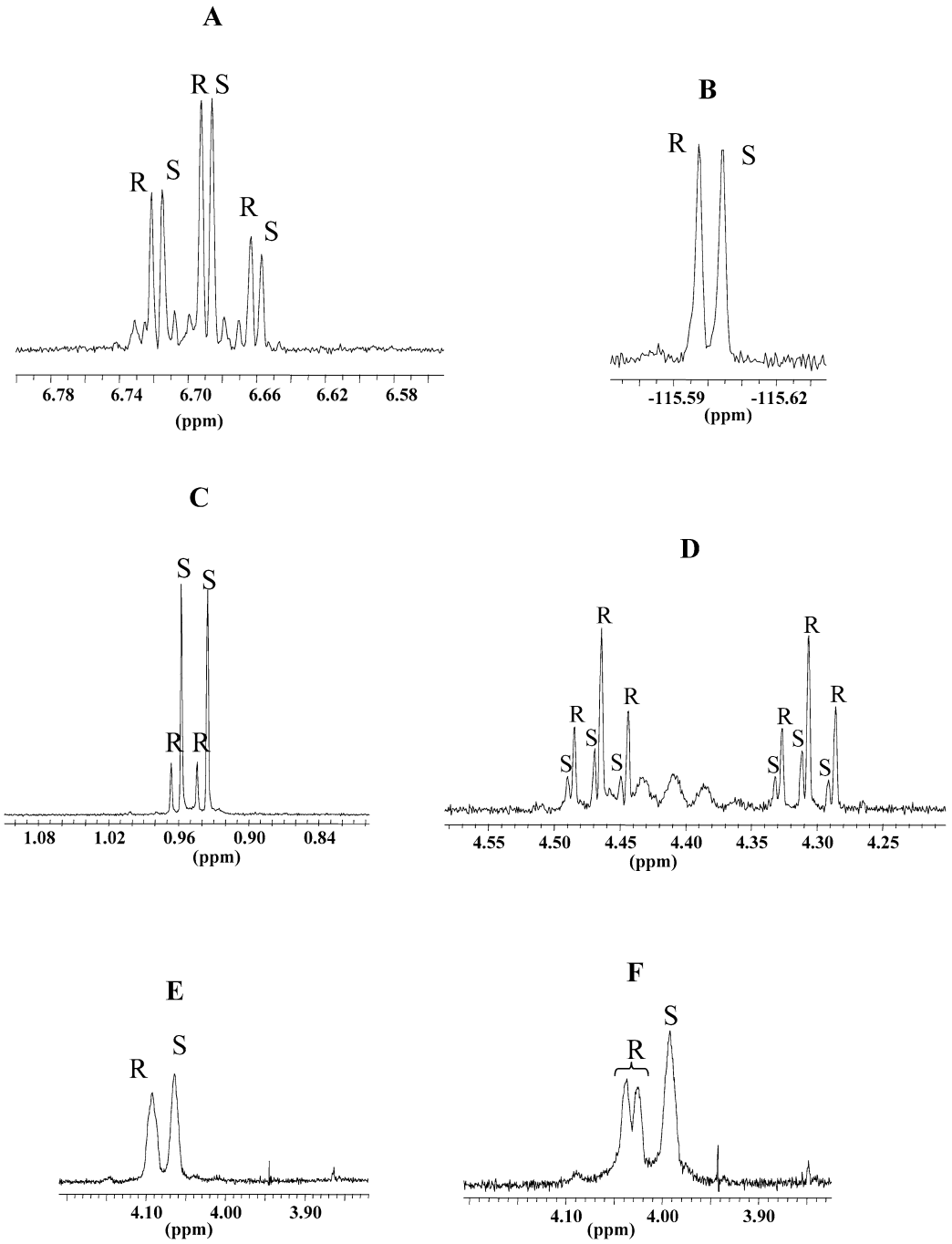


Fig. 3. Enantiomeric discrimination of the ^{1}H and ^{19}F NMR signals of thiazolines **1Aa**, **1Ab**, **1Ac**, **1Ad** and **1Ba** in the presence of the CSA **(R)-5b**. Assignments were made using pure enantiomers. A: H(3′) ^{1}H NMR signal of **1Aa**, [**5**]/[**1**] = 2/1, in C_6D_6 solutions at 296 K. B: $^{19}F[^{1}H]$ NMR signal of **1Ba**, [**5**]/[**1**] = 6/1, in C_6D_6 solutions at 296 K. C: CH_3–C(4) ^{1}H NMR signal of **1Ab**, [**5**]/[**1**] = 2/1 with [(**R**)-**1Ab**]/[(**S**)-**1Ab**] = 1/5, in CCl_4CDCl_3 solutions at 276 K. D: H(12) ^{1}H NMR signal of **1Ac**, [**5**]/[**1**] = 2/1 with [(**R**)-**1Ac**]/[(**S**)-**1Ac**] = 4/1, in $CCl_4/CDCl_3$ solutions at 296 K. E: H(6) ^{1}H NMR signal of **1Ad**, [**5**]/[**1**] = 2/1, in $CC_4/CDCL_3$ solutions at 296 K. F: H(6) ^{1}H NMR signal of **1Ad**, [**5**]/[**1**] = 5/1, in $CCl_4/CDCl_3$ solutions at 296 K.

Table 4
Temperature effect on enantiomeric discrimination of ^{1}H NMR signals of Δ^2-thiazolines-1,3 **1** by solvatation with chiral solvating CSA **5b**

	Temperature (K)	Achiral solvent	(**R**)-**1**/ (**S**)-**1**	[(**R**)-**5b**] /[**1**]	Proton of **1**	$\Delta\delta$[a] (ppm)	$\Delta\nu$ (Hz)	$\delta^{1/2b}$ (Hz)	h %	Assay
1Ab	**296**	C_6D_6	Racemate	2/1	H_5	−0.07	1.80	0.05	23	**1**
			1/1		CH_3–C_4	−0.01	1.00	0.99	58	
				5/1	**H_5**	**−0.15**	**4.30**	**0.90**	**0**	**2**
					CH_3–C_4	−0.12	2.70	0.72	6	
	296		**1/4**	**5/1**	H_5	n.u.[c]	–	0.80	–	
					CH_3–C_4	n.u.	–	0.42	–	
	286				**H_5**	**−0.12**	**3.20**	**0.33**	**0**	**3**
					CH_3-C_4	**−0.09**	**0.60**	**0.55**	**0**	
	276				**H_5**	**−0.11**	**4.40**	**0.72**	**0**	
1Ac	**296**	CCl_4/ $CDCl_3$	**1/1**	**2/1**	**H_5**	**−0.06**	**3.10**	**0.73**	**0**	**4**
					H_{12}	**−0.02**	**1.70**	**0.45**	**6**	
	296		**1/4**	**2/1**	**H_5**	**−0.06**	**3.00**	**0.82**	**0**	**5**
					H_{12}	**−0.02**	**1.60**	**0.86**	**5**	
	286/276				H_5	n.u.	–	14.4	–	**6**
					H_{12}	n.u.	–	9.37	–	

In bold characters are indicated the usable signals for enantiomeric NMR discrimination.
[a] See Table 1 for initial chemical shifts without CSA.
[b] Width at half peak height.
[c] n.u.: discrimination not usable due to overlapping with other signals of thiazoline or with the **5b** hydroxyl group.

3.2.2.2. Enantiomeric excess determination Enantiomerically enriched samples of thiazolines **1** served also to appreciate the enantiomeric composition of thiazolines **1** and the precision on e.e. determination using chiral NMR analysis. Since, with CSAs, line widthes are equivalent for enantiomers, the enantiomeric composition can be determined from the relative intensities of the anisochronous resonances themselves obtained either from area or peak height. It was previously estimated that "as little as 0.02 ppm nonequivalence is adequate for a $\pm 2\%$ determination of ee on well-resolved singlets at 100 MHz" [19]. From this work, we consider to be in a position to furnish a $\pm 1\%$ determination of e.e. on well-resolved multiplets i.e. with $\Delta\nu$ as little as 4–3 Hz at 300 MHz ($\Delta\Delta\delta \sim 0.01$ ppm), and a $\pm 3\%$ determination of e.e. with even smaller $\Delta\nu$ of about 1.8 Hz. Thus we confirmed by the observation of e.e. ranging from 96 to 99 $\pm 1\%$ for the enantiomers of thiazolines **1Ac** and **1Aa**, and the regiomer **1Ba** that the reaction induced by Lawesson reagent occur without racemisation and probably with retention of configuration as previously observed for oxazolines [18]. More precisely there is retention and inversion of configuration for the C(4) stereocenter of **1Aa–c**, and for the C(5) stereocenter of **1Ba**, respectively.

3.2.2.3. Improvement of the enantiomeric discrimination As expected, overloading assays resulted in a diminishing of resolution ($h\%$ increases). Therefore, with the thiazolines presenting a more difficult analysis, further assays were conducted to improve their enantiomeric discrimination by varying the ratio [**5**]/[**1**], the temperature, the achiral solvent and the CSA **5**.

At room temperature, with the racemate of thiazoline **1Ab** it was sufficient to increase the [**5b**]/[**1**] ratio from 2/1 to 5/1 to obtain a baseline resolution for H_5 signal (see assays 1 and 2 in Table 4). For an enriched solution of **1Ab** a decrease in temperature of 10K was sufficient for providing a baseline resolution of this signal for the same [**5b**]/[**1**] ratio = 5 (see assay 3 in Table 4). This behaviour agrees

with the known effect of reduction in temperature that provides a $\Delta\nu$ enhancement resulting generally in an improvement of the resolution [19]. However this favourable effect can be outmatched by a simultaneous broadening of the signal arising from the increase in the viscosity of the solution, as presently observed for H_5 and H_{12} signals of CCl_4 solutions of thiazoline **1Ac** (see assays 5 and 6 in Table 4). In fact, with thiazoline **1Bc** no enantiomeric discrimination could be obtained by varying the previous factors or changing the CSA **5b** for **5a**.

4. Conclusion

The good complementarity existing between the functionalities of thiazolines **1** and the Pirkle's trifluorocarbinol **5b** resulted in a good enantiomeric discrimination for five of the six thiazolines studied namely **1Aa–d** and **1Ba**. This simple and rapid chiral method has firstly shown that the synthesis of thiazolines using Lawesson reagent and hydroxylamides **6A** and **6B** enantiomerically pure occurs without racemization. In a proinsecticide perspective it will be soon applied to extracts of locust tissues incubated with racemates of thiazolines to determine the possible occurrence of a chiral recognition during their enzymatic hydrolysis.

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