Journal of Molecular Structure 1261 (2022) 132892

Contents lists available at ScienceDirect

Journal of Molecular Structure

journal homepage: www.elsevier.com/locate/molstr

Rotamerization equilibrium in novel *N,N*-disubstituted chloroacetamides: An NMR spectroscopic study

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ARTICLE INFO

Article history:
Received 20 November 2021
Revised 18 March 2022
Accepted 20 March 2022
Available online 24 March 2022

Keywords:
Chloroacetamides
Rotamers
NMR
ASIS effect
Molecular modeling

ABSTRACT

We studied the *E/Z* rotamers of new *N*-substituted 2–chloro-*N*-(2-nitrobenzyl)acetamides by applying ^{1}H-NMR and ^{13}C-NMR techniques. The restricted rotation around the amide bond generates a mixture of *E/Z* diastereomers visualized as a set of appreciably separated signals in the NMR spectra. In order to assign each signal to its corresponding rotamer, the criterion of the anisotropic solvent-induced shift (ASIS) effect, observed in the ^{1}H-NMR spectra, was used. The effects of solvent and substituent on the relative *E/Z* ratios were also analyzed.

1. Introduction

The synthesis of new small bioactive molecules is a continuous challenge for the synthetic organic chemistry community [1]. *N*-substituted 2-chloroacetamides are molecules with a high impact on biochemistry. Several examples of molecules acting as targeted covalent inhibitors (TCIs) with proven pharmacological effects have been reported in recent years [2]. Thus, the synthesis of novel chloroacetamides is still a field to be investigated in order to enlarge the molecular diversity of this kind of molecule. As biological receptors interact with bioactive molecules in a precise, unique way, the molecular structure and conformations of potentially bioactive synthetic organic compounds must be analyzed in all their complexity [3]. Structural features related to isomeric equilibriums play a key role in many biological processes. For example, *E/Z* isomerization in substituted amides was reported as crucial in protein folding and biocatalytic processes [4]. The (O)C-N moiety adopts a partial double bond electronic configuration with a hindered rotation [5], and this particular electronic feature causes a rotational barrier established in 15–25 kcal/mol [6] that can be studied by spectroscopic techniques [7]. When a substituent is present in the nitrogen atom, the asymmetrical substructure could be represented as a non-isolable pair of *E/Z* rotamers (Fig. 1).

This equilibrium could be seen in NMR spectra as different sets of signals when rotamers are appreciably present. Therefore, NMR spectrometric analysis, including dynamic techniques, complemented with theoretical calculations brings valuable information about structures and their conformations [8]. Recently, we have observed these spectral signals in some α-chloroacetamides synthesized in our laboratory. As part of our ongoing efforts in the synthesis and in-depth structure elucidation of new bioactive compounds [9], we report herein a complete NMR and molecular modeling study of nine new *N*-substituted 2-chloro-*N*-(2-nitrobenzyl)acetamides.

2. Experimental

2.1. Chemistry

The preparation of novel chloroacetamides was performed as previously described [9g] *N*-substituted *N*-(2-nitrobenzyl)amines were prepared by reaction with *o*-nitrobenzaldehyde in ethanol at room temperature. After the aldehyde was consumed, $NaBH_4$ was added and 2-nitrobenzylamines were obtained after 2 h when R= alkyl or aralkyl and 24 h when R= aryl. The amine (3 mmol) and triethylamine (3 mmol) were dissolved in 5 mL of toluene

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https://doi.org/10.1016/j.molstruc.2022.132892
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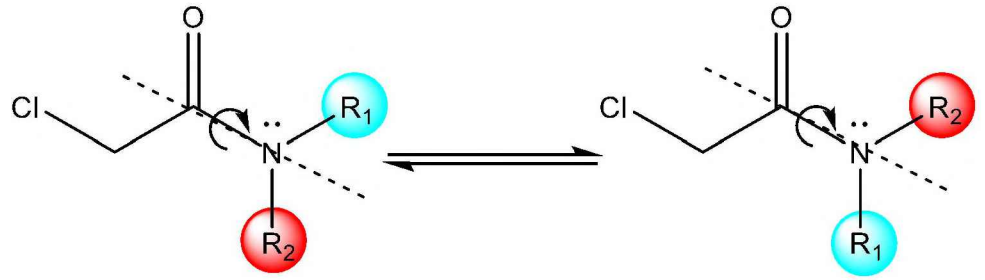


Fig. 1. Rotamerization equilibrium in N,N-disubstituted chloroacetamides.

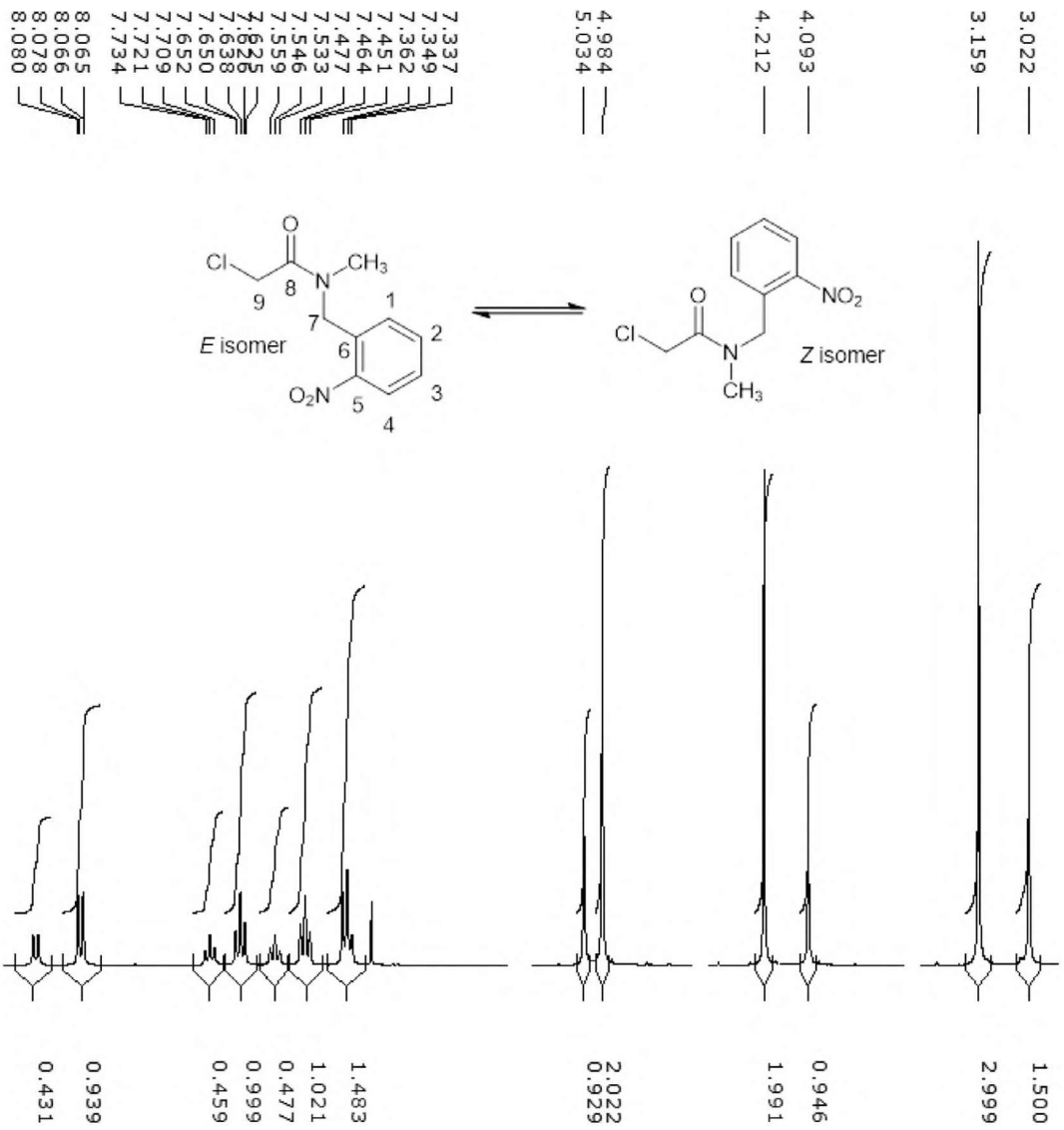


Fig. 2. ^{1}H NMR spectrum of compound **1a** in $CDCl_3$.

and a solution of 3 mmol of chloroacetyl chloride in 5 mL of toluene was added. The mixture was heated to reflux for 2–24 h, cooled and washed with water (10 mL), HCl 10% (10 mL) and water (2 × 10 mL). Solution was dried over sodium sulfate and concentrated to dryness at reduced pressure to obtain crude 2–chloro-*N*-(2-nitrobenzyl)acetamides (**1**) that were purified by column chromatography (hexane-EtOAc 8:2) or crystallization from methanol. The desired products were obtained in good yields (Scheme 1).

2.2. NMR spectra

^{1}H- and ^{13}C-NMR spectra were measured at 600.13 MHz and 150.91 MHz, respectively, on a Bruker UltraShield 14.1 T with shim Boss II (Probe multinuclear Bruker Smart Probe BBFO (5 mm) and Bruker Avance III acquisition and control system unless noted otherwise. *J* values are given in Hz. All assignments were confirmed by a combination of 2D spectra (HSQC and NOESY) using $CDCl_3$ or C_6D_6 solution. Chemical shifts are indicated in parts per mil-

Table 1
^{1}H chemical shifts and coupling constants (Hz) of compounds **1** in $CDCl_3$. (*: *E/Z* overlapped signals).

Compound	H_1	H_2	H_3	H_4	H_7	H_9	$H_{Aromatics}$	R
1a (*Z*)	7.35–7.30 (m)*	7.61 (td, *J* = 7.6, 1.3)	7.46–7.41 (m)	8.05 (dd, *J* = 8.2, 1.3)	4.96 (s)	4.19 (s)		**H_{10}:** 3.14 (s)
1a (*E*)	7.35–7.30 (m)*	7.70 (td, *J* = 7.6, 1.4)	7.55–7.50 (m)	8.17 (dd, *J* = 8.2, 1.4)	5.01 (s)	4.07 (s)		**H_{10}:** 3.00 (s)
1b (*Z*)	7.36–7.33 (m)*	7.60 (td, *J* = 7.6, 1.4)	7.45–7.40 (m)	8.05 (dd, *J* = 8.2, 1.4)	4.94 (s)	4.18 (s)		**H_{10}:** 3.46 (q, *J* = 7.1) **H_{11}:** 1.26 (t, *J* = 7.1)
1b (*E*)	7.36–7.33 (m)*	7.69 (td, *J* = 7.6, 1.4)	7.54–7.50 (m)	8.17 (dd, *J* = 8.2, 1.4)	4.99 (s)	4.01 (s)		**H_{10}:** 3.42 (q, *J* = 7.1) **H_{11}:** 1.17(t, *J* = 7.1)
1c (*Z*)	7.37–7.32 (m)*	7.60 (td, *J* = 7.6, 1.3)	7.45–7.41 (m)	8.05 (td, *J* = 8.1, 1.3)	4.94 (s)	4.18 (s)		**H_{10}:** 3.33 (m)* **H_{11}:** 1.67 (m) **H_{12}:** 0.94 (t, *J* = 7.4)
1c (*E*)	7.37–7.32 (m)*	7.69 (td, *J* = 7.6, 1.3)	7.54–7.50 (m)	8.18 (td, *J* = 8.1, 1.3)	5.00 (s)	4.00 (s)		**H_{10}:** 3.33 (m)* **H_{11}:** 1.62 (m) **H_{12}:** 0.90 (t, *J* = 7.4)
1d (*Z*)	7.32 (dd, *J* = 7.8, 1.1)	7.57 (td, *J* = 7.8, 1.2)	7.41–7.37 (m)	8.06 (td, *J* = 8.2, 1.2)	4.85 (s)	4.21 (s)		**H_{10}:** 4.32 (hept, *J* = 6.6) **H_{11}, H_{12}:** 1.22 (d, *J* = 6.6)
1d (*E*)	7.46 (dd, *J* = 7.7, 1.1)	7.68 (td, *J* = 7.7, 1.2)	7.53–7.49 (m)	8.17 (td, *J* = 8.2, 1.2)	4.91 (s)	3.87 (s)		**H_{10}:** 4.79 (hept, *J* = 6.6) **H_{11}, H_{12}:** 1.12 (d, *J* = 6.6)
1e (*Z*)	7.36–7.32 (m)*	7.60 (td, *J* = 7.6, 1.4)	7.46–7.40 (m)	8.05 (dd, *J* = 8.2, 1.4)	4.95 (s)	4.18 (s)		**H_{10}:** 3.36 (t)* **H_{11}:** 1.66 – 1.53 (m)* **H_{12}:** 1.37 – 1.28 (m)* **H_{13}:** 0.94 (t, *J* = 7.4)
1e (*E*)	7.36–7.32(m)*	7.68 (td, *J* = 7.7, 1.4)	7.54–7.50 (m)	8.18 (d, *J* = 8.2, 1.4)	4.99 (s)	4.00 (s)		**H_{10}:** 3.36 (t)* **H_{11}:** 1.66 – 1.53 (m)* **H_{12}:** 1.37 – 1.28 (m)* **H_{13}:** 0.91 (t, *J* = 7.4)
1f	7.67- 7.62 (m)	7.43 (ddd, *J* = 8.5, 7.2, 1.3).	7.62 - 7.67 (m)	7.93 (dd, *J* = 8.1, 1.3)	5.33 (s)	3.92 (s)	**H_{12}, H_{14}:** 7.15 - 7.14 (m) **H_{11}, H_{13}, H_{15}:** 7.40 – 7.36 (m)	
1 g	7.54–7.49 (m)	7.60 - 7.65 (m)	7.60 - 7.65 (m)	7.95 (dd, *J* = 8.2, 1.4)	5.29 (s)	3.90 (s)	**H_{12}, H_{14}:** 7.54–7.49 (m) **H_{11}, H_{15}:** 7.04 (d, *J* = 8.6)	
1 h (*Z*)	7.38–7.16 (m)*	7.60 (td, *J* = 7.6, 1.3)	7.46–7.41 (m)	8.06 (dd, *J* = 8.2, 1.3)	5.00 (s)	4.02 (s)	**H_{13-17}:** 7.38 - 7.16 (m)*	**H_{10}:** 3.62 (t, *J* = 7.1) **H_{11}:** 2.94 – 2.91 (m)
1 h (*E*)	7.38–7.16 (m)*	7.65 (td, *J* = 7.7, 1.3)	7.54–7.48 (m)	8.15 (dd, *J* = 8.2, 1.3)	4.82 (s)	3.77 (s)	**H_{13-17}:** 7.38 - 7.16 (m)*	**H_{10}:** 3.59 - 3.56 (m) **H_{11}:** 2.85 (t, *J* = 7.1)

Table 2
^{13}C chemical shifts assigned with the aid of HSQC for selected compounds **1** in $CDCl_3$.

	Compound					
	1c		1d		1f	
Position	^1H δ (ppm)	^{13}C δ (ppm)	^1H δ (ppm)	^{13}C δ (ppm)	^1H δ (ppm)	^{13}C δ (ppm)
1	(*Z*) 7.34	128.93	(*Z*) 7.32	133.95	7.43	128.45
	(*E*) 7.34	128.27	(*E*) 7.46	134.07		
2	(*Z*) 7.60	133.94	(*Z*) 7.57	133.76	7.62 - 7.67	133.57
	(*E*) 7.69	134.48	(*E*) 7.68	134.28		
3	(*Z*) 7.43	127.64	(*Z*) 7.39	127.94	7.62 - 7.67	130.05
	(*E*) 7.52	128.22	(*E*) 7.51	128.81		
4	(*Z*) 8.05	125.30	(*Z*) 8.06	125.24	7.93	124.94
	(*E*) 8.18	126.03	(*E*) 8.17	125.99		
7	(*Z*) 4.94	46.34	(*Z*) 4.85	41.72	5.33	50.63
	(*E*) 5.00	50.63	(*E*) 4.91	44.44		
9	(*Z*) 4.18	40.97	(*Z*) 4.21	41.41	3.92	41.64
	(*E*) 4.00	41.38	(*E*) 3.87	42.00		
10	(*Z*) 3.33	49.39	(*Z*) 4.32	50.07		131.91
	(*E*) 3.33	48.91	(*E*) 4.79	47.34		
11	(*Z*) 1.67	20.56	(*Z*) 1.22	21.27	7.36 - 7.40	130.19
	(*E*) 1.62	22.34	(*E*) 1.12	19.88		
12	(*Z*) 0.94	11.27	(*Z*) 1.22	21.27	7.14 - 7.15	130.19
	(*E*) 0.90	11.30	(*E*) 1.12	19.88		
13					7.36 - 7.40	127.66
14					7.36 - 7.40	127.66
15					7.36 - 7.40	129.10
8		167.34 167.37		167.33 167.46		167.00
5 & 6		132.68 132.78 147.90 148.49		134.07 134.06 147.62 147.47		140.93 148.65

lion relative to TMS. The multiplicities are expressed as: singlet (s), doublet (d), triplet (t), quartet (q), hexuplet (h), multiplet (m), double doublet (dd) and triple doublet (td).

2.3. Computational details

Compounds **1c** and **1f** have been taken as a model for theoretical calculations using Gaussian Software [10a]. After a preoptimization process, the C–N-C(O) torsion angle was scanned using AM1 calculations. In both cases, two minima were found, which correspond to *E* and *Z* conformers. These structures were optimized using DFT method [10b], B3LYP functional and 6-311++G(d,p) as basis set [10c-d].

Chemical shifts (δ_{calc}) relative to tetramethylsilane were then calculated for ^1H and ^{13}C nuclei for the two optimized conformers for both compounds. The isotropic magnetic shielding tensors were evaluated in chloroform and benzene by means of GIAO NMR calculations at the B3LYP/6-31+G(d,p) level of theory using the polarization continuum model, SCRF-PCM [10f]. Each scalar isotropic shielding value (σ_i) was turned into a chemical shift by subtracting it from the corresponding σ of ^1H or ^{13}C of TMS (these σ were calculated using the same method and basis set).

3. Results and discussion

The NMR spectra of all new compounds were carefully analyzed. Due to the slow rotation around the N–C(O) bond, these compounds exist in solution as a mixture of two *E* and *Z* isomers (rotamers) that were detected by ^1H and ^{13}C-NMR. As we expected, duplicated signals could be attributed to the two *E*/*Z* isomers, as shown in Fig. 2.

^1H and ^{13}C chemical shifts and multiplicities for both isomers in $CDCl_3$ are given in Tables 1 and 2, respectively. At this point, to establish the correspondence of the signals with *E* or *Z* rotamers, we assume the reported tendency for model compounds, which indicates that, concerning to the carbonyl moiety, *cis* methylene protons are more deshielded than those in the *trans* isomer [11]. The relative proportions of rotamers in $CDCl_3$ were calculated by integrating the C7 and C9 methylene signals, as listed in Table 3. In all cases, the signal assignments were made with the aid of gHSQC, and NOESY experiments.

Table 3
E/*Z* ratios at 25 °C for compounds **1** in $CDCl_3$ and C_6D_6.

	$CDCl_3$		C_6D_6	
Compound	%*E*	%*Z*	%*E*	%*Z*
1a	31	69	32	68
1b	36	64	37	63
1c	35	65	36	64
1d[a]	70	30	67	33
1e	34	66	35	65
1h	32	68	40	60

[a] Note that the rotamers visualized for compound 1d have inverted names because of priority rules.

Although some reports demonstrate good correlations between chemical shifts and *cis*/*trans* relative location of *N*-substituents in amides, this information could not be assumed to be undoubtedly certain [12]. Recording the NMR spectra in C_6D_6 solution could provide more structural information. The results obtained are summarized in **Tables 1SI** and **2SI**.

As shown in Tables 1 and **1SI**, the signal of R substituent *trans* to the carbonyl moiety (*Z*) suffers a diamagnetic shift ($\Delta\delta$) when chloroform was replaced with benzene as the deuterated solvent (Fig. 3). This $\Delta\delta$ was smaller in the rotamer where R substituent is *cis* to the carbonyl moiety (*E*). This phenomenon was extended (Table 4), and it agrees with the anisotropic solvent-induced shifts (ASIS) effect [13].

Assuming that the ASIS effect is a piece of reliable information, we could describe and undoubtedly assign all signals expressed in the NMR spectra to both diastereomers in equilibrium. In the ^1H NMR spectrum of **1a**, the protons of the methyl group appear as two singlet signals at 3.00 (*E*) and 3.14 (*Z*) ppm. These characteristic two sets of signals were also observed in both C7 and C9 methylenes. The benzyl protons appear at 4.96 (*Z*) and 5.01 (*E*) ppm and the chloromethylene at 4.07 (*E*) and 4.19 (*Z*) ppm. Also, aromatic protons were distinguished in both rotamers. Near the nitro group, the H4 signals at 8.17 (*Z*) and 8.05 (*E*) appear as

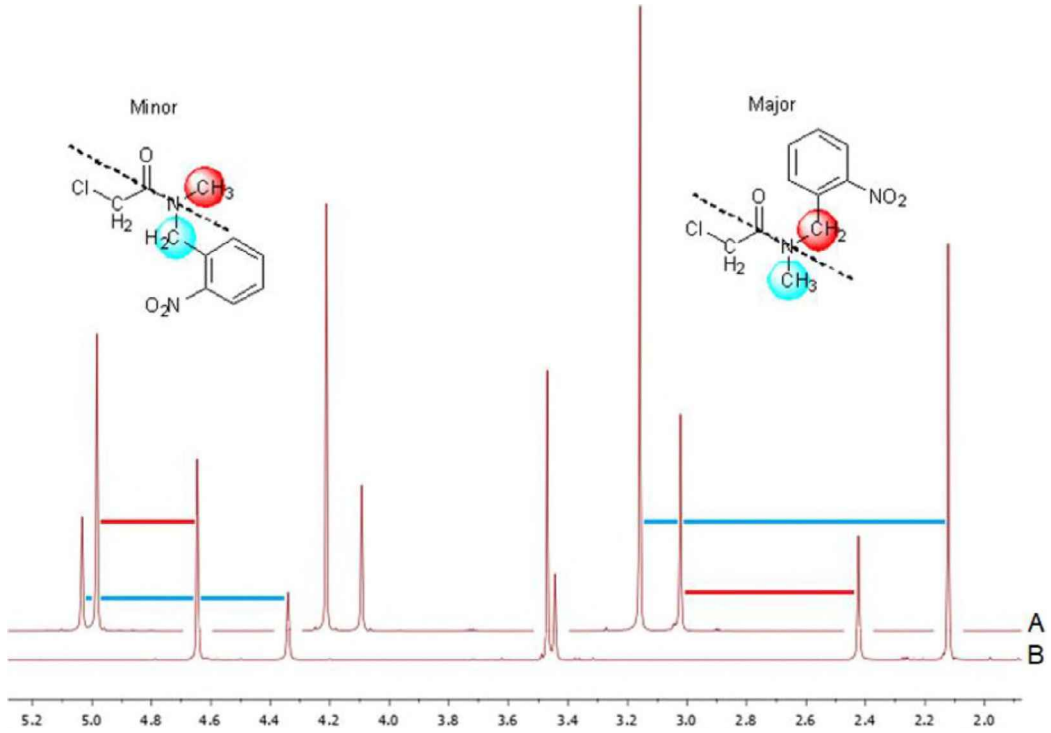


Fig. 3. Detail of the ^{1}H NMR spectra of **1a** ($R=CH_3$) in $CDCl_3$ (A) and C_6D_6 (B). The blue lines indicates the signal experiencing the strongest ASIS for each rotamer.

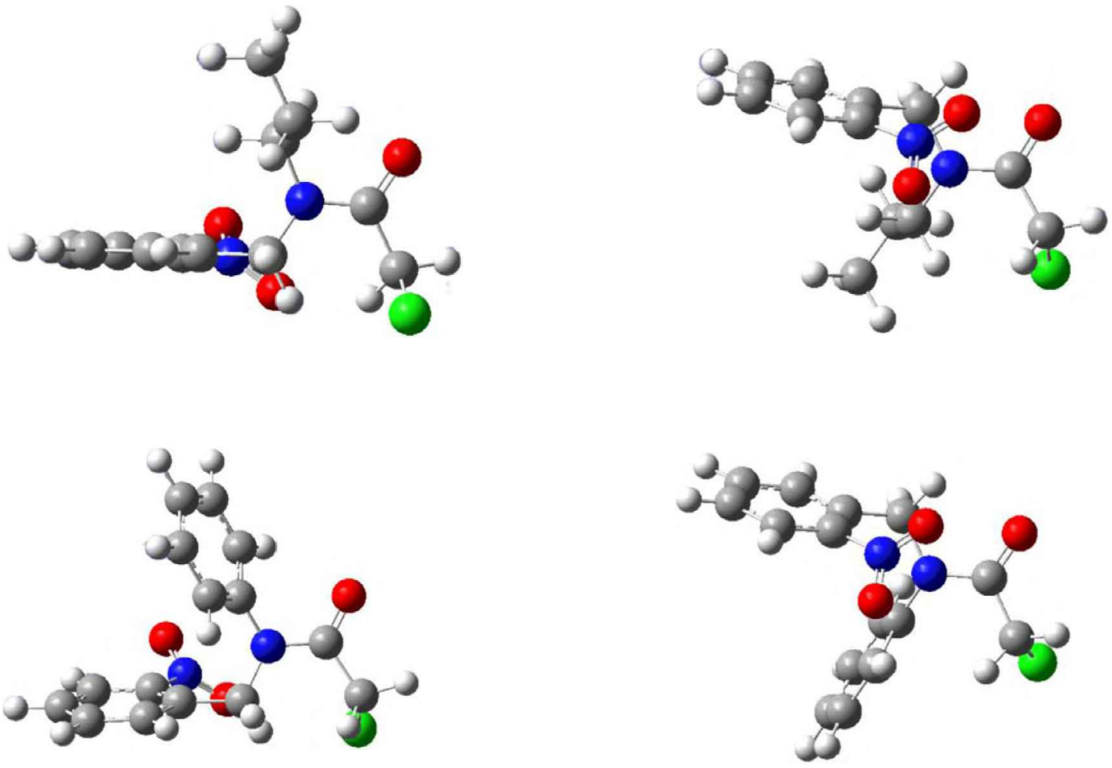


Fig. 4. Conformational minima for compounds **1c** and **1f** at the B3LYP/6-311++G(d,p) level.

Table 4
$\Delta\delta$ values, calculated from $CDCl_3$ and C_6D_6 ^1H NMR spectra, indicating the ASIS effect for compounds **1**.

Compound	δ (ppm) $H_7C_6D_6$	δ (ppm) H_7CDCl_3	$\Delta\delta$ (ppm) H_7	δ (ppm) $H_9C_6D_6$	δ (ppm) H_9CDCl_3	$\Delta\delta$ (ppm) H_9
1a (*Z*)	4.65	4.96	0.31	3.48	4.19	0.71
1a (*E*)	4.34	5.01	0.67	3.45	4.07	0.62
1b (*Z*)	4.73	4.94	0.21	3.53	4.18	0.65
1b (*E*)	4.45	4.99	0.54	3.40	4.01	0.61
1c (*Z*)	4.78	4.94	0.16	3.60	4.18	0.58
1c (*E*)	4.54	5.00	0.46	3.41	4.00	0.59
1d (*Z*)	4.50	4.91	0.41	3.35	3.87	0.52
1d (*E*)	4.79	4.85	0.06	3.59	4.21	0.62
1e (*Z*)	4.72	4.95	0.23	3.48	4.18	0.70
1e (*E*)	4.41	4.99	0.58	3.35	4.00	0.65
1 h (*Z*)	4.84	5.00	0.16	3.40	4.02	0.62
1 h (*E*)	4.42	4.82	0.40	3.27	3.77	0.50

Table 5
Calculated and experimental chemical shifts (ppm) for *E/Z* rotamers of compound **1c** in $CDCl_3$.

Carbons	*E*		*Z*		Hydrogens	*E*		*Z*	
	Calc.	Exp.	Calc.	Exp.		Calc.	Exp.	Calc.	Exp.
1	128.6	128.27	127.2	128.93	1	7.61	7.34	7.68	7.34
2	129.5	134.48	129.3	133.94	2	7.97	7.69	7.86	7.60
3	125.6	128.22	124.1	127.64	3	7.72	7.52	7.83	7.43
4	122.2	126.03	122.1	125.30	4	8.18	8.18	8.03	8.05
5	147.9	148.49	147.5	147.90	7	4.88	5.00	4.93	4.94
6	129.8	132.68	128.7	132.68	9	4.22	4.00	3.81	4.18
7	51.4	49.39	46.30	46.34	10	2.65	3.33	2.61	3.33
8	161.7	167.37	161.2	167.34	11	1.40	1.62	1.51	1.67
9	47.3	41.38	46.30	40.97	12	0.62	0.90	0.56	0.94
10	46.2	48.91	49.00	50.63					
11	19.7	22.34	23.10	20.56					
12	11.3	11.30	11.00	11.27					

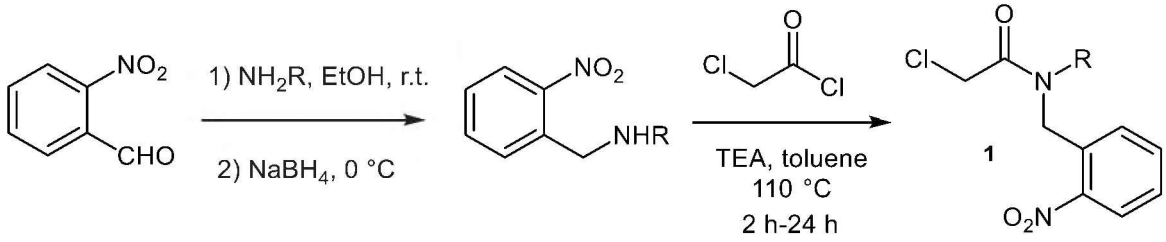
Table 6
Calculated and experimental chemical shifts (ppm) for *E/Z* rotamers of compound **1f** in $CDCl_3$.

Carbons	*E*	*Z*		Hydrogens	*E*	*Z*	
	Calc.	Calc.	Exp.		Calc.	Calc.	Exp.
1	127.6	131.2	128.45	1	7.11	7.59	7.43
2	129.1	129.5	133.57	2	7.65	7.77	7.64
3	124.0	124.6	130.05	3	7.65	7.50	7.64
4	121.9	122.2	124.94	4	8.03	8.22	7.93
5	147.9	146.3	148.65	7	5.31	5.50	5.33
6	129.4	129.6	140.93	9	3.50	3.97	3.92
7	51.80	54.70	50.63	Ph	6.72 - 7.69	6.93 - 7.38	7.15–7.40
8	161.7	161.10	167.00				
9	45.10	46.60	41.64				
Ph	124.3 - 135.7	120.8 - 136.5	127.66–131.91				

two doublets (J^3_{HH} =8.2 Hz), then two pairs of triplets at 7.44 (*Z*) and 7.52 (*E*) (J^3_{HH}=7.4) for protons H3 and 7.62 (*Z*) and 7.70 (*Z*) with J^3_{HH}=7.3 for protons H2 were present. The H1 peak was the only signal in the spectra that could not differentiate between the isomers, and a multiplet with the correct integration appears at 7.35–7.31 ppm. This assignment was also confirmed with data obtained from NOESY spectra. In all the examples examined, with *N*-alkyl substituents, appreciable signal correlations with the CH_2Cl methylene were detected only in the major isomer, assuming it as *Z*. The correlations extracted from gHSQC analysis provide the information to correctly assign all ^{13}C signals. After integrating and comparing all signals for compound **1a**, we assume that 69% of molecules in equilibrium are the *Z* isomer. The *Z* rotamers predominate in all cases, as shown in Table 3, except for R=isopropyl due to a change in substituent priority. This expected result is consistent with the steric influence of the groups that surrounded the amide bond. The bulky chloromethyl moiety preferred the *trans* position towards the *o*-nitrobenzyl group as this is more voluminous than the R-alkyl substituent. The populations of this major conformation are all around 65–70%. When aryl *N*-substituent (**1f** and **1g**) is present, the electronic effects produce a delocalization of the nitrogen electrons so *E/Z* diastereomers were not detected in the NMR time scale [14].

The experimental information collected reveals a slow molecular equilibrium at room temperature, visible by NMR, and it may be attributed to the presence of relatively stable *E* and *Z* rotamers with respect to rotation about the (O)C–N bond. Although all NMR spectra were completely assigned, and the chemical shifts and ASIS effect are consistent with the proposed structures, we can obtain more information from theoretical calculations and analyze and compare it with the experimental observations [15]. Theoretical quantum calculations reveal crucial data to support experimental values, and hence elucidating the molecular structures is easier [16]. In this work, molecular modeling was performed to obtain the relative stability of the two rotamers and calculate the chemical shifts of carbon and hydrogen atoms. Model compounds 1c (R=propyl) and 1f (R=phenyl) were studied computationally using Gaussian Software as previously described (Fig. 4).

These structures were optimized using the DFT method, B3LYP functional, and 6-311++G(d,p) as basis set. ΔG at 25 °C was calculated in both cases by taking the differences between the *Z* and E rotamer energies, the results obtained being 1.55 kcal.mol^{-1} and



Scheme 1. Synthetic route for preparation of *N*-substituted 2-chloro-*N*-(2-nitrobenzyl)acetamides.

-0.29 kcal.mol^{-1} for compounds **1c** and **1f**, respectively. These calculations were in full accordance with the experimental results obtained by NMR spectroscopy, and the ΔG values obtained show the same tendency as experimental ones in solution. The ^{1}H and ^{13}C NMR chemical shifts were calculated using chloroform and benzene as solvents, and a good correlation with experimental δ was found, as shown in Tables 5 and 6.

The experimental and theoretical data collected are consistent values in all cases. For the structures analyzed herein, the prediction fits well with the expected abundance of rotamers and their chemical shifts. We calculated their relative abundance in $CDCl_3$ and C_6D_6 solution, as well as the ASIS effect, and found that the chemical shifts provide the necessary information to distinguish both rotamers. Also, the theoretical calculations supported the spectroscopic experimental information: the most stable rotamer was the most abundant in solution, and all calculated chemical shifts clearly differentiated the *E* and *Z* diastereomers with the same experimental pattern.

4. Conclusions

We report here the complete ^{1}H and ^{13}C-NMR characterization of a series of new *N*-substituted 2-chloro-*N*-(2-nitrobenzyl)acetamides. Such compounds display *E/Z* stereoisomerism due to partial (O)C–N double bond character, showing two unequally populated sets of signals in their ^{1}H and ^{13}C-NMR spectra. The ^{1}H-NMR resonances of both rotamers were assigned on the basis of the ASIS effects. For all the compounds, the *E/Z* equilibrium favors the *Z* diastereomer. This preference of chloromethylene and 2-nitrobenzyl moieties in *cis* position is sensitive to steric hindrance. Chloroacetamides carrying an aryl substituent to nitrogen atom did not show *E/Z* stereoisomerism. Results of the complete conformational study for compunds **1c** and **1f** performed at DFT-B3LYP/6-36–311++G(d,p) level of theory were in good agreement with the experimentally determined *E/Z* ratios.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

CRediT authorship contribution statement

Guido G. Fraga: Investigation, Formal analysis. **Diego D. Colasurdo:** Software, Formal analysis. **Cintia C. Santiago:** Data curation, Writing – original draft, Writing – review & editing. **Agustín Ponzinibbio:** Resources, Validation, Writing – original draft, Writing – review & editing. **Leandro D. Sasiambarrena:** Conceptualization, Methodology, Visualization, Supervision.

Acknowledgment

Authors wish to thank CICPBA, UNLP and CONICET of Argentina for financial support. We express deep thanks to Lic. Omar E. Guaymás (CICPBA) for assistance in the preparation and NMR characterization of some compounds and Prof. Dr. Sergio L. Laurella (UNLP) for valuable advice. A. P. is a research fellow of CONICET.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.molstruc.2022.132892.

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