Journal of Molecular Structure 1059 (2014) 176–184

Contents lists available at ScienceDirect

Journal of Molecular Structure

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

NMR and computational studies on tautomerism of 3-hydroxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one

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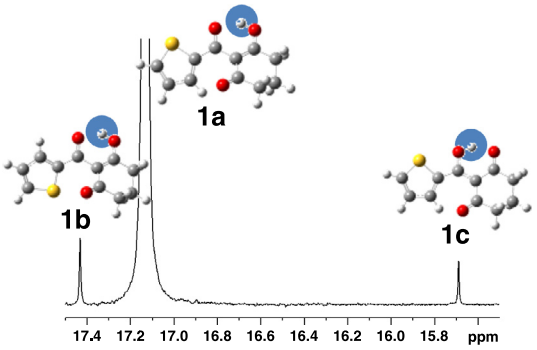
HIGHLIGHTS

- We report the first experimental study of the tautomerism of a 2-acylcyclohexane-1,3-dione.
- In theoretical calculations, monoenols showed the lowest energy.
- The endocyclic enol is the isomer in higher proportion.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 19 September 2013
Received in revised form 28 November 2013
Accepted 28 November 2013
Available online 3 December 2013

Keywords:
Acylcyclohexanediones
Tautomerism
NMR
Computational chemistry

ABSTRACT

The tautomeric system of 3-hydroxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one **1** has been investigated by NMR spectroscopy between 224 and 298 K. At all temperatures an endocyclic enol tautomer was the major isomer; however, at low temperatures two other enol isomers were found. Conformational search of the potential energy surfaces of all tautomers of cyclohexenone **1** was also carried out. Extensive calculations were performed for two triketones and four *cis*-endocyclic double bond enol tautomers with the lowest energies. Syntheses of 3-methoxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one **2** and 2-benzoyl-3-hydroxycyclohex-2-en-1-one **3** were carried out to analyze the features of thienyl group rotation and structural differences with a symmetrical substituent, respectively.

1. Introduction

The chemistry of 2-acylcycloalkane-1,3-diones has acquired increased importance due to the creation and wide application of a number of modern plant growth regulators based on 2-acylcyclohexane-1,3-dione derivatives [1–3]. The mode of action of those compounds is the inhibition of (4-hydroxyphenyl)pyruvate dioxygenase (HPPD), the enzyme that catalyzes the conversion of (4-hydroxyphenyl)pyruvate to homogentisate [4], which is a key step in the tyrosine catabolism pathway. One of the severe human hereditary diseases connected with the tyrosine catabolism is tyrosinemia type I [5], which is a fatal autosomal recessive disorder that causes hepatic failure, liver cirrhosis, and an early onset of primary liver cancer [6]. The biological activity of a molecule generally depends only of a particular isomer, among all possible isomers [7,8]. This bioactive conformation is not necessarily the most stable; nevertheless, it cannot be very high in energy since it would be excluded from the population of conformers in solution. In a great number of compounds, the biological activity is related to their tautomeric structure [9–11].

The keto-enol tautomerization is one of the most well studied topics both from experimental and theoretical viewpoints [12,13]. An important number of publications was focused on

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http://dx.doi.org/10.1016/j.molstruc.2013.11.057

tautomerism of 1,3-diones [14–21]. Nevertheless, 2-acylcyclohexane-1,3-diones are triketones that present several tautomeric forms and have been barely studied [22–24].

One of the ways of modifying the keto-enol tautomerism equilibrium is with the variation of solvent polarity. In solvents like D_2O and $CHCl_3$, the monoenol of β,β′-triketones is the most stable form (Fig. 1), whereas triketone form could not be detected [25].

On the other hand, theoretical calculations performed by Huang et al. on benzoylcyclohexane-1,3-dione and similar triketones [23] suggested two enol forms in solution, the exocyclic and endocyclic enols, but these have not been experimentally determined. It has been reported that substitution in position 2 of benzoyl group affects the interconversion of endocyclic and exocyclic enol forms [23]. The tautomeric equilibrium tends to favor the enol tautomer with endocyclic double bond, despite the substituent character. This trend is more obvious when solvent is considered. In addition, it has been found that the inhibitory activity of HPPD in animals increases when position 2 of benzoyl group is substituted, being the most active compound the one with a nitro group in such position [23,26,27]. The NTBC ((2-nitro-4-trifluoromethylbenzoyl)cyclohexane-1,3-dione) is a strong competitive inhibitor of HPPD. Crystallographic data of an analog of NTBC, 2-benzoyl-5,5-dimethylcyclohexane-1,3-dione [28], and other synthesized derivatives [24,27], like 2-(2-nitro-benzoyl)cyclohexane-1,3-dione, demonstrated that this kind of compounds exists predominantly as endocyclic enol.

To extend our knowledge of the tautomeric equilibrium of β-triketones, we have studied the tautomerism of a thienyl-carbonyl derivative of cyclohexane-1,3-dione. NMR measurements, supported by computational calculations, have provided new information on its solution structure. The present article documents the first experimental variable temperature-NMR determination of 2-acylcycloalkane-1,3-dione isomers, as well as the first computational study of energy barriers in such compounds.

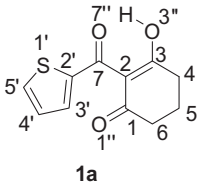


Fig. 2. 3-Hydroxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one **1a**.

Table 1
Representative isomers of **1** with their respective energies (kJ mol^{-1}, HF/6-31G(d)).

	Isomer	ΔE^A		Isomer	ΔE
Monoenol	 1a	0.0	Triketone	 1e	1.3
	 1b	10.6		 1f	10.9
	 1c	12.7	Dienol		100.8
	 1d	16.2			112.9
		44.8	Trienol		220.7
		48.1			222.4

[A] ΔE = Energy difference respect to **1a**.

2. Results and discussion

2.1. Conformational analysis

3-Hydroxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one (Fig. 2, enol form **1a**) has 8 possible tautomeric forms (one triketo, six keto-enol and one trienol). Considering the orientation of the OH hydrogen (in the same or the opposite side of carbonyl $C_7{=}O_{7''}$), the number of isomers ascends to 28, and taking into account the thienyl group rotation, to 56 (see Fig. S1).

All geometries of isomers were submitted to a complete optimization using HF/6-31G(d), where 44 of the total of 56 isomers evolved to stable structures (see Fig. S2 and Table S1). Table 1 shows a summary of some representative calculated isomers, grouped by tautomeric form. The most stable isomer was taken as reference to obtain the energy of stabilization relative to the other isomers. Monoenol and triketone forms were the most stable ones, whereas all dienols and trienols were 100–167 and 205–246 kJ mol^{-1} higher than monoenols, respectively.

The energy of the same enol form varied with the orientations of hydroxyl groups. In general, monoenols and dienols with an intramolecular hydrogen bond showed lower energies than the ones that do not have. In addition, *trans*-triketones were more stables than the *cis* ones (see Fig. S3 and Table S1).

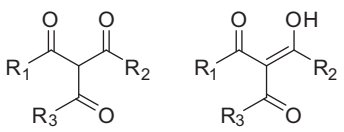


Fig. 1. β,β′-Triketone structure and their monoenol form.

Trans-triketones (**1e–f**) and 4 monoenols (**1a–d**), which can have an intramolecular hydrogen bond, had the lowest energies. These compounds were analyzed by different methods (HF, DFT) and bases set (6-31G(d), 6-31+G(d,p)) (Table S2). Endocyclic monoenol **1a** was the most stable in all used methods, indicating that this isomer could be the predominant tautomeric form of **1**. In all cases, rotamers that have the sulfur atom of the thienyl group opposite to carbonyl $C_7{=}O_{7''}$ were less stables than the ones in the same direction.

On the other hand, comparing optimized geometries obtained with different methodologies of calculation, the results were analogous. For ulterior analysis, we decided to work with HF/6-31+G(d,p), same as literature [23], for comparative purposes.

According to the calculated dipoles of *endo* and *exo* forms (Table 2), it is probable that the *endo* form (higher dipolar moment) increase in water. Energy calculations of these isomers in solution were in agreement with their dipolar moment.

Table 2
Relative energies (Δ*E*) of the most stable tautomers in solution (kJ mol^{-1}), with their respective dipolar moments (D).

Tautomer	ΔΔ*G*	Δ*E* ($CHCl_3$)	Δ*E* (H_2O)	[D]
1a	0.0	0.0	0.0	3.98
1b	7.0	5.2	4.6	2.88
1c	11.8	15.6	18.4	2.06
1d	16.0	19.8	22.8	1.07

2.2. Tautomerism and rotation energy barriers

Scheme 1 shows the proposed tautomeric interconversion between endocyclic enol (**1a, 1b**) and exocyclic enol forms (**1c**, **1d**), and the rotation process (**1a** → **1b** and **1c** → **1d**).

Tautomers with endocyclic double bond (**1a** and **1b**) are the most stable in gas phase, and both processes are endergonic (Fig. 3).

In the analysis of the reaction coordinate of thienyl group rotation for both endocyclic and exocyclic enols (**1a** and **1c**) (Scheme 1), two maxima were found, which correspond to transitional states (**TS3** and **TS4**, respectively). Dipolar moments of these transitional states were 3.81 D and 2.42 D, respectively.

By comparing with the rotation process (Fig. 3), the *endo–exo* tautomerization is more feasible to occur.

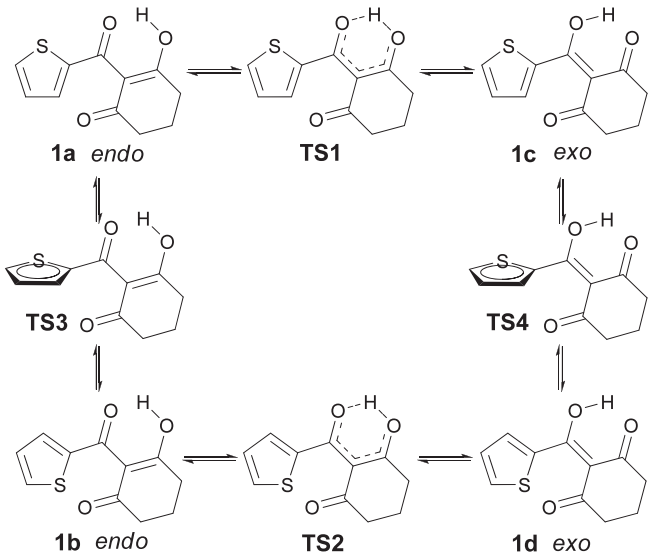
In order to determine the influence of solvation in tautomerization and rotation barriers of **1**, computational calculations with $CHCl_3$ and H_2O were carried out (Table 3). In both solvents the rotation barriers decreased, whereas the opposite effect was observed in tautomerism. Nevertheless, according to the reaction free energies, solvation effect was more important in tautomerism than in rotation.

Table 3
Gibbs free energies of interconversion (ΔG) and activation energies of tautomerization and rotation($\Delta G^{\ddagger}$) in different solvents. Values are given in kJ mol^{-1}.

	Tautomerization				Rotation			
	1a ↔ 1c		**1b ↔ d**		**1a ↔ 1b**		**1c ↔ 1d**	
	ΔG	$\Delta G^{\ddagger}$	ΔG	$\Delta G^{\ddagger}$	ΔG	$\Delta G^{\ddagger}$	ΔG	$\Delta G^{\ddagger}$
Gas	9.3	12.8	7.7	13.0	5.5	24.0	4.0	18.3
$CHCl_3$	13.1	17.5	13.4	18.4	3.7	19.8	4.0	13.0
H_2O	15.9	21.1	17.0	22.0	3.1	17.2	4.2	9.6

In summary, isomer **1a** was the most stable of all calculated isomers, among rotamers and tautomers, both in gas phase as in solution.

Since the energy barriers of rotation and tautomerism processes showed a small energy difference between them, both processes should be carried out at room temperature. Conformers' relationship was calculated using ΔG. Thus, the endocyclic isomer is expected to be found in a high excess, with a relation of **1a**:**1c** ≈ 98:2 and **1a**:**1b** ≈ 90:10.



Scheme 1.

2.3. NMR analysis

With the aim to determine experimentally the predominant tautomeric form of **1** in solution, different 1D and 2D NMR experiments were carried out. In its ^{1}H NMR spectrum at 298 K (Fig. 4), the highly deshielded peak at 17.28 ppm corresponds to an OH proton, characteristic for the enolic form of tricarbonylic systems with an intramolecular hydrogen bond [22,24,27]. A methine proton absence ($\delta \cong 6$ ppm) in ^{1}H NMR spectra rules out triketone form in solution. These results confirmed that **1** is fully enolized in solution at room temperature.

A number of factors can affect the enolization, e.g., nature of solvent, sample concentration and temperature. Additionally, the relation of the enol form is inversely proportional to the solvent polarity [17]. Hence, ^{1}H and ^{13}C experiments were performed at room temperature in $CDCl_3$, $(CD_3)_2CO$, CD_3CN and $(CD_3)_2SO$ (DMSO-d_6) (Table 4). Our results indicated that **1** is completely enolized in all the solutions studied, no matter the polarity of the solvent, because the signal corresponding to the keto form was

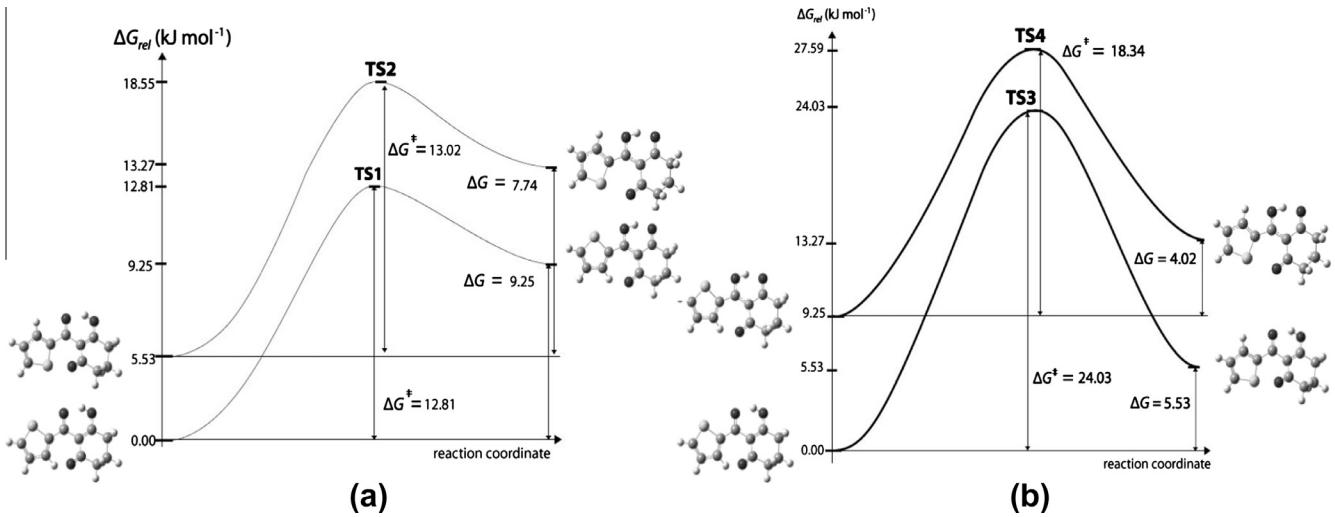


Fig. 3. Plot of energy barriers of tautomerization (a) and rotation (b) in gas phase.

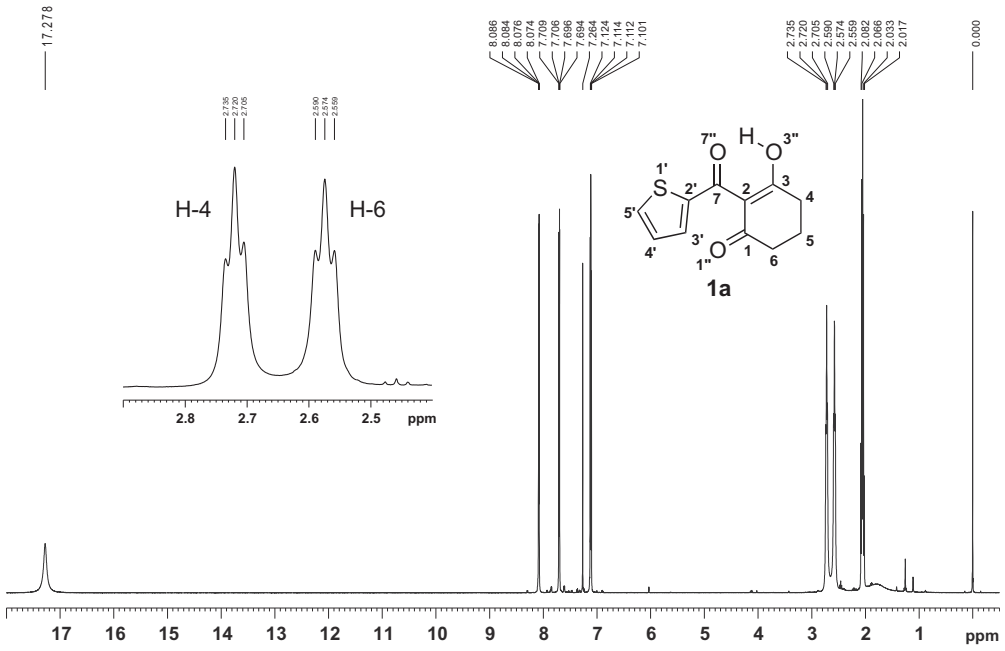


Fig. 4. ^{1}H NMR spectrum of **1** at 298 K in $CDCl_3$.

Table 4
Proton chemical shifts (ppm) of the enolic hydrogen of **1** in different solvents at room temperature.

Hydrogen	$CDCl_3$ (4.8)[A]	$(CD_3)_2CO$ (21.0)[A]	CD_3CN (36.6)[A]	DMSO-d_6 (47.2)[A]
3″ (s, 1H (OH))	17.28	16.10	15.79	11.76

[A] Dielectric constant (ε). Values taken from literature [29].

not observed. In the spectra, the shifts were changed due to solvent exchange.

Assignment of all carbons and hydrogens was accomplished by HSQC and HMBC (data recently published) [30]. Exocyclic carbon C_7 and hydrogen $H_{3'}$ of thienyl group were correctly assigned due to their mutual long-range correlation C–H (HMBC). A considerable difference of ca. 10 ppm can be noticed between the more deshielded carbons C_1 and C_3 respect to the exocyclic carbon C_7 [24] as shown in Table 5.

As can be seen in Fig. 4, both separated shielded triplet signals at ≈2.6 and 2.7 ppm correspond to methylene hydrogens (H_4 and H_6), confirming that each of these hydrogens have a different environment. In ^{13}C NMR spectrum, the carbons C_1 and C_3 appear as separated peaks, as well as C_4 and C_6. Unfortunately, it was unable to establish an accurate assignment of H_4, H_6 and quaternary carbons C_1 and C_3 at room temperature. Nevertheless, the higher chemical shift (196.44 ppm) is expected to be assigned to enol C_3, according to the information found in literature [16,24,27]. NOESY experiment at room temperature showed no correlation between the hydroxylic hydrogen with any proton, which made impossible to confirm the presence of the endocyclic enol as the major isomer through this experiment.

Table 5
^{1}H and ^{13}C chemical shifts (ppm) of **1** relative to TMS at 298 K.

Atom	δ_H (ppm), mult. (J (Hz))	δ_C (ppm)	COSY	HMBC
1		194.48		
2		112.78		
3		196.44		
4	2.72, t (6.2)	32.97	5, 6	2, 3, 5, 6
5	2.05, quint. (6.2)	19.02	4, 6	1, 3, 4, 5
6	2.57, t (6.2)	38.45	4, 5	1, 4, 5
7		187.37		
2′		141.07		
3′	8.08, dd (4.0, 1.1)	136.38	4′, 5′	7, 2′, 4′, 5′
4′	7.11, dd (5.0, 4.0)	127.63	3′, 5′	2′, 3′, 5′
5′	7.70, dd (5.0, 1.1)	135.49	3′, 4′	2′, 3′, 4′

A series of ^{1}H NMR experiments was performed, decreasing the temperature from 298 K to 224 K (Fig. 5). As the temperature went down, two new highly deshielded signals appeared in the range between 15 and 18 ppm, corresponding to hydroxylic hydrogens of other isomeric species (enol-**I** and enol-**II**). Proton signals corresponding to 17.16 and 17.44 ppm coalesced at 270 K, whereas the major peak corresponding to the OH at 15.75 ppm kept practically constant at the same chemical shift by varying the temperature.

Contrary to the room temperature spectrum, a NOE effect was observed between OH at 17.16 ppm and one of the methylene groups (2.72 ppm) in the NOESY spectrum at low temperature (Fig. 6). This correlation demonstrated that the major enol is endocyclic (**1a**, Scheme 1) and thus confirms the assignment of carbons C_1 and C_3 as well as hydrogens H_4 and H_6 (Table 5).

According to the calculated geometry of **1a**, hydrogens H_4 and H_5 were 2.45 Å between each other, whereas the distance between hydrogens H_4 and $H_{3''}$ was 3.36 Å. The relationship between the calculated NOE ($NOE_{4-5}/NOE_{4-3''} = r_{4-5}^{-6}/r_{4-3''}^{-6}$) with these distances was of approx. 7:1. Thus, the NOE effect between H_4 and H_5 is 7 times higher than for H_4 and $H_{3''}$. A series of NOESY spectra was performed at low temperature (218.5 K, Fig. 6). In this case, the NOE induced by hydrogens H_4 and $H_{3''}$ (comparing with H_4 and H_5) was proportional to the calculated distances, confirming the structure **1a**, where the endocyclic enol can form an intramolecular hydrogen bond. In the isomer where OH is not forming an

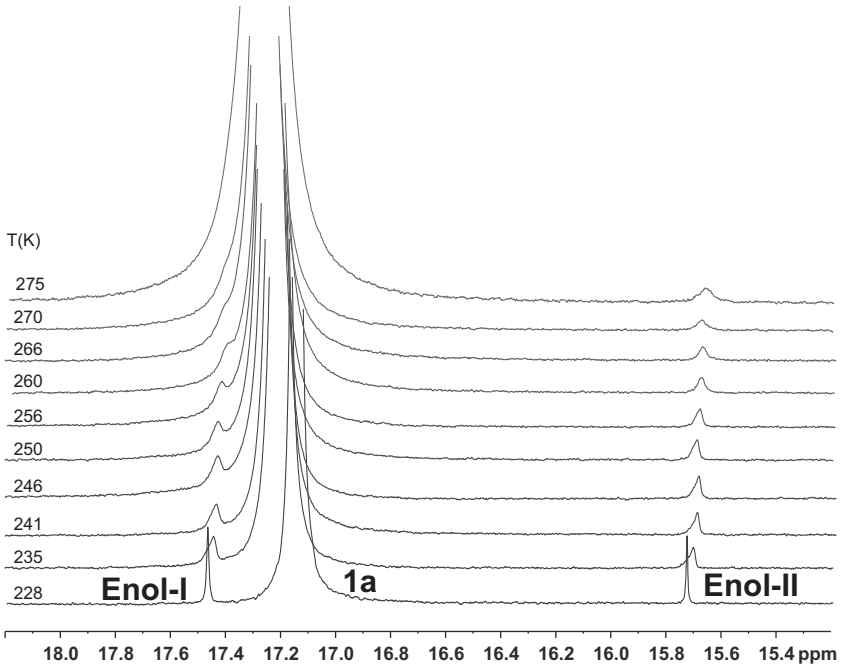


Fig. 5. ^{1}H NMR spectra of **1** in $CDCl_3$ at different temperatures.

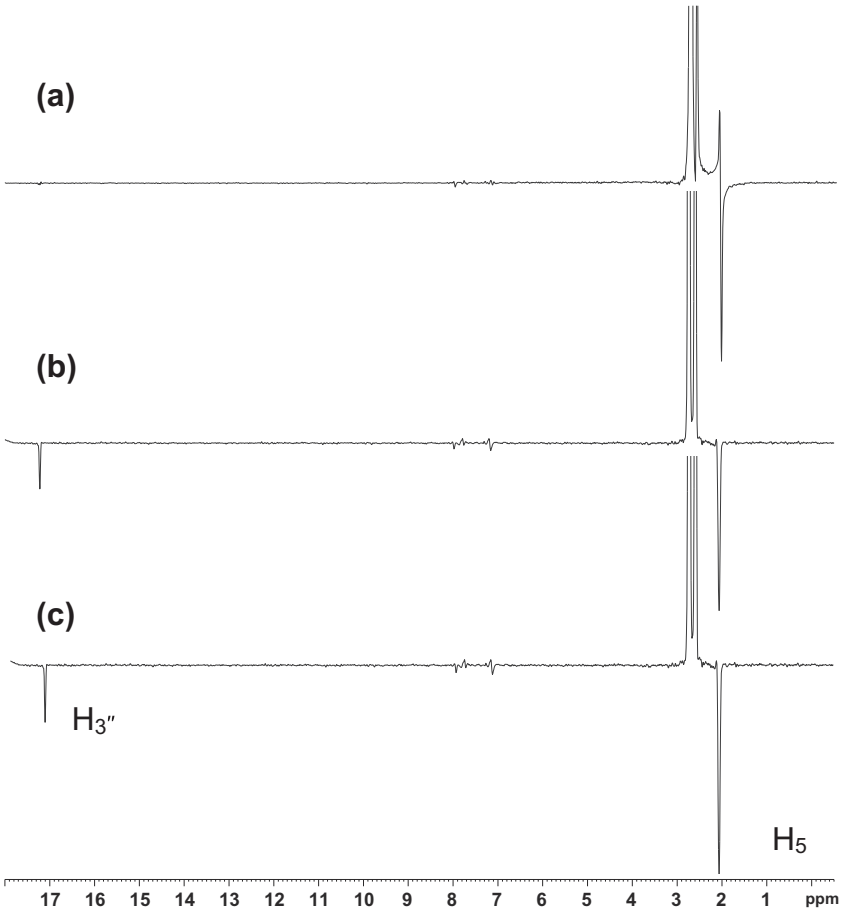


Fig. 6. NOESY spectra of **1** at 218.5 K using different mixing times: (a) 50, (b) 250 and (c) 900 ms [31].

intramolecular hydrogen bond (see Fig. S2, isomer 1K2,3-E(*t*)7KTh(*c*)), the calculated distance of $H_{3''}$–H_4 is 2.21 Å long; therefore, a NOE effect of similar intensity is expected to occur just as in H_4-H_5.

In the next step, an exhaustive analysis of the 2D spectra was carried out, with the aim to determine the structure of both minor isomers (enol-**I** and enol-**II**, Fig. 5) found at low temperature. Four of the calculated most stable isomers of **1** are shown in Scheme 1.

A HMBC spectrum at low temperature (224 K) is shown in Fig. 7. Interactions between carbons at 33.0, 112.8 and 196.4 ppm and hydroxyl of major species **1a** (17.16 ppm) and the most deshielded minor isomer (17.44 ppm, enol-**I**) are very similar. Hence, it can be implied that the carbon skeleton in the proximity of the OH hydrogen for both isomers is the same. Therefore, the structure assigned for enol-**I** was rotamer **1b**.

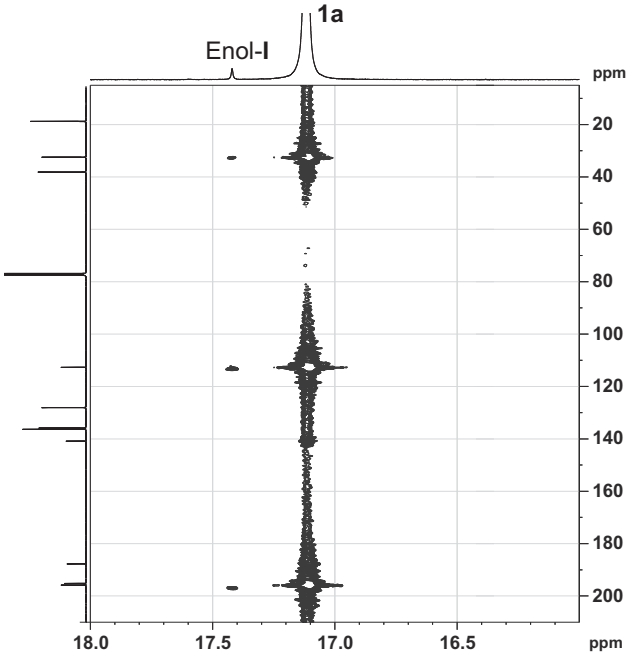


Fig. 7. HMBC spectrum of **1** at 224 K in $CDCl_3$.

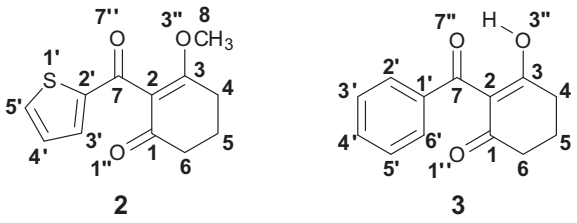


Fig. 8. 3-Methoxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one (**2**) and 2-benzoyl-3-hydroxycyclohex-2-en-1-one (**3**).

2.4. Synthesis of derivatives *2* and *3*

With the aim to block the keto-enol tautomerism of **1**, this compound was methylated with dimethylsulfate (DMS) according to a methodology based on the Khlebnicova procedure [32], generating 3-methoxy-2-(thienylcarbonyl)cyclohex-2-en-1-one (**2**) (Fig. 8), which could allow to analyze the features of the thienyl group rotation.

The main differences between spectra of **1** and **2** are the disappearance of a signal corresponding to an OH and the presence of a singlet signal corresponding to a methyl group at 3.78 ppm (Table 6). A NOE effect was also observed between methoxyl group hydrogens and methylene hydrogens H_4 in the NOESY experiment of **2** at room temperature, confirming the methoxyl group in C_3.

On the other hand, the hydrogen signal $H_{3'}$ of thienyl group in **2** is shifted 0.53 ppm to higher fields than in **1** (Fig. 9). This result could be explained by a ring current effect produced by the C_1 carbonyl group in **1**, whereas this effect is not observed in **2**.

Computational calculations indicated that thiophene-2-carbonyl substituent is orthogonal to the cyclohexenyl system in **2**, and its rotamers (**2a** and **2b**, Fig. S4) can coexist at room temperature. These structures could be very well correlated with the NMR experimental results. According to its relationship with **1**, this study allowed us to corroborate the coplanarity between the thiophene-2-carbonyl substituent and the endocyclic carbonyl of **1** (C_1=$O_{1''}$) and the interaction of this carbonyl group with $H_{3'}$.

In order to simplify the study of rotamers and tautomers of **1**, 2-benzoyl-3-hydroxycyclohex-2-en-1-one (**3**) was also synthesized [30]. NMR analysis of this compound could give clear information about the species present in solution, excluding the possibility of rotamers' signals due to the symmetry of phenyl substituent. Signals assignments of **3** are shown in Table 6. The signal at 16.96 ppm corresponded to a hydroxyl group, indicating that the major species is an enol with an intramolecular hydrogen bond. A NOE effect was also observed between hydroxyl $H_{3''}$ and methylene hydrogens H_4 in the NOESY experiment at 228 K, indicating that the enol is endocyclic. These results were comparable to the obtained for **1**.

From the ^{1}H NMR analysis of hydrogens $H_{2'}$ and $H_{6'}$ of phenyl group, we can observe that these protons are more shielded (7.51 ppm) than in acetophenone (7.94 ppm). Consequently, it can be inferred that the phenyl substituent is displaced from the plane of the molecule and does not have the influence of the carbonyl group.

Computational calculations of **3** were carried out in solution ($CHCl_3$) for a better comparison with NMR experiments. From the analysis similarly performed for **1**, we could determine that the most stable structure was **3a** (endocyclic enol), being 11.97 kJ mol^{-1} more stable than the exocyclic enol. In this conformation, the phenyl group is out of the plane. These results are in agreement with the experimental NMR measurements (Fig. 10).

In the ^{1}H NMR spectra of **3** at 224 K, a minor peak was observed at higher fields (16.11 ppm) than the major OH signal (16.96 ppm) (relation of 0.3% between each other), which corresponds to a hydroxyl group of an isomer of **3**. This peak remains at all the studied

Table 6
^{1}H and ^{13}C chemical shifts (ppm) comparison of **1** with **2** and **3** at room temperature.

		1	2	3 (OH)	4	5	6	7	8	2′	3′	4′	5′
1	δ_H	–	–	17.28, s, 1H	2.72, t, 2H	2.05, quint, 2H	2.57, t, 2H	–	–	–	8.08, dd, 1H	7.11, dd, 1H	7.70, dd, 1H
	δ_C	194.48	112.78	196.44	32.97	19.02	38.45	187.37	–	141.07	136.38	127.63	135.49
2	δ_H	–	–	–	2.68, t, 2H	2.12, quint, 2H	2.45, t, 2H	–	3.78, s, 3H	–	7.55, dd,1H	7.08, d, 1H	7.63, dd, 1H
	δ_C	196.01	120.31	175.08	25.89	20.61	36.46	187.08	56.66	145.13	133.84	128.18	134.31
		1	2	3 (OH)	4	5	6	7	–	1′	2′/6′	3′/5′	4′
3	δ_H	–	–	16.96, s, 1H	2.75, t, 2H	2.07, quint, 2H	2.50, t, 2H	–	–	–	7.51, m, 2H	7.39, m, 2H	7.49, dd, 1H
	δ_C	194.32	113.39	196.38	32.57	19.27	38.27	199.14	–	138.30	128.40	127.84	132.04

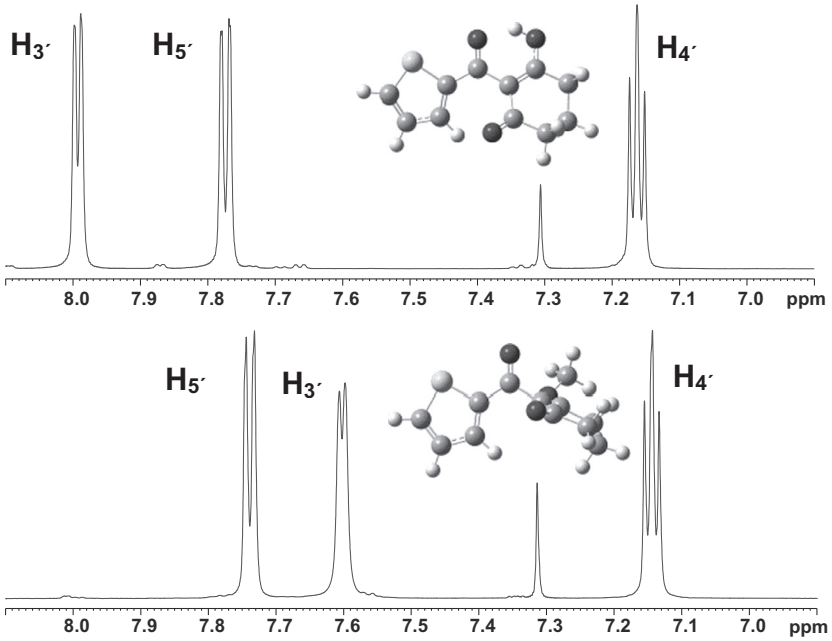


Fig. 9. ^{1}H NMR of **1** and **2** (224 K), amplified between 6.9 and 8.1 ppm.

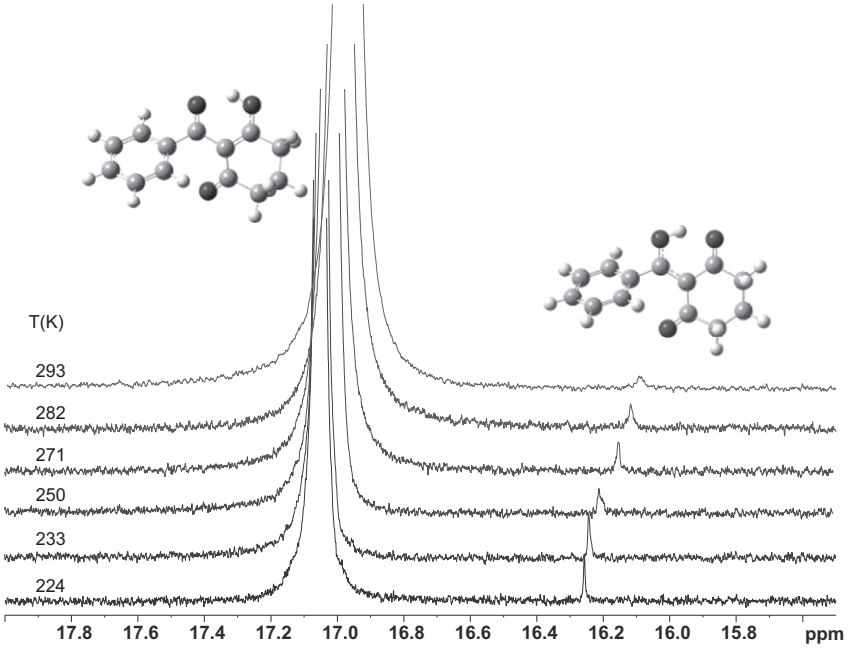


Fig. 10. ^{1}H NMR spectra of **3** in $CDCl_3$ at different temperatures.

temperatures (224–293 K, Fig. 10) and it does not coalesce. Thus, for this compound, it is possible the formation of the exocyclic enol **3c** (Fig. 10). Taking into account these findings, by analogy, the enol-**II** of **1** could be proposed to be the exocyclic enol **1c**.

3. Conclusions

This work represents the first experimental study of the tautomerism of a 2-acylcyclohexane-1,3-dione, using NMR and computational studies concurrently.

In theoretical calculations, monoenols showed the lowest energy, being the most stable the endocyclic tautomer with their respective rotamer. It was shown that the endocyclic enol **1a** is the isomer in higher proportion (>95%) from thermodynamic analysis, rotation study and tautomerism barriers. Nevertheless, the involved energies in both processes are quite small.

Analyzing NMR spectra, it is possible to establish that the predominant isomer is the endocyclic enol with the sulfur atom of thienyl ring in the same direction of the exocyclic carbonyl group.

At low temperatures, other two isomers were observed. The most deshielded signal corresponds to rotamer **1b**, whereas enol-II was assigned to the *exo* tautomer **1c**.

Thus, the first analysis of 2-acylcyclohexanediones involving other enol isomers is presented. The important tautomeric information gathered from the study here disclosed could be a

significant contribution for the investigation of interaction mechanisms comprising these compounds with the HPPD enzyme.

4. Experimental

4.1. Synthesis

Triketones **1** and **3** were synthesized by treating the appropriate acyl chloride with dried potassium cyanide in the presence of anhydrous acetonitrile under ultrasound irradiation at 50 °C to form the corresponding acylcyanide. Then, triethylamine and cyclohexane-1,3-dione were added in situ and the mixture was stirred overnight at room temperature, according to the conditions previously described [30].

4.2. Synthesis of 3-methoxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one **2**

Calcined K_2CO_3 (0.379 g; 0.274 mmol) and dimethyl sulfate (0.065 g; 0.051 mmol) were added to a solution of 3-hydroxy-2-(2-thienylcarbonyl)cyclohex-2-en-1-one (**1**) (0.102 g; 0.045 mmol) in absolute toluene (10 mL). The reaction mixture was boiled for 10 h, the solid was filtered off, and washed with toluene. After removing the toluene on the rotary evaporator, the residue was dissolved in ethyl acetate and then purified by chromatographic column using ethyl acetate:hexane [9:1] as eluent. White solid. ^{1}H NMR ($CDCl_3$): δ = 2.12 (quint., J = 6.5, 2H, H-5), 2.46 (t, J = 6.5, 2H, H-6), 2.68 (t, J = 6.5, 2H, H-4), 3.78 (s, 3H, H-8) 7.08 (dd, J = 4.9, 3.9, 1H, H-4′), 7.55 (dd, J = 3.9, 0.9, 1H, H-3′), 7.63 (dd, J = 4.9, 0.9, 1H, H-5′) ppm. ^{13}C NMR ($CDCl_3$): δ = 20.61 (C-5), 25.89 (C-4), 36.46 (C-6), 56.66 (C-8), 120.31 (C-2), 128.18 (C-4′), 133.84 (C-3′), 134.31 (C-5′), 145.13 (C-2′), 175.08 (C-3), 187.08 (C-7), 196.01 (C-1) ppm. MS (EI): m/z (%) = 236 $[M]^+$ (28), 203 (21), 180 (9), 151 (21), 139 (14), 123 (9), 111 (100), 97 (27), 83 (16), 55 (15).

4.3. Computational calculations

All calculations of the investigated species were performed using the Gaussian03 program package [33], employing HF and DFT (B3LYP) levels of theory, with 6-31+G(d,p) basis set for thermodynamic calculations. The solvent effect was taken into account using the PCM of Tomasi et al. [34]. The enol hydrogen was treated explicitly (radii taken from the UFF force field) [35]. Total energies were corrected for the zero-point energies and for thermal and entropic contributions at 298.15 K calculated at the same levels of theory as were used for optimization.

AM1 method [36] was used to obtain the initial geometries of the 8 tautomeric forms of **1**. Subsequently, a conformational analysis was carried out, considering all the possible torsion angles (see Fig. S1 in Supporting Information).

4.4. NMR

All spectra were taken with a Bruker Avance II spectrometer with a BVT3000 temperature controller (solution, 9 T, 400.16 and 100.56 MHz for ^{1}H and ^{13}C NMR, respectively), equipped with an inverse detection probe H-X (BBI) of 5 mm and a z-gradient coil for ^{1}H and ^{13}C. Chemical shifts (δ) are reported in ppm and coupling constants in Hz. Chemical shifts are given by internal solvents, $CDCl_3$ (7.26, 77.16), CD_3COCD_3 (2.05, 29.84), and DMSO-d_6 (2.50, 39.52) ppm for ^{1}H and ^{13}C, respectively. The internal standard was TMS for $CDCl_3$ and CD_3COCD_3, whereas in the other solvents, the signal corresponding to the undeuterated residual peak was used as standard.

The following parameters were used to measure ^{1}H NMR spectra: spectral width, 8.220 Hz (20 ppm); pulse P_1 = 7.5 μs, digital resolution, 0.39 Hz/point; number of scans, 16. For ^{13}C NMR spectra: spectral width, 20,500 Hz; pulse P_1 = 10.6 μs, digital resolution, 0.63 Hz/point; number of scans >2.000. In the case of ^{13}C NMR, partially saturated spectra were recorded with the application of WALTZ 16 proton decoupling. FIDs were multiplied by an exponential weight (lb = 1 Hz for ^{1}H and lb = 1.2 Hz for ^{13}C) before Fourier transformation.

Accessory publication

General procedure for the synthesis of these new compounds with their corresponding ^{1}H, ^{13}C and 2D NMR spectra and the computational calculations are available on the Journal's Website.

Acknowledgments

This work was financially supported by the National Research Council of Argentina (CONICET) and the Secretary of Science and Technology – Universidad Nacional de Córdoba (SECyT-UNC). G. M. C. thanks CONICET for a doctoral fellowship. The authors are grateful for the contribution of Ph. D. Walter J. Peláez and the technical assistance provided by Ph. D. Gloria Bonetto.

Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.molstruc.2013.11.057.

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