Deuterium Isotope Effects in ^{13}C NMR Spectra of *trans-N*-Salicylideneaniline Isotopomers

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A series of isotopically labelled *trans-N*-salicylideneaniline (SAN) iso-
topomers has been prepared (2'-^{2}H-, 3'-^{2}H-, 4'-^{2}H- and 2',3',4',5',6'-
^{2}H$_5$-SAN) and their ^{13}C NMR spectra recorded and analyzed. Deu-
terium isotope effects (DIEs) in ^{13}C NMR spectra of SAN isotopo-
mers have been compared with those observed in the correspond-
ing isotopomers of related binuclear aromatic molecules. Most of
DIEs in SAN resemble those previously reported for similar mole-
cular systems, except for some effects over three bonds. It is sug-
gested that the participation of SAN nitrogen lone pair in the in-
tramolecular hydrogen bond is responsible for the observed diffe-
rence in DIEs through three bonds at the aniline ring *ortho* carbon.

Key words: NMR, ^{1}H, ^{13}C, deuterium isotope effects, salicylidene-
aniline, hydrogen bond.

INTRODUCTION

Isotope effects on NMR spectral parameters have gained a considerable
attention due to their relation to the molecular structure and dynamics.[1–5]
Deuterium is particularly interesting because of the large fractional change

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in mass upon deuterium labelling (^{1}H/^{2}H ratio) and the relative ease of its introduction into a molecule. Using high magnetic field spectrometers, it became possible to detect minute long-range deuterium isotope effects (DIEs) in NMR spectra.[2] In that respect DIEs through six bonds ($^6\Delta_{ie}$) were recently[5,6] determined for a series of binuclear aromatic compounds with one phenyl group labelled by deuterium in *para* position and then used to estimate the >C=N–Ph or >C=C–Ph torsional angles according to the observed linear dependence of $^6\Delta_{ie}$ on the *ab initio* calculated >C=N–Ph or >C=C–Ph torsional angle.

trans-N-Salicylideneaniline (SAN, **1** in Scheme 1) was chosen as a prototype Schiff base with an intramolecular hydrogen bond. The aim of this study was to investigate isotope effects in a molecule that forms an intramolecular hydrogen bond and to compare them to those already found in the corresponding systems without a hydrogen bond.
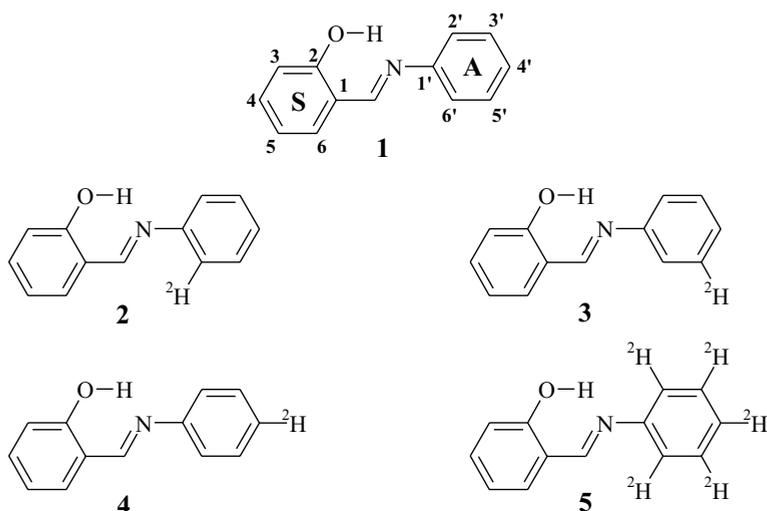
Scheme 1.

EXPERIMENTAL

NMR spectra were recorded on a Bruker AVANCE DRX500 spectrometer equipped with a *z*-gradient accessory (operating at 125.757 MHz for ^{13}C) from samples in 5 mm NMR tubes. Measurements were performed at 295 K with sample concentrations of 30 mg ml^{-1} in ^{2}H$_6$-benzene. ^{13}C signals were referred to TMS as internal standard. The standard pulse sequences with WALTZ-16 decoupling and relaxation delay of 1.5 s were used. Narrow region spectra with spectral widths of 8000 Hz (number of data points was 64K) were zero-filled to 128K, thus giving a digital resolution better than 0.07 Hz per point after Fourier transformation.

In order to study DIEs on ^{13}C chemical shifts, five deuterium labelled isotopomers of the model compound were prepared (Scheme 1). Enumeration of atoms and notation of phenyl rings is shown in Scheme 1, where **A** denotes the aniline and **S** the salicylidene ring. The sign of DIEs has been determined on the basis of equation

$$^n\Delta_{ie}{}^{13}C(^2H) = \delta\,^{13}C(H) - \delta\,^{13}C(^2H) \tag{1}$$

where »n« denotes the number of bonds between the observed atom and the isotopic substitution site, δ is chemical shift and Δ_{ie} is DIE (an experimental example for DIE determination can be found in Ref. 7). All prepared isotopomers were synthesized by addition of specifically deuterium labelled anilines[8] to salicylaldehyde.

Geometry optimizations and energy calculations were carried out using the DFT quantum-chemical method with the B3LYP functional[9] and the 6-31G(d) basis set using the Gaussian 94 program package.[10] In order to simulate experimental conditions by estimating the solvent influence on the molecular geometry and energy, two additional approaches, including theoretical treatment of the solvent-solute interactions, were employed: Onsager[11] and SCI-PCM[12] models. The solvent was represented by an infinite dielectric and polarizable continuum characterized by its dielectric constant (ε). A dielectric constant of 2.247 was used in calculations, representing deuterium labelled benzene. Relative energies of solvation (E_{sol} / kJ mol^{-1}) and the hydrogen bond ($E_{H\text{-}bond}$ / kJ mol^{-1}) were obtained using equations (2) and (3), respectively.

$$E_{sol} = E_{(\text{Onsager or SCI-PCM})} - E_{(in\ vacuo)} \tag{2}$$

$$E_{H\text{-}bond} = E_{(\text{with H-bond})} - E_{(\text{without H-bond})} \tag{3}$$

$E_{(\text{Onsager or SCI-PCM})}$, $E_{(in\ vacuo)}$, $E_{(\text{with H-bond})}$, and $E_{(\text{without H-bond})}$ are the total energies calculated.

RESULTS AND DISCUSSION

All DIEs presented here are secondary isotope effects (hereafter referred to only as isotope effects). Owing to the observation that some DIEs in NMR spectra of SAN isotopomers were dependent on the solvent used, here we report the results obtained for SAN in deuterium labelled benzene, only. Solvent dependence of DIEs will be treated in a separate paper.[13] Furthermore, Katritzky and coworkers[14] recently reported also on the temperature dependence of DIEs in a series of salicylaldehyde Schiff bases. They studied isotopomers in which an intramolecular hydrogen-bonded hydrogen atom of the salicylidene ring hydroxyl group was replaced by deuterium and observed that temperature effects were not larger than 5 ppb.[14] Therefore, to check whether DIEs in SAN isotopomers **4** and **5** are affected by temperature, NMR spectra were measured at 20 °C and 60 °C. The difference of 1 ppb was the largest one found between the corresponding observed DIEs at these

two temperatures and was considered non-significant. It was concluded that, in this temperature range, isotope effects in SAN are temperature independent in benzene-d_6.

DIEs ($^n\Delta_{ie}$ or $^{t(n)}\Delta_{ie}$ / ppb, where t denotes the total isotope effect arising from all present deuterium atoms in a molecule) in ^{13}C NMR spectra of deuterium labelled SAN isotopomers are presented in Table I. A variety of short range DIEs (*i.e.*, effects up to three bonds, $^3\Delta_{ie}$) was observed. Most of them resemble those found in the corresponding *trans-N*-benzylideneaniline (tBA) isotopomers. The only exception is the effect at C-6' in **2**, which is now –5.7 ppb compared to –10.0 ppb in the related 2'-^{2}H-tBA isotopomer. This can be rationalized by a change of interaction involving nitrogen and *ortho* hydrogen/deuterium of the aniline ring, since the nitrogen lone pair in SAN participates in the intramolecular hydrogen bond (Scheme 2).

In general, the observed values of short range DIEs are similar to those found in the related aromatic systems of the Ph–R and Ph–Z–Ph types (where Ph is phenyl and R stands for –CH_3, –NH_2, –CHO, –COOH, and –$COCH_3$, while Z is –CH=CH–, –CH=N–, >C=O, and –N=N–).[5]

2'-^{2}H-tBA   2'-^{2}H-SAN
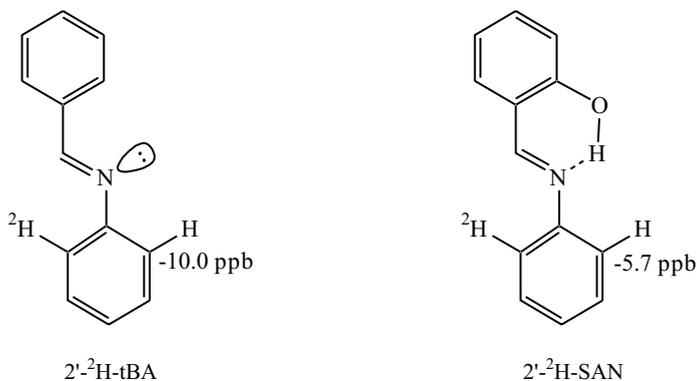
Scheme 2.

From Table I one can see that only a few long-range DIEs in SAN have been detected. Accordingly, only two long-range DIEs were observed in each of the mono-deuterium labelled isotopomers **2**, **3** and **4.** The longest range of all DIEs is over six bonds, *i.e.*, at C-   in **4**. The same was observed also for the corresponding tBA isotopomer.[15] In contrast, the range of DIEs is much longer, even up to ten bonds,[5] for *para* mono-deuterium labelled isotopomers in other related binuclear aromatic compounds.

However, four long-range DIEs we observed in isotopomer **5** with one per-deuterium labelled phenyl ring. Three of them, at C-1 (–4.5 ppb), C-4

TABLE I

Deuterium isotope effects ($^{n}\Delta_{ie}$ or $^{t(n)}\Delta_{ie}$ / ppb)[a] in ^{13}C NMR spectra of deuterium labelled *trans-N*-salicylideneaniline isotopomers

Site	Δ_{ie} / ppb			
	2'-^{2}H (**2**)	3'-^{2}H (**3**)	4'-^{2}H (**4**)	$^2H_{5(A)}$ (**5**)
C-	$^4\Delta_{ie} = 3.2$	$^5\Delta_{ie} = 3.4$	$^6\Delta_{ie} = 3.2$	$^{t(4)}\Delta_{ie} = -4.9$
C-1				$^{t(5)}\Delta_{ie} = -4.5$
C-1'	$^2\Delta_{ie} = 58.4$	$^3\Delta_{ie} = 3.1$	$^4\Delta_{ie} = -4.1$	$^{t(2)}\Delta_{ie} = 121.5$
C-2				
C-6				$^{t(6)}\Delta_{ie} = 6.5$
C-2'	$^1\Delta_{ie} = 296.5$	$^2\Delta_{ie} = 103.7$	$^3\Delta_{ie} = 7.6$	$^{t(1)}\Delta_{ie} = 405.5$
C-6'	$^3\Delta_{ie} = -5.7$	$^4\Delta_{ie} = 4.6$	$^3\Delta_{ie} = 7.6$	$^{t(1)}\Delta_{ie} = 405.5$
C-3				
C-5				
C-3'	$^2\Delta_{ie} = 109.5$	$^1\Delta_{ie} = 283.5$	$^2\Delta_{ie} = 111.2$	$^{t(1)}\Delta_{ie} = 502.5$
C-5'	$^4\Delta_{ie} = 5.0$		$^2\Delta_{ie} = 111.2$	$^{t(1)}\Delta_{ie} = 502.5$
C-4				$^{t(8)}\Delta_{ie} = 8.7$
C-4'	$^3\Delta_{ie} = 4.8$	$^2\Delta_{ie} = 110.2$	$^1\Delta_{ie} = 288.5$	$^{t(1)}\Delta_{ie} = 513.5$

[a] n denotes the number of bonds between the carbon under observation and the closest deuterium atom, while t denotes the total (cumulative) isotope effect arising from all deuterium atoms present in a molecule; standard deviation is better than 2.0 ppb for $^1\Delta_{ie}$ and $^{t(1)}\Delta_{ie}$, and better than 1.0 ppb for the other effects.

(8.7 ppb) and C-6 (6.5 ppb), were not observed at the corresponding carbon atoms in the mono-deuterium labelled isotopomers. Effects at these carbons in **5** arise from the simultaneous action of five deuterium atoms, while in mono-deuterium labelled molecules from one deuterium only, and hence are too small to be detected.

Out of all long range DIEs ($>^3\Delta_{ie}$), the effect over six bonds ($^6\Delta_{ie}$) was previously found to be useful in conformational studies. Namely, it has been reported[5,6] that $^6\Delta_{ie}$ in *para*-deuterium labelled isotopomers of the Ph–Z–Ph type compounds can be correlated with the *ab initio* calculated torsional angle φ of the $-C_{2'}=C_{1'}-N=C$ – (Scheme 1) or $-C_2=C_1-C$ =C – intervening bond fragments. The –CH=N–Ph torsional angle in SAN was estimated by the above mentioned correlation[6] using $^6\Delta_{ie}$ in isotopomer **4** and the value of 63.8 was obtained. On the other hand, this torsional angle was calculated using different quantum-chemical approaches, starting from molecular conformations with and without the intramolecular hydrogen bond (Scheme 3). The obtained values are given in Table II. In order to reproduce experimental

TABLE II

DFT (B3LYP/6-31G*) calculated values of –CH=N–Ph and Ph–CH=N– torsional angles (φ) for *trans-N*-salicylideneaniline with and without the hydrogen bond[a]

Torsional angle	with H-bond	without H-bond
	in vacuo	
$\varphi_{(N\text{-}Ph)}$ / °	33.302	37.119
$\varphi_{(C\text{-}Ph)}$ / °	0.375	8.474
	Onsager	
$\varphi_{(N\text{-}Ph)}$ / °	33.251	37.094
$\varphi_{(C\text{-}Ph)}$ / °	0.330	8.335
	SCI – PCM	
$\varphi_{(N\text{-}Ph)}$ / °	32.446	37.721
$\varphi_{(C\text{-}Ph)}$ / °	0.192	9.499

[a] N-Ph stands for –CH=N–Ph, and C-Ph for Ph–CH=N–.

conditions, benzene was selected as a solvent in calculations using the Onsager and SCI-PCM models. Relative energies of solvation and the hydrogen bond calculated by these models are collected in Table III. These calculations demonstrate that the solvation effect stabilizes the molecule in both cases, *i.e.*, with and without the intramolecular hydrogen bond. In the former case, the stabilization is accompanied by a small decrease (less than 1°) in –CH=N–Ph and Ph–CH=N– torsional angles (Table II). In the latter case, the decrease is even smaller (0.1° or less) in the Onsager model, while an increase in torsional angles (*ca.* 1°) was calculated by the SCI-PCM model. The difference in torsional angles upon solvation obtained in the Onsager and SCI-PCM models reflects changes in the relative energies of solvation, as well as that of H-bond. It should be noted that in the case of the hydrogen bonded structure (Scheme 3), the salicylidene ring and imino moiety are

SAN without H-bond   SAN with H-bond
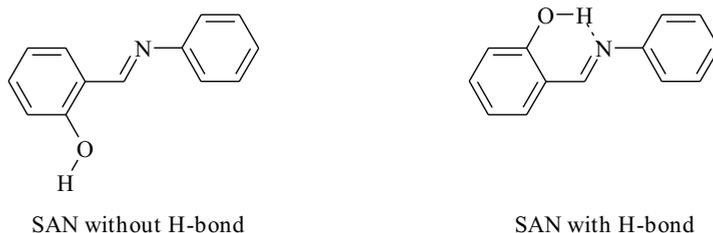
Scheme 3.

TABLE III

DFT (B3LYP/6-31G*) calculated values of relative energies of solvation
(E_{sol} / kJ mol^{-1})[a] and the hydrogen bond (E_{H-bond} / kJ mol^{-1})[b] in
trans-*N*-salicylideneaniline[c]

Relative energy	with H-bond	without H-bond
	in vacuo	
$_{H-bond}$ / kJ mol^{-1}	−48.60	−
	Onsager	
$_{sol}$ / kJ mol^{-1}	−0.82	−0.21
$_{H-bond}$ / kJ mol^{-1}	−49.21	−
	SCI − PCM	
$_{sol}$ / kJ mol^{-1}	−6.57	−8.98
$_{H-bond}$ / kJ mol^{-1}	−46.19	−

[a] $E_{sol} = E_{(Onsager\ or\ SCI-PCM)} − E_{(in\ vacuo)}$.
[b] $E_{H-bond} = E_{(with\ H-bond)} − E_{(without\ H-bond)}$ (for each model separately).
[c] Minus sign denotes lower energy (stabilization); $E_{(Onsager\ or\ SCI-PCM)}$, $E_{(in\ vacuo)}$, $E_{(with\ H-bond)}$, and $E_{(without\ H-bond)}$ are the total energies calculated.

nearly coplanar, while the angle between these two planes is 8.5°–9.5° when there is no intramolecular hydrogen bond. It can be clearly seen from Table II that the –CH=N–Ph torsional angles do not exceed 38° in any of the calculations used. Therefore, it is suggested that the difference between the values of the –CH=N–Ph torsional angle calculated by DFT approach and the above mentioned correlation arises from the hydrogen bonding influence on the magnitude of isotope effects. As already reported for benzoic acid isotopomers,[16] electron density redistribution induced by the hydrogen bonding affects the overall magnitude of isotope effects. Since the magnitude of isotope effects reflects the presence of the intramolecular hydrogen bond in SAN, $^6\Delta_{ie}$ in SAN isotopomer **4** cannot fit into the correlation and hence cannot be used to estimate the –CH=N–Ph torsional angle in SAN. The same is also expected to hold for other similar systems with intramolecular hydrogen bond(s).

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SAŽETAK

Deuterijski izotopni učinci u ^{13}C NMR spektrima izotopomera *trans-N*-salicilidenanilina

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Pripravljen je niz izotopno obilježenih (2'-^{2}H-, 3'-^{2}H-, 4'-^{2}H- i 2',3',4',5',6'-^{2}H$_5$-) izotopomera *trans-N*-salicilidenanilina (SAN) te su snimljeni i analizirani njihovi ^{13}C NMR spektri. Deuterijski izotopni učinci (DIE) u ^{13}C NMR spektrima izotopomera SAN uspoređeni su s izotopnim učincima opaženima u odgovarajućim izotopomerima srodnih binuklearnih aromatskih molekula. Većina opaženih DIE u skladu je s odgovarajućim ranije opisanim učincima u sličnim molekulskim sustavima, osim nekih efekata kroz tri veze. Opažena razlika DIE kroz tri veze na *orto*-atomu ugljika anilinskog prstena objašnjena je sudjelovanjem slobodnog para elektrona na atomu dušika u intramolekulskoj vodikovoj vezi.