1-Phenyl-3-methyl-5-*N*-benzylideneaminopyrazoles. Substituent effects and protonation sites studied by NMR and *ab initio* (6-31G*) MO calculations

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1-Phenyl-3-methyl-5-*N*-benzylideneaminopyrazole and its derivatives 11 prepared by condensation of 1-phenyl-3-methyl-5-aminopyrazole and aromatic aldehydes have been studied by multinuclear (^{1}H, ^{13}C, $^{14/15}$N and ^{17}O) magnetic resonance spectroscopy. The ^{13}C NMR chemical shifts and the direct spin–spin coupling constants ^1J(C,H) of the azomethine carbon of these Schiff bases (SB) correlate significantly with the Hammett substituent constants, σ_p, of the *para*-substituents in the aryl ring bound to the azomethine carbon. The assignments of the ^{15}N NMR chemicals shifts of SBs in $CDCl_3$ were based on ^2J(N,H)s observed for the azomethine nitrogen as well as ^{1}H, ^{15}N HMBC experiments. Based on the present ^{1}H, ^{13}C and ^{15}N NMR data these SBs can be transformed to single and double protonated forms in trifluoroacetic acid (TFA). The protonation sites (the first one at the unsubstituted nitrogen of the pyrazole ring and the second one at the azomethine nitrogen) deduced from the NMR data are supported by *ab initio* MO calculations at HF/6-31G* level with a full geometry optimization performed for a model compound, 1,3-dimethyl-5-*N*-benzylideneaminopyrazole.

Introduction

Schiff bases (SB) showing unique protonation properties,[1–3] occurring as structural fragments in significant chromophores[2] and intermediates in many enzymatic reactions[4] are of continuing interest. The main part of the studies on SBs deals with the *N*-benzylideneanilines[1,3–9] while few reports describe other structures like aliphatic[2] or heteroaromatic systems.[10–13] Especially interesting are pyrazole derivatives[11–13] used as versatile building blocks for photochemically and antibacterially important bispyrazolopyridines.[14–17]

The biological activity of pyrazole itself and its derivatives is well known.[12,18] Recently, ^{13}C NMR parameters of pyrazoles have been exhaustively reviewed by Begtrup *et al.*[19] Similarly, the protonation sites of pyrazole and its amino derivatives have been determined by ^{13}C NMR spectroscopy.[20] Recently, ^{15}N NMR studies on pyrazole and its derivatives have also received remarkable attention.[21–24] Quantum chemical *ab initio* studies at 6-31G level on substituted pyrazoles including the correlation of electronic parameters with ^{15}N NMR characteristics also have been reported.[22]

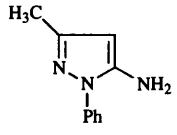
So, it is reasonable to expect that NMR spectroscopy together with *ab initio* MO calculations could provide a deep insight into the structural and protonation characteristics of the present series of SBs.

Experimental

Synthesis of the compounds 1–13

1-Phenyl-3-methyl-5-aminopyrazole, $C_{10}H_{11}N_3$ (1). Compound **1** was synthesized as described previously.[25,26] The structure and purity of **1** were checked by its melting point as well as ^{1}H, ^{13}C NMR and mass spectra. Condensation reactions of commercially available aromatic aldehydes with **1** were done by refluxing a 1:1 molar mixture of starting materials in ethanol for 3 h.

1-Phenyl-3-methyl-5-*N*-benzylideneaminopyrazole (=SB), $C_{17}H_{15}N_3$ (2). Compound **2**, mp (lit. value in parentheses)/°C 68–72 (oil[14]), crystallized from hexane–AcOEt (v:v 5:1) with a 57.5% yield, elemental analysis (calc. values in parentheses)/%: C 78.13 (78.05), H 5.79 (5.70), N 16.08 (15.92).



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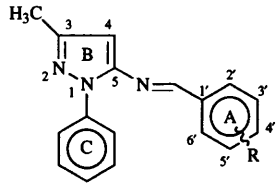
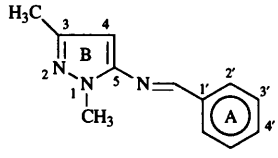


2–13



Model compund **14**

***p*-F-SB, $C_{17}H_{14}N_3F$ (3).** Compound **3**, mp/°C oil (new compound), crystallized from hexane–AcOEt (v:v 5:1) with a 51.0% yield, elemental analysis not done owing to traces of starting materials observed in ^{1}H NMR spectrum.

***p*-Me-SB, $C_{18}H_{17}N_3$ (4).** Compound **4**, mp/°C 78–78.5 (new compound), crystallized from EtOH–H_2O (v:v 5:1) with a 40.5% yield, elemental analysis/%: C 78.52 (78.43), H 6.22 (6.31), N 15.26 (15.01).

***p*-MeO-SB, $C_{18}H_{17}N_3O$ (5).** Compound **5**, mp/°C oil (oil[14]), crystallized from hexane–AcOEt (v:v 5:1) with a 44.0% yield, elemental analysis not done owing to traces of starting materials observed in the ^{1}H NMR spectrum.

***p*-NMe_2-SB, $C_{19}H_{20}N_4$ (6).** Compound **6**, mp/°C 113–115 (172–174[14]), crystallized from EtOH–H_2O (v:v 5:1) with a 45.0% yield, elemental analysis/%: C 74.97 (75.08), H 6.62 (6.41), N 18.41 (18.62).

***p*-Cl-SB, $C_{17}H_{14}N_3Cl$ (7).** Compound **7**, mp/°C 99.5–101 (102–103[14]), crystallized from EtOH–H_2O (v:v 5:1) with a 76.0% yield.

***p*-Br-SB, $C_{17}H_{14}N_3Br$ (8).** Compound **8**, mp/°C 91–93 (new compound), crystallized from EtOH with a 59.5% yield, elemental analysis/%: C 60.02 (60.19), H 4.15 (4.31), N 12.35 (12.14).

***p*-NO_2-SB, $C_{17}H_{14}N_4O_2$ (9).** Compound **9**, mp/°C 135–136.5 (134–135[14]), crystallized from EtOH with an 80.5% yield.

***p*-$MeCO_2$-SB, $C_{19}H_{17}N_3O_2$ (10).** Compound **10**, mp/°C 86–87.5 (new compound), crystallized from EtOH–H_2O (v:v 5:1) with a 49.5% yield, elemental analysis/%: C 71.46 (71.32), H 5.36 (5.22), N 13.16 (13.24).

***p*-CN-SB, $C_{18}H_{14}N_4$ (11).** Compound **11**, mp/°C 135–137 (new compound), crystallized from EtOH–H_2O (v:v 5:1) with an 82.0% yield, elemental analysis/%: C 75.50 (75.39), H 4.93 (5.12), N 19.57 (19.63).

***o*-OH-SB, $C_{17}H_{15}N_3O$ (12).** Compound **12**, mp/°C 110–112 (new compound), crystallized from EtOH–H_2O (v:v 5:1) with an 83.5% yield, elemental analysis/%: C 73.63 (73.58), H 5.45 (5.39), N 15.15 (15.01).

***o*-Br-SB, $C_{17}H_{14}N_3Br$ (13).** Compound **13**, mp/°C 82.5–85 (new compound), crystallized from EtOH–H_2O (v:v 5:1) with a 61.5% yield, elemental analysis/%: C 60.02 (60.24), H 4.15 (3.98), N 12.35 (12.30).

NMR spectroscopy

All 1-D NMR and 2-D ^{13}C–^1H HETCOR measurements were done with a Jeol JNM GSX-270 FT NMR spectrometer working at 270.17 MHz in ^1H, 67.94 MHz in ^{13}C, 36.63 MHz in ^{17}O and 27.38 MHz in ^{15}N NMR experiments, respectively. All samples were measured for 0.5 M solutions in $CDCl_3$ (99.5%, Reidel-de-Haën). The ^1H and ^{13}C NMR measurements were done in 5 mm diameter NMR tubes using a dual C/H probehead and ^{17}O and ^{15}N NMR experiments in 10 mm diameter NMR tubes using a tunable multinuclear probehead. The protonation experiments were done with trifluoroacetic acid, TFA (Fluka AG), in varying concentrations. Internal reference $SiMe_4$ ($\delta = 0$ ppm) for ^1H and ^{13}C NMR. External reference H_2O ($\delta = 0$ ppm) in 1 mm diameter capillary tube inserted coaxially inside the 10 mm NMR tube for ^{17}O NMR. Reference CH_3NO_2 ($\delta = 0$ ppm) measured at 30 °C in 10 mm diameter tube for ^{15}N NMR.

In ^1H NMR measurements the spectral width was 3500 Hz and the number of data points 32 K giving a digital resolution of 0.21 Hz. The number of scans (NS) was 4 with a pulse delay of 1 s and a flip angle of 90°. The FIDs were digitally filtered using an exponential window function of the digital resolution prior to FT to improve the signal to noise ratio (S/N) in the frequency spectra. The ^1H spectra were analysed on a first-order basis.

In ^{13}C NMR measurements the spectral width was 15 000 Hz and the number of data points 32 K giving a digital resolution of 0.92 Hz. NS for proton broad band decoupled (BBD) spectra was 100 and for fully coupled spectra *ca.* 8000. The pulse delay in BBD experiments was 3 s and in experiments without BBD 1 s. The FIDs were digitally filtered using an exponential window function of the digital resolution prior to FT to improve S/N in the frequency spectra.

The ^{13}C–^1H HETCOR experiments transmitted *via* $^1J(C,H)$ couplings were performed using a standard pulse sequence. The spectral widths/data points were 1200 Hz/512 (^{13}C) and 350 Hz/128 (^1H), respectively. The FIDs of both directions were digitally filtered by the exponential window functions of the digital resolutions prior to FT to improve S/N in the 2-D contour map.

In ^{17}O NMR measurement the spectral width was 36 000 Hz and the number of data points 8 K giving a digital resolution of 9 Hz and an acquisition time of 0.11 s. The pulse delay was set to 10 ms. NS for BBD spectrum was 400 000. The FIDs were digitally filtered using an exponential window function of 50 Hz prior to FT to improve S/N in the frequency spectra.

In 1-D ^{15}N NMR measurements the spectral width was 15 000 Hz and the number of data points 64 K giving a spectral resolution of 0.46 Hz. NS in inverse gated proton decoupled experiments (to prevent NOE) was *ca.* 1000 and pulse delay 30 s. In proton coupled spectra NS > 2000. The FIDs were digitally filtered using an exponential window function of the digital resolution prior to FT to improve S/N in the frequency spectra. The inverse 2-D ^1H, ^{15}N HMBC (heteronuclear multiple bond coherence) field gradient (FG) enhanced experiments with WALTZ decoupling were performed with a Jeol Eclipse 400 spectrometer working at 399.78 MHz. The matrix size was 1024 (^1H) × 64 (^{15}N).

The ^{14}N NMR experiments were done with a Bruker Avance DRX500 spectrometer working at 36.14 MHz. The spectral width was 50 000 Hz, number of data points 16 K giving a 3.0 Hz digital resolution and NS > 200 000. Reference CH_3NO_2 ($\delta = 0$ ppm) measured at 30 °C in 10 mm diameter tube for ^{14}N NMR.

Ab initio MO calculations

Ab initio MO calculations were carried out with the GAUSSIAN92[27] program set running in VAX cluster computers of University of Jyväskylä. A full geometry optimization for neutral and four protonated forms of model compound **14** was obtained with gradient techniques. All calculations were performed at the HF/6-31G* level of theory.

Results and discussion

The ^1H and ^{13}C NMR chemical shifts of **1–13** in $CDCl_3$ are given in Tables 1 and 2, respectively. The assignment of ^1H and ^{13}C NMR chemical shifts of 1-phenyl-3-methyl-5-aminopyrazole **1** and SBs **2–13** is based on the literature data,[14,28] characteristic substituent chemical shifts (SCS), mutual comparisons as well as fully coupled ^{13}C NMR spectra and ^{13}C, ^1H-HETCOR experiments.

Table 1 ^1H NMR chemical shifts (δ from internal $SiMe_4$) of compounds **1–13** in 0.5 M $CDCl_3$ solution at 30 °C

		Ring *A*				Ring *B*		Ring *C*		
No	R	H-2/6	H-3/5	Others	=CH	H-4	CH_3	H-2/6	H-3/5	H-4
1		—	—	—	—	5.44	2.33	7.54	7.45	7.31
2	H	7.39	7.76	7.38	8.51	6.09	2.33	7.75	7.40	7.24
3	F	7.71	7.06	—	8.44	6.06	2.32	7.73	7.39	7.26
4	Me	7.20	7.69	2.34	8.51	6.09	2.35	7.76	7.41	7.25
5	OMe	6.90	7.73	3.78	8.48	6.07	2.34	7.77	7.40	7.26
6	NMe_2	6.61	7.61	2.93	8.41	6.02	2.33	7.82	7.39	7.25
7	Cl	7.36	7.70[a]	—	8.49	6.12	2.34	7.71[a]	7.44	7.28
8	Br	7.51	7.62	—	8.46	6.11	2.34	7.71	7.41	7.30
9	NO_2	7.92	8.23	—	8.62	6.23	2.36	7.69	7.44	7.31
10	CO_2Me	7.83	8.06	—	8.57	6.17	2.35	7.72	7.52	7.32
11	CN	7.85	7.66[a]	—	8.56	6.20	2.35	7.67[a]	7.43	7.30
12	*o*-OH	*b*	*b*	*b*	8.61	6.17	2.34	7.72	7.52	7.32
13	*o*-Br	*c*	*c*	*c*	8.91	6.15	2.33	7.71	7.38	7.24

[a] Overlapping signals. [b] 6.91 (H-3), 7.34 (H-4), 6.89 (H-5) and 7.30 ppm (H-6). [c] 7.48 (H-3), 7.14 (H-4), 7.24 (H-5) and 8.00 ppm (H-6).

Table 2 ^{13}C NMR chemical shifts (δ from internal $SiMe_4$) of compounds **1–13** in 0.5 M $CDCl_3$ solution at 30 °C

		Ring *A*					Ring *B*				Ring *C*			
No	R	C-1	C-2	C-3	C-4	=CH	C-3	C-4	C-5	CH_3	C-1	C-2	C-3	C-4
1		—	—	—	—	—	149.4	90.8	145.3	13.9	138.8	123.9	129.4	127.0
2	H	135.8	128.7	129.0	131.7	160.0	149.0	93.3	150.3	14.1	139.5	124.0	128.5	126.3
3	F	131.1	128.9	116.0	164.9	158.5	149.1	93.3	150.2	14.1	139.5	124.0	128.5	126.4
4	Me[a]	133.5	129.6	129.1	142.5	160.1	149.1	93.2	150.6	14.2	139.6	123.9	128.5	126.2
5	OMe[b]	128.8	130.8	114.3	162.7	159.5	150.8	93.0	149.0	14.2	139.6	123.9	128.4	126.2
6	NMe_2[c]	123.8	130.8	111.4	152.8	160.1	148.9	92.6	151.7	14.2	139.8	123.7	128.3	125.8
7	Cl	137.9	129.1	130.1	134.3	158.4	149.2	93.3	150.0	14.1	139.4	126.1	128.5	126.5
8	Br	134.7	130.2	132.1	126.4	158.5	149.1	93.4	149.9	14.2	139.4	124.0	128.5	126.5
9	NO_2	141.2	129.5	124.0	149.4	156.7	149.3	93.8	149.2	14.1	139.2	124.2	128.6	126.8
10	CO_2Me[d]	139.6	128.8	130.0	132.7	158.5	149.2	93.5	149.8	14.2	139.4	124.1	128.6	126.6
11	CN[e]	139.5	129.1	132.5	114.6	157.3	149.3	93.7	149.3	14.1	139.2	124.1	128.6	126.7
12	*o*-OH[f]	118.9	160.6	117.3	133.9	162.3	149.5	93.3	147.9	14.1	138.7	124.6	129.0	127.6
13	*o*-Br[g]	134.0	126.3	133.2	132.6	158.5	149.1	93.7	149.9	14.1	139.3	124.0	128.4	126.4

[a] 21.6 ppm (CH_3). [b] 55.3 (OCH_3). [c] 39.9 [$N(CH_3)_2$]. [d] 52.3 (CH_3), 166.3 (CO). [e] 118.3 (CN). [f] 119.4 (C-5) and 132.7 ppm (C-6). [g] 127.6 (C-5) and 128.9 ppm (C-6).

Table 3 ^1H NMR chemical shifts (δ from internal $SiMe_4$) of compounds **1**, **2**, **4**, **5**, **6** and **9** in TFA solution at 30 °C

		Ring *A*				Ring *B*		Ring *C*		
No	R	H-2/6	H-3/5	Others	=CH	H-4	CH_3	H-2/6	H-3/5	H-4
1a		—	—	—	—	5.91	2.45	7.70	7.52	7.67
2b	H	8.03	7.63	7.80	9.92	5.91	2.46	7.7[a]	7.5[a]	7.7[a]
4b	Me	7.92	7.46	—	9.81	5.91	2.50	7.7[a]	7.5[a]	7.7[a]
5b	OMe	8.06	7.19	4.04	9.74	5.91	2.46	7.7[a]	7.5[a]	7.7[a]
6b	NMe_2	8.26	7.93	3.54	10.09	5.91	2.45	7.7[a]	7.5[a]	7.63
9	NO_2	7.82	8.43	—	Not obs	7.32	2.46	7.7[a]	7.5[a]	7.7[a]
9a	NO_2	8.17	8.37	—	9.17	6.86	2.72	7.7[a]	7.5[a]	7.7[a]
9b	NO_2	8.26	8.48	—	10.17	5.93	2.47	7.7[a]	7.5[a]	7.67

[a] Strongly second-order pattern. Bold face letters **a** and **b** refer to singly and doubly protonated forms, respectively. The values for forms **2a**, **4a**, **5a** and **6a** are not reported due to strongly overlapping signals with doubly protonated forms.

Table 4 ^{13}C NMR chemical shifts (δ from internal $SiMe_4$) of selected Schiff bases in TFA solution at 30 °C

		Ring *A*					Ring *B*				Ring *C*			
No	R	C-1	C-2	C-3	C-4	=CH	C-3	C-4	C-5	CH_3	C-1	C-2	C-3	C-4
2b	H	139.0	133.1	131.3	136.9	200.8	*a*	94.6	153.5	11.7	133.5	127.6	133.0	134.2
4b	Me	*a*	133.4	132.0	134.3	200.1[b]	*a*	94.7	153.6	11.8	133.5	127.5	132.8	134.0
6b	NMe_2	139.1	135.0	123.1	150.7	197.2[c]	*a*	94.7	153.6	11.8	133.2	127.3	132.7	133.8
9b	NO_2	133.8	133.6	126.4	141.7	198.1	*a*	94.5	153.3	11.7	133.5	127.6	132.9	134.1

δ(4-Me) in **4** = 22.5 ppm and δ(4-NMe_2) in **6** = 49.1 ppm. Bold face letter **b** refers to the doubly protonated form of each Schiff base. [a] Not visible in experimental conditions. [b] ^1J(C-α,H-α) = 175.0. [c] ^1J(C-α,H-α) = 181.5.

Among the ^1H NMR chemical shifts of **1** only H-4 is markedly influenced by the condensation with aromatic aldehydes. Similarly, the ^1H NMR chemical shifts of SBs **2–13** stay very constant except for those in substituted ring *A*. Small substituent effects are also transmitted to the azomethine proton, the *p*-NMe_2 and *p*-NO_2 derivatives showing the most different values in agreement with their different electronic character. In contrast, the ^1H NMR chemical shifts are drastically changed by protonation in TFA (Table 3).

The ^{13}C NMR chemical shifts of substituents-bearing ring *A* show typical variation depending on the character of the substituent (Table 4). There is also some variation in the chemical shift of the azomethine or imidoyl carbon (C-α to ring *A*), which can be related with the Hammett substituent constant σ_p[29] as in the case of *N*-benzylideneanilines.[7] In **2–11** a clear correlation was observed, eqn. (1), with multiple $R = 0.88$ and

$$\delta(^{13}C\text{-}\alpha) = (-2.2 \pm 0.4)\sigma_p + 159.0 \pm 0.2 \qquad (1)$$

number of cases $n = 10$. This correlation is practically the same as in *N*-benzylideneanilines (slope −2.3, $R = 0.89$ and $n = 9$),[7] for which this unexpected finding is explained by an inversion of the ordinary conjugative interaction of the azomethine moiety.[7]

The ^1J(C-α,H-α)/Hz of azomethine carbon varying between 156.4 [in *p*-$N(CH_3)_2$] and 162.7 (in *p*-NO_2) also shows a clear correlation with σ_p, eqn. (2), with multiple $R = 0.90$ and number

$$^1J(\text{C-}\alpha,\text{H-}\alpha)/\text{Hz} = (4.0 \pm 0.8)\sigma_p + 160.2 \pm 0.4 \qquad (2)$$

of cases $n = 8$ (*p*-F and *p*-OCH_3 derivatives are omitted owing to the traces of starting materials and poor S/N ratio in coupled ^{13}C NMR spectra). The opposite dependencies of $\delta(^{13}C\text{-}\alpha)$ and ^1J(C-α,H-α) *vs.* σ_p reveal that the electron withdrawing nature of the substituent causes a deshielding effect on the carbon by diminishing the p-electron density and similarly increasing the s-character of the C–H bond and ^1J(C-α,H-α).

^{15}N NMR spectroscopy can provide useful information for the present SBs containing three nitrogens and the sensitivity of nitrogen chemical shifts to solvent, substituents as well as protonation.[1,9,21–24] The $\delta(^{15}N)$ of **2**, **4**, **6**, **9** and **12** are collected in Table 5. The $\delta(^{15}N)$ assignment is based on literature values[1,9,21–24] and proton coupled spectra. The close signals of N-2 in the pyrazole ring and in $N{=}C_\alpha H_\alpha$ are assigned *via*

Table 5 ^{15}N NMR chemical shifts (δ from external CH_3NO_2) of compounds **2**, **4**, **6**, **9** and **12** in 0.5 M $CDCl_3$ solution at 30 °C

No	R	N-1	N-2	N=C	Others
2	H	−172.8	−84.3	−86.0	—
2[a]	H	−209.9	−211.2	−330.2	—
4	Me	−173.4	−87.1	−87.9	—
6	NMe_2	−175.4	−90.7	−103.0	−325.6
9	NO_2	−171.3	−82.4	−73.0	−13.3
12	*o*-OH	−173.7	−84.4	−112.7[b]	—

[a] In TFA, the chemical shift assignment of N-1 and N-2 is ambiguous. [b] Triplet.

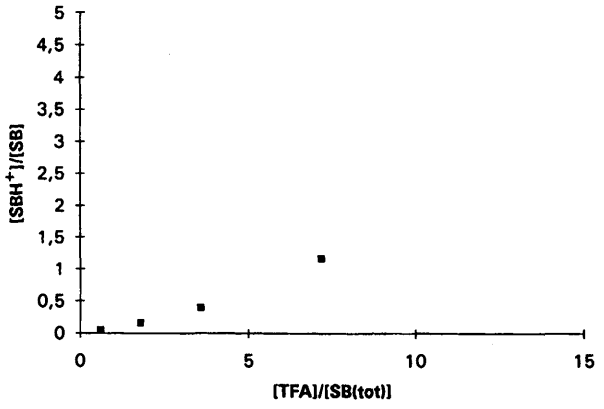


Fig. 1 $[SBH^+]/[SB]$ *vs.* [TFA]/[SB(tot)] for **4** (*p*-CH_3 derivative) based on integration of proton signals of N=CH–moiety

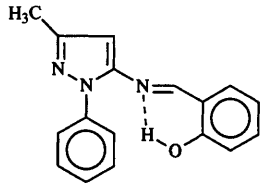


Fig. 2 Intramolecular hydrogen bonding in **12**

$^2J(N,H_\alpha)/Hz = 3.7$ in **2**, **4**, **6** and $^2J(N,\alpha\text{-}H)/Hz = 4.1$ in **9** while N-2 did not show any resolved coupling. These assignments were ascertained by inverse 2-D ^1H, ^{15}N HMBC field gradient (FG) enhanced experiments for **2**, **4**, **6** and **9**, where three correlation peaks were found, *viz.* between (*i*) the azomethine proton and nitrogen and in pyrazole ring between (*ii*) H-4 and N-1 and (*iii*) 3-CH_3 and N-2.

The $\delta(^{15}N)$ of N=CH shows a 30 ppm variation range in **2**–**9**; *p*-$N(CH_3)_2$ and *p*-NO_2 derivatives representing the extreme values −103.0 and −73.0 ppm, respectively. The $\delta(^{15}N)$ of azomethine nitrogen in *o*-OH derivative **12** exhibits a clearly shielded value, −112.7 ppm. This can be explained by intramolecular hydrogen bonding between the *o*-OH and azomethine nitrogen (Fig. 2) as in *o*-OH *N*-benzylideneanilines.[1] This hydrogen bond formation is also detected by ^{17}O NMR spectroscopy of **12** giving $\delta(o\text{-}^{17}OH) = 92.6$ ppm, which is clearly deshielded from that of 3-hydroxybenzaldehyde, $\delta(m\text{-}^{17}OH) = 80$ ppm.[30]

Protonation studies by NMR

The degree of protonation *vs.* [TFA]/[SB(tot)] has been studied for *p*-CH_3 derivative **4** by titrating its 0.5 M $CDCl_3$-solution with TFA (**4** was selected instead of **2** to avoid a second-order ^1H NMR spectral analysis). Fig. 1 shows $[SBH^+]/[SB]$ *vs.* [TFA]/[SB(tot)] for **4** based on integration of the methine proton signals of N=CH. At [TFA]/[SB(tot)] = 15 there still exists *ca.* 20% of the total amount of **4** in its neutral form. This finding is in agreement with a previous observation,[2] where trichloroacetic acid ($pK_a = 0.66$) was unable to fully protonate simple aliphatic SBs. Further, for **9** (200 mg in 700 μl TFA) three forms, *viz.* unprotonated **9**, singly protonated **9a** and doubly protonated **9b** were present in the molar ratios of 0.3/0.4/1.0. The spectral assignments of these forms are obtained by a gradual dilution of the sample and finally measuring it in a solution where only the form **9b** was visible (7 mg of **9** in 700 μl TFA). Thus, the double protonation of these SBs can be achieved in very dilute solutions ([SB] < 0.02 M) in TFA.

The forms **9a** and **9b** both differ by their ^1H chemical shifts from those of the neutral form **9** (Table 3). Therefore, it is not straightforward to determine the preferred protonation site. The differences, $\delta(\mathbf{9}) - \delta(\mathbf{9a}) < \delta(\mathbf{9a}) - \delta(\mathbf{9b})$, for the methine protons of N=CH (8.62 ppm in $CDCl_3$), however, suggest that the most preferred site is N-2 in ring *B* and the second one the azomethine nitrogen, respectively.

^{13}C NMR spectra in TFA were also measured for some SBs although problems arose in the spectral assignment owing to the simultaneous existence of different forms. An extreme dilution of the sample as in ^1H NMR was impractical because accumulation times tend to increase rapidly in ^{13}C NMR. In spite of these difficulties, the ^{13}C NMR chemicals shifts for the doubly protonated forms of four SBs were obtained (Table 4). As in ^1H NMR spectroscopy, clear chemical shift changes were found especially for C-α in CH=N showing clearly deshielded values at 197.2–200.8 ppm.

The protonation sites in 3- and 5-amino-1-methylpyrazoles determined *via* ^{13}C NMR spectroscopy have been reported by de Mendoza *et al.*[20] According to their report the most preferred protonation site is N-2 in the pyrazole ring in accord with the above ^1H data. Further, this protonation strongly deactivates the exocyclic nitrogen atom, so that no double protonation is observed in 3- and 5-aminopyrazoles, even in sulfuric acid. This is in contrast with the present ^{13}C NMR (and ^1H) data since the SBs studied now clearly show double protonation properties. Unfortunately, the $\delta(^{13}C)$ of singly protonated forms remained uncertain owing to the overlap of different forms and consequently a comparison with the literature data[20] was impossible.

^{15}N NMR spectroscopy has been successfully used in protonation studies of pyrazoles.[22,24] The poor sensitivity of ^{15}N NMR at natural abundance and the simultaneous existence of several forms of SBs formed a limiting factor in the present study. Nevertheless, for dilute **2** in TFA all ^{15}N NMR resonances have been observed. The protonation shifts observed for N-1 (−37.1 ppm) and N-2 (−126.9 ppm) in TFA are in agreement with those reported by Claramunt *et al.*[22] A large shift of azomethine nitrogen (−244.2 ppm) verifies that the protonation also occurs on that site. Unfortunately, all attempts to obtain the proton coupled ^{15}N NMR spectrum of **2** in TFA failed.

In order to improve the sensitivity of the ^{15}N NMR, inverse 2-D ^1H, ^{15}N HMQC (hetero multiple quantum coherence) experiments with and without FG enhancement have been done for **2** in TFA. Although the value of transmitting coupling was varied between 70 and 120 Hz, these experiments did not give any additional information, which probably is due to proton exchange between the nitrogens and the solvent.

Next, ^{14}N NMR spectroscopy (99.63% natural abundance) was applied to avoid the sensitivity problems of ^{15}N NMR spectroscopy. Unfortunately, **2** in TFA gave only one signal at −72.5 ppm characterized by a 70 Hz linewidth. The *p*-NO_2-derivative **9** (for which the systematic dilution study was made) was also checked by ^{14}N NMR spectroscopy in $CDCl_3$ giving only two signals at −14.5 ppm (width 360 Hz) and −71.4 ppm (width 70 Hz), respectively.

Ab initio MO calculations

To assess the order and sites of protonation from the above, *ab initio* MO calculations at the 6-31G* level with full geometry optimization have been performed for a model compound **14**, 1,3-dimethyl-5-*N*-benzylideneaminopyrazole, which was selected instead of 1-phenyl derivative **2** to shorten the CPU-demanding calculations needed for **2** and its possible con-

Table 6 Energies E(HF/6-31G*)/au and ΔE/kJ mol^{-1} between the non-protonated form **I** and the protonated forms **II–V** of **14**

Form	Protonation site	E/au	ΔE/kJ mol^{-1}
I	Non-protonated	−625.308 840 281	0.0
II	N of azomethine	−625.674 602 823	−960.2 ($E_2 - E_1$)
III	N-2 of pyrazole	−625.701 121 812	−1030.1 ($E_3 - E_1$)
IV	C-4 of pyrazole	−625.691 648 135	−1004.9 ($E_4 - E_1$)
V	N-1 of pyrazole	−625.696 017 567	−1016.7 ($E_5 - E_1$)

Table 7 Optimized (HF/6-31G*) bond lengths/Å and bond angles/° for non-protonated form I and the protonated forms **II–V** of **14**[a]

Form	**I**	**II**	**III**	**IV**	**V**
Bond length/Å					
N(1)–N(2)	1.334	1.333	1.333	1.396	1.433
N(2)–C(3)	1.305	1.302	1.302	1.262	1.275
C(3)–C(4)	1.408	1.415	1.415	1.506	1.461
C(4)–C(5)	1.375	1.362	1.362	1.507	1.331
C(5)–N	1.389	1.413	1.413	1.343	1.359
N=C	1.258	1.290	1.290	1.288	1.279
Bond angle/°					
N(1)–N(2)–C(3)	106.14	107.21	107.21	107.70	105.82
N(2)–C(3)–C(4)	110.96	110.59	107.44	111.77	113.36
C(3)–C(4)–C(5)	104.31	103.42	103.42	99.88	106.78
C(4)–C(5)–N	134.42	130.20	130.20	131.77	140.25
C(5)–N=C	120.03	125.37	125.37	119.10	119.93

[a] The full set of (HF/6-31G*) structural parameters are available from R. S. on request.

formers. The HF/6-31G* energy values in au as well as the energy differences ΔE in kJ mol^{-1} are collected in Table 6. The optimized HF/6-31G* geometries of the unprotonated and all three protonated forms of **14** (except the protons of the methyl groups and the forms where an additional proton is located at C-4 or N-1) are almost planar. The bond lengths and angles for the pyrazole ring and the azomethine moiety in forms **I–V** of **14** are collected in Table 7, respectively.

Based on these data the preferred protonation site of SBs studied is N-2 in the pyrazole ring. The other sites of the pyrazole ring are also preferred over the azomethine group in singly protonated structures. However, it is reasonable to assume that after protonation at N-2 the next site will be the nitrogen of the azomethine moiety rather than other sites in the pyrazole ring. Consequently, these theoretical calculations support the conclusions drawn based on the NMR spectral data.

Conclusions

Schiff bases (SB) derived by condensation of 1-phenyl-3-methyl-5-aminopyrazole and aromatic aldehydes show double protonation characteristics in trifluoroacetic acid (TFA). According to the present NMR and *ab initio* (HF/6-31G*) studies, the first protonation site is the unsubstituted N-2 in the pyrazole ring and the second one the nitrogen of the azomethine moiety. Over a large [TFA]/[SB(tot)] range the neutral as well as singly and doubly protonated forms were simultaneously observed. In fact, the doubly protonated forms of these SBs were observed in dilute solutions ([SB] < 0.02 M) in neat TFA. Protonation studies by ^{15}N NMR spectroscopy at natural abundance proved to be problematic owing to low sensitivity and simultaneous existence of several forms of the compounds in the relatively concentrated solutions needed for ^{15}N NMR spectroscopy. The multiple protonation characteristics of the SBs studied in this work may also be of importance in predicting the binding of metal cations, as in for example dioxo-vanadium(v) Schiff base complexes.[31]

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