Reactions of indolic nitrones and *N*-heteroaromatic bases under irradiation and chemical oxidation

Angelo Alberti,[a] Paola Astolfi,[b] Dietrich Döpp,[c] Lucedio Greci,*[b] Dante Macciantelli[a] and Rita Petrucci[d]

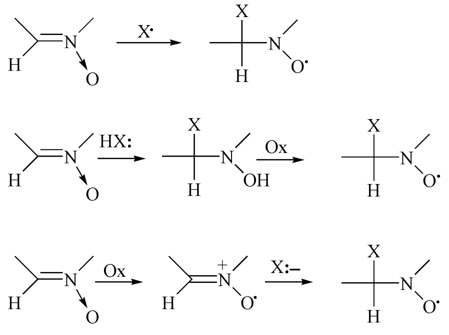
[a] *ISOF-CNR, Area della Ricerca, Via P. Gobetti 101, I-40129, Bologna, Italy*
[b] *Dipartimento di Scienze dei Materiali e della Terra, Università di Ancona, Via Brecce Bianche, I-60131, Ancona, Italy. E-mail: greci@unian.it; Fax: +39 071 220 4714; Tel: +39 071 220 4408*
[c] *Fachgebiet Oirganische Chemie, Universität Duisburg, D-47048, Duisburg, Germany*
[d] *Dipartimento di Ingegneria Chimica, Università di Roma "La Sapienza", Via del Castro Laurenziano 7, I-00161, Roma, Italy*

Received (in Montpellier, France) 19th December 2002, Accepted 21st February 2003
First published as an Advance Article on the web 2nd June 2003

The spin adducts formed under UVA irradiation or in the presence of mild oxidants in the reaction of a number of heteroaromatic bases and two indolic nitrones have been characterized by means of ESR spectroscopy. The formation of the spin adducts is explained *via* the Forrester–Hepburn mechanism, while the occurrence of inverted spin trapping is excluded. Photolysis of nitrones **1** and **2** or of the corresponding cyclic hydroxamic acids resulted in the formation of the related acyl nitroxides.

Introduction

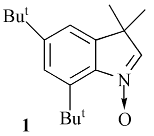
First introduced in the early 70's, spin trapping has become through the years the most widely used technique for the study of transient radicals.[1–3] The most popular spin traps are nitroso compounds, either aliphatic or aromatic, and nitrones. Typical examples of the former group are 2-methyl-2-nitrosopropane (MNP), nitrosobenzene (NB) and the sodium salt of 3,5-dibromo-4-nitrosobenzenesulfonic acid (DBNBS), while the most popular members of the latter group are *N*-*tert*-butyl-α-phenylnitrone (PBN) and 5,5-dimethyl-1-pyrroline *N*-oxide (DMPO). These two last compounds have been widely used in the study of radical processes in biological systems.[4,5] In recent years, 5-diethoxyphosphoryl-5-methyl-1-pyrroline *N*-oxide (DEPMPO)[6] has successfully joined the family of nitrone-based spin traps, its main advantage over DMPO consisting in the greater persistence of its spin adducts with oxygen-centred radicals, and in particular the superoxide radical anion.[7]



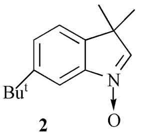
Scheme 1

Although spin trapping is now a well-established technique, there are instances in which the mechanism of formation of the spin adducts to nitrones remains the object of controversy. Indeed, while in most cases the spin adducts are formed by homolytic addition of an $X^•$ radical to the nitrone (Scheme 1, first reaction), under oxidative conditions they may also originate through two different alternative routes. Thus, if the nucleophile :XH is present in the reaction medium, its reaction with the nitrone would give a hydroxylamine, which under the experimental conditions (*i.e.*, irradiation, presence of oxidants) is subsequently oxidised to the spin adduct (Scheme 1, second reaction). This process is known as the Forrester and Hepburn mechanism.[8] On the other hand, the oxidation of the nitrone to its radical cation might initially take place, followed by nucleophilic addition by the nucleophile $:X^-$ present in solution (Scheme 1, third reaction). This process, which also results in the formation of a spin adduct, goes under the name of "inverted spin trapping" and was first hypothesised by Eberson and coworkers[9] in order to explain the formation of spin adducts when the common traps were irradiated in the presence of nucleophiles.

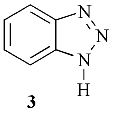
Here, a study is reported aimed at gaining more information on the mechanism of the spin adduct formed in the reaction between the cyclic nitrones **1** and **2** and a number of *N*-heteroaromatic bases (**3**–**9**) with high oxidation potentials, under either UVA irradiation or mild chemical oxidising conditions. From the redox potentials of the reagents and from the results obtained, it may be excluded that the reactions follow the "inverted spin trapping" mechanism, but, more likely, the formation of the spin adducts could be explained by the Hepburn–Forrester mechanism.



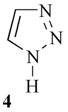
1



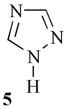
2



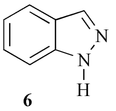
3



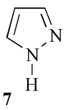
4



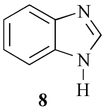
5



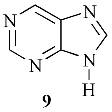
6



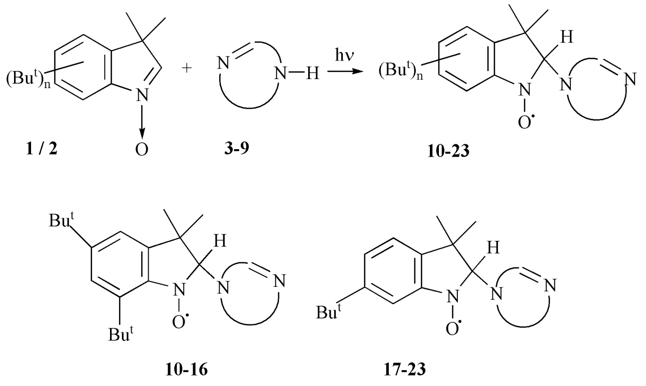
7



8



9



Scheme 2

Results

UV photolysis of dichloromethane solutions of either **1** or **2** and one of the heteroaromatic bases **3–9** inside the cavity of the ESR spectrometer led to the observation of rather intense and well-resolved signals of the spin adducts **10–23** (Scheme 2). The hyperfine spectral parameters of the adducts, which could also be observed in the absence of UV irradiation upon addition of a small amount of chloranil to the solutions, are collected in Table 1.

The ESR spectra of the adducts from the two spin traps exhibited a similar spectral pattern within each series and two typical examples are shown in Fig. 1. In the adducts of **1**, coupling is observed between the unpaired electron and the nitroxidic nitrogen (N_1), the C-2 hydrogen, two ring hydrogen atoms ($H_{4,6}$), and a nitrogen atom from the $X^•$ group, while in those of **2** the unpaired electron is coupled with the nitroxidic nitrogen (N_1), the C-2 hydrogen, three ring hydrogen atoms (H_4, H_5 and H_7), and a nitrogen atom from the $X^•$ radical.

In a few cases prolonged irradiation of the samples resulted in a modification of the spectral pattern, and eventually new spectra were observed that were characterized by a much smaller overall splitting (see Fig. 2). These spectra were attributed to the acyl nitroxide radicals **24** [$a(2H_{4,6}) = 0.085$ mT, $a(N_1) = 0.599$ mT, $g = 2.0059_9$] and **25** [$a(H_4) = 0.081$ mT, $a(H_5) = 0.238$ mT, $a(H_7) = 0.284$ mT, $a(N_1) = 0.592$ mT, $g = 2.0060_5$], and could also be observed by UVA irradiation of the solutions of the spin traps or of the corresponding hydroxylamines **26** and **27** in the absence of a base or by treating the last two compounds with PbO_2.

The knowledge of the oxidation potentials of the two spin traps and of the different nucleophiles is critical for the rationalisation of the mechanism leading to the formation of the spin adducts. We have therefore measured the oxidation potentials for those compounds for which they were not available. All the values are collected in Table 2.

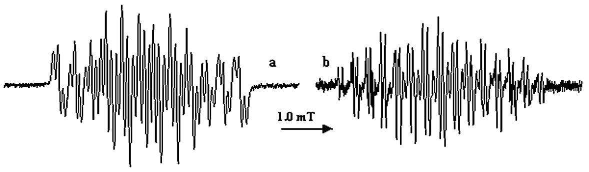


Fig. 1 ESR spectra observed at room temperature upon photolysis of a CH_2Cl_2 solution of benzotriazole containing a small amount of **1** (a) or **2** (b).

Discussion

The formation of spin adducts **10** to **23** is not to be viewed as a normal spin trapping process, that is it cannot be accounted for by a direct homolytic addition of the heteroaromatic base derived radicals to the nitrones. All the *N*-heteroaromatic bases that have been the object of the present study have oxidation potentials higher than 1.7 V *vs.* NHE, and it can therefore be excluded that the action of chloranil can convert them to radicals. Indeed, with an $E_{red} = 0.25$ V *vs.* NHE, chloranil is a very weak oxidant and its action in organic solvents is similar to that of PbO_2 in $CH_3CN–H_2O$. For example, the latter species is unable to oxidise indolic nitroxides, for example compounds with an $E_{ox} = 1.0$ V *vs.* NHE, to the corresponding quinoneimine *N*-oxides.[12]

An alternative explanation for the detection of these spin adducts might be that they are formed through an inverted spin trapping process. Such a possibility can be immediately ruled out in the case of chloranil on the basis of Marcus theory, according to which an electron transfer process is feasible when the acceptor (the oxidant) has a reduction potential more positive than that of the donor (the reductant).[13] The reduction potential of chloranil and those of **1** and **2** are clearly incompatible. Compounds **1** and **2** have oxidation potentials of about 1.8 V *vs.* NHE and by analogy with PBN[14,15] their excited states are expected to have an E_{red} that is even more positive. Thus, under UVA irradiation conditions one might expect the excited state of **1** or **2** to act as an oxidant towards the ground state molecules. However, upon irradiation of solutions of nitrones **1** or **2** and a compound with an oxidation potential lower than those of the two nitrones, and amenable of being easily oxidised to a very stable radical cation, no ESR signal from such a radical cation could be detected.

Table 1 Hyperfine spectral parameters of spin adducts **10–23**[a]

Radical	Spin trap	X	$a(N_1)$	$a(H_2)$	$a(H_{4,6})$	$a(H_5)^{10}$	$a(H_7)^{10}$	a(other)	g
10	**1**	Benzotriazol-1-yl	0.993	1.117	0.095[b]			0.334(1N)	2.0054_3
11	**1**	1,2,3-Triazol-1-yl	1.020	0.900	0.095[b]			0.425(N)	2.0053_5
12	**1**	1,2,4-Triazol-1-yl	1.023	0.964	0.102[b]			0.367(N)	2.0054_0
13	**1**	Indazol-1-yl	0.993	0.919	0.097[b]			0.360(N)	2.0054_8
14	**1**	Pyrazol-1-yl	0.993	0.932	0.082[b]			0.365(N)	2.0055_2
15	**1**	Benzimidazol-1-yl	1.030	0.990	0.102[b]			0.312(N)	2.0053_9
16	**1**	Purin-1-yl	1.025	1.125	0.075[b]			0.300(N)	2.0054_3
17	**2**	Benzotriazol-1-yl	0.997	1.323	0.099[c]	0.344	0.305	0.341(N)	2.0054_0
18	**2**	1,2,3-Triazol-1-yl	1.021	1.130	0.103[c]	0.344	0.314	0.420(N)	2.0053_5
19	**2**	1,2,4-Triazol-1-yl	0.941	1.042	0.093[c]	0.336	0.281	0.344(N)	2.0054_9
20	**2**	Indazol-1-yl	1.025	1.112	0.098[c]	0.364	0.349	0.331(N)	2.0051_0
21	**2**	Pyrazol-1-yl	1.028	1.202	0.101[c]	0.345	0.309	0.353(N)	2.0054_4
22	**2**	Benzimidazol-1-yl	1.004	1.305	0.101[c]	0.349	0.304	0.322(N)	2.0053_8
23	**2**	Purin-1-yl	0.975	1.390	0.120[c]	0.345	0.320	0.360(N)	

[a] Coupling constants in mT. [b] 2H. [c] $1H(H_4)$.

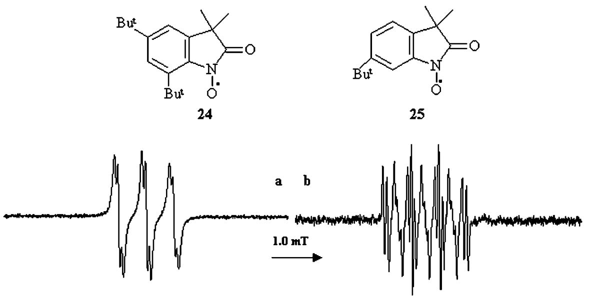
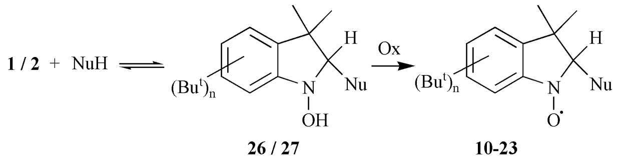


Fig. 2 ESR spectra of acyl nitroxides **24** (a) and **25** (b) as observed at room temperature upon prolonged photolysis of a CH_2Cl_2 solution of either **1** or **2**.



Scheme 3

The compound used in the present case was 2,2-diphenyl-5-*tert*-butoxyindolin-3-one, which has an oxidation potential $E_{1/2\text{ox}} = 0.78$ V in CH_3CN *vs.* Ag/Ag^+.[16] As a consequence, the formation of spin adducts **10–23** upon simple UVA irradiation of solutions of nitrones **1** or **2** cannot be explained with the occurrence of an inverted spin trapping process.

We are therefore left with the Hepburn–Forrester mechanism, that is the nucleophilic addition of the base to the nitrones, followed by oxidation of the resulting hydroxylamines **26** and **27**, as the most likely route to nitroxides **10–23** under the present experimental conditions (see Scheme 3).

It remains unclear, however, what is the nature of the oxidant that promotes the last step of the reaction under UVA irradiations, that is in the absence of either chloranil or PbO_2. Although the excited state of the nitrone(s) may be seen as possible candidates, we believe it more likely that they are trapped by the ground state nitrone(s) to give the acyl nitroxides **24** or **25** (see Scheme 4). The fact that the signal of the acyl nitroxide(s) is immediately observed upon UVA irradiation of the nitrone(s) alone gives support to the proposed mechanism.

Therefore, the acyl nitroxides **24/25** must be the actual oxidants that convert hydroxylamines **26/27** to nitroxides **10–23**. In order to verify that acyl nitroxides can oxidise the intermediate hydroxylamines, **24** was prepared by oxidising the corresponding hydroxamic acid. When the acyl nitroxide was added to a solution of nitrone **1** and benzotriazole **3**, an intense signal of the spin adduct **10** could be immediately detected.

Last but not least, it seems worth stressing that irradiation of the hydroxamic acids **32/33** alone inside the EPR cavity also leads to detection of the spectra of the corresponding acyl nitroxides **24/25** (Scheme 5), thus implying that the hydroxamic acids can be oxidised to the corresponding nitrone. Disproportionation of **34/35** followed by the loss of water eventually leads to the corresponding indolones.

Similarly, the formation of the 3-*H* indoles (deoxygenated nitrones) **30/31** and of the hydroxamic acids **32/33** (see Scheme 4) can be explained *via* the disproportionation of **28/29**.

Conclusions

Based on the redox potentials of the reagents and on the present results, it can be excluded that the process leading to **10–23** involves "inverted spin trapping". The reaction of indolic nitrones **1** and **2** with the *N*-heteroaromatic bases **3–9** in the presence of weak oxidants represents a typical case of the Hepburn–Forrester mechanism, consisting in the nucleophilic addition of the base to the trap followed by oxidation of the resulting hydroxylamino derivative. The initial addition is presumably an equilibrium that, even if shifted to the left (the macroscale reaction did not lead to any isolable product), affords the intermediate hydroxylamine to an extent sufficient to be oxidised to the corresponding spin adduct.

The Forrester–Hepburn mechanism is also believed to be operative for the reactions carried out under irradiation, the acyl nitroxides being the most likely oxidants of the intermediate hydroxylamines.

Experimental

Materials

N-Heteroaromatic bases **3–9** were purchased from Aldrich and were used as received. Indolic nitrones **1**[17] and **2**[18] were prepared according to literature procedures. For the preparation of acyl nitroxides **24** and **25** the appropriate hydroxamic acid (20 mg) was dissolved in 10 mL of dichloromethane and oxidised with PbO_2 (50 mg) at room temperature under stirring for 15 min. The mixture was filtered and washed with dichloromethane; the conversion was considered complete.

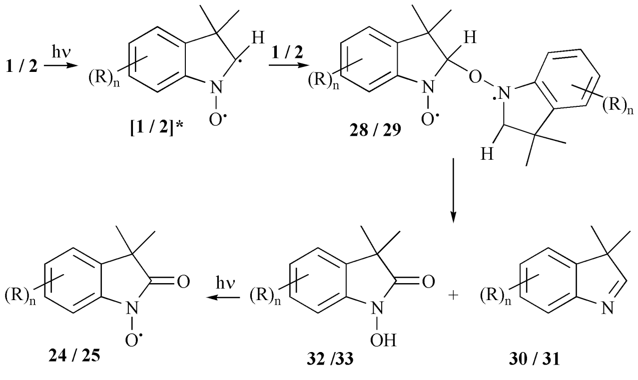
ESR measurements

ESR spectra were recorded on a Varian E-4 or on an upgraded Bruker ER 200 D spectrometer. In the case of chemical oxidation, an inverted U cell[19] was used: 5 mg of the nitrone and 10 mg of the base were dissolved in 2 mL of dichloromethane in one of the two legs of the cell, whereas in the other leg 10 mg of

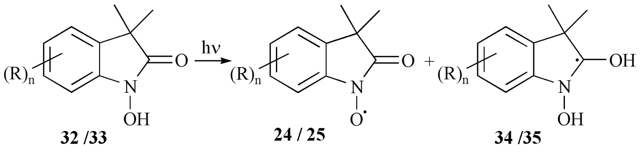
Table 2 Half-wave oxidation potentials *vs.* Ag/Ag^+ for compounds **1**–**9** in acetonitrile/TEAP[a]

Compound	$E_{1/2}$(ox)/V	Reference
1	1.91	This work
2	1.75	This work
3	1.70	Ref. 11
4	1.80	Ref. 11
5	2.36	This work
6	1.15	Ref. 11
7	1.70	Ref. 11
8	1.10	Ref. 11
9	2.10	Ref. 11

[a] NHE = $Ag/Ag^+ + 0.54$ V



Scheme 4



Scheme 5

PbO$_2$ or 2 mg of chloroanil, respectively, were added to 2 mL of the same solvent. The solutions were degassed with argon, mixed and transferred into the aqueous cell that was placed into the ESR cavity. For the irradiation experiments 5 mg of the nitrone and 10 mg of the base were dissolved in 2 mL of dichloromethane and an aliquot of 400 μL were transferred to a quartz tube. The solution was carefully deoxygenated with argon and the tube was irradiated directly inside the ESR cavity with UVA light from a Philips Original Home Solarium (model HB 406/A) equipped with a 400 W ozone-free Philips HPA lamp, UV type 3.

The solutions used for the measurements carried out with the Bruker spectrometer were prepared, using the same amounts of each reactant, as described above; 0.4 mL of these solutions were carefully degassed with argon and the tubes were put into the ESR cavity and irradiated with a UVA lamp: the ESR spectra of the spin adducts were immediately recorded. The same spin adduct spectra were also obtained when solutions of the nitrone–base mixture were oxidised with acyl nitroxide **24**.

Electrochemical measurements

Electrochemical measurements were carried out at room temperature under nitrogen in a three-electrode cell,[20] using 10^{-3} mol·dm^{-3} acetonitrile solutions of **1**, **2** or **5** containing tetraethylammonium perchlorate (TEAP; 0.1 mol·dm^{-3}). The working electrode was a stationary platinum disk (Amel 492), the reference electrode was Ag/0.1 mol·dm^{-3} AgClO$_4$ and the auxiliary electrode was a platinum wire. The experiments were performed using an Amel 472/WR multipolarograph coupled with an Amel 863 digital x/y recorder.

References

1 E. G. Janzen, *Acc. Chem. Res.*, 1971, **4**, 31.
2 V. E. Zubarev, V. N. Belevskii and L. T. Bugaenko, *Russ. Chem. Rev.*, 1979, **48**, 729.
3 M. J. Perkins, *Adv. Phys. Org. Chem*, 1980, **17**, 1.
4 J. A. Degray and R. P. Mason, in *Electron Spin Resonance*, ed. N. M. Atherton, M. J. Davies and B. C. Gilbert, Atheaeun Press Ltd., Cambridge, 1994, vol. 14, p. 246.
5 G. R. Buettner, *Free Radical Biol.*, 1987, **2**, 259.
6 (*a*) C. Frejaville, H. Karoui, F. Le Moigne, M. Culcasi, S. Pietri, R. Lauricella, B. Tuccio and P. Tordo, *J. Chem. Soc., Chem. Commun.*, 1994, 1973; (*b*) C. Frejaville, H. Karoui, F. Le Moigne, M. Culcasi, S. Pietri, R. Lauricella, B. Tuccio and P. Tordo, *J. Med. Chem.*, 1995, **38**, 258.
7 B. Tuccio, B. Lauricella, C. Frejaville, J. C. Bouteiller and P. Tordo, *J. Chem. Soc., Perkin Trans. 2*, 1995, 295.
8 A. R. Forrester and S. P. Hepburn, *J. Chem. Soc. (C)*, 1971, 701.
9 (*a*) L. Eberson, *J. Chem. Soc., Perkin Trans. 2*, 1992, 1807; (*b*) L. Eberson and M. Nilsson, *Acta Chem. Scand.*, 1993, **47**, 1129; (*c*) L. Eberson, *J. Chem. Soc., Perkin Trans. 2*, 1994, 171; (*d*) L. Eberson, M. P. Hartshorn and O. Persson, *J. Chem. Soc., Perkin Trans. 2*, 1996, 141.
10 (*a*) C. Berti, M. Colonna, L. Greci and L. Marchetti, *Tetrahedron*, 1977, **33**, 3149; (*b*) C. Berti and L. Greci, *J. Org. Chem.*, 1981, **46**, 3060.
11 A. Alberti, P. Carloni, L. Eberson, L. Greci and P. Stipa, *J. Chem. Soc., Perkin Trans. 2*, 1997, 887.
12 A. Alberti, R. Andruzzi, L. Greci, P. Stipa, G. Marrosu, A. Trazza and M. Poloni, *Tetrahedron*, 1988, **44**, 1503.
13 (*a*) L. Eberson, *Adv. Phys. Org. Chem.*, 1982, **18**, 79; (*b*) M. Juillard and M. Chanon, *Chem. Rev.*, 1983, **83**, 425; (*c*) L. Eberson, *Electron Transfer in Organic Chemistry*, Springer, Heidelberg, 1987; (*d*) J. K. Kochi, *Acta Chem. Scand.*, 1990, **44**, 409.
14 D. Rehm and A. Weller, *Ber. Bunsen-Ges. Phys. Chem.*, 1969, **73**, 834.
15 L. Eberson, J. Lind and G. Merenyi, *J. Chem. Soc., Perkin Trans. 2*, 1994, 1181.
16 C. Berti, L. Greci, R. Andruzzi and A. Trazza, *J. Org. Chem.*, 1985, **50**, 368.
17 D. Döpp and K-H. Sailer, *Chem. Ber.*, 1975, **108**, 301.
18 D. Döpp and E. Brugger, *Chem. Ber.*, 1973, **106**, 2166.
19 G. A. Russel, E. G. Janzen and E. T. Strom, *J. Am. Chem. Soc.*, 1964, **86**, 1807.
20 (*a*) R. Andruzzi, A. Trazza, L. Greci and L. Marchetti, *Ann. Chim. (Rome)*, 1979, **69**, 583; (*b*) R. Andruzzi and A. Trazza, *J. Electroanal. Chem.*, 1977, **77**, 101; (*c*) R. Andruzzi and A. Trazza, *J. Electroanal. Chem.*, 1978, **90**, 389.