

Spin Trapping by 5,5-Dimethylpyrroline-*N*-oxide in Fenton Media in the Presence of Nafion Perfluorinated Membranes: Limitations and Potential

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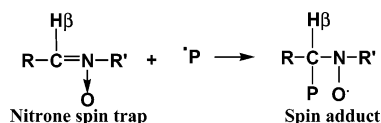
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Spin trapping by 5,5-dimethylpyrroline-*N*-oxide (DMPO) was used for the detection of radicals in Fenton media in the presence and absence of Nafion perfluorinated ionomers. For ethanol as solvent, the same types of spin adducts were detected in the presence or absence of Nafion. Solvent-derived adducts, DMPO/ $\cdot\text{OC}_2\text{H}_5$ and DMPO/ $\cdot\text{CH}(\text{OH})\text{CH}_3$, were identified, and their presence was rationalized by Fe(III)-catalyzed nucleophilic addition of ethanol to the spin trap and hydrogen abstraction by $\cdot\text{OH}$ radicals; oxygen radical adducts, DMPO/ $\cdot\text{O}_2^-$ and DMPO/ $\cdot\text{OOH}$, were also detected. In Fenton media with methanol as solvent (and no Nafion), the DMPO/ $\cdot\text{O}_2^-$ adduct dominated immediately after sample preparation, and a mixture consisting of DMPO/ $\cdot\text{OCH}_3$, DMPO/ $\cdot\text{CH}_3$, DMPO/ $\cdot\text{O}_2^-$, and DMPO/ $\cdot\text{OOH}$ adducts was detected after 30 min. In the presence of Nafion, only the adduct DMPO/ $\cdot\text{OH}$ was detected. For water as solvent, only the DMPO/ $\cdot\text{OH}$ adduct was detected, in both the absence and the presence of Nafion. The full hyperfine tensor components of this adduct were determined in Fenton media in the presence of Nafion with water and methanol as solvents. In Nafion/water exposed to the Fenton reagent at 358 K for 3 h, a DMPO adduct of a carbon-centered radical was also identified and assigned to a Nafion-derived fragment; its exact nature is under investigation. Variations of the ^{14}N and H_β hyperfine splittings of a given adduct with the local polarity were key to the identification of some DMPO adducts, in particular DMPO/ $\cdot\text{O}_2^-$. Both $\cdot\text{OOH}$ and $\text{O}_2^{\cdot-}$ adducts, with different ^{14}N and H_β splittings, were detected simultaneously in some samples, for the first time in the spin trapping literature. Comparison with the results of a direct electron spin resonance study of Nafion exposed to the Fenton reagent indicated that spin trapping by DMPO can provide complementary information on the type of radicals present during Nafion degradation. The spin trapping approach described in this paper is limited, however, to systems that do not contain organic solvents.

Introduction

Since its discovery in the late 1960s,^{1,2} the method of spin trapping has been extensively used for the detection and identification of short-lived free radicals in chemistry, biology, and medicine.^{1–10} The method is based on the scavenging of radicals, $\cdot\text{P}$, by a spin trap, leading to the formation of a spin adduct with higher stability, typically a nitroxide radical. Nitroso and nitron compounds are the most commonly used spin traps. The process of trapping and adduct formation is shown below for a nitron spin trap.



In most cases the ESR spectra of spin adducts exhibit hyperfine splittings (hfs's) from the ^{14}N and H_β nuclei. The goal in spin trapping experiments is to translate these splittings into an identity of the radical $\cdot\text{P}$. Normally it is easy to establish the presence of the radical and more of a challenge to identify it.⁵ The stability of the spin trap under conditions necessary for the formation of radicals is an important consideration. Careful sample preparation, the collection of hfs's from a given system in a series of media and as a function of pH and temperature,

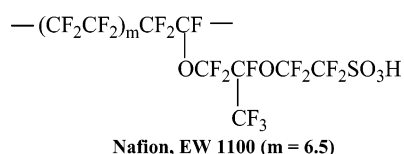
and isotopic enrichment by ^2H , ^{13}C , ^{15}N , and ^{17}O have been used for positive identification. The spin trapping database is a useful resource.¹¹

α -Phenyl-*tert*-butylnitron (PBN) is a useful spin trap whose advantage is its stability under UV irradiation and its selectivity toward trapping of carbon-centered radical species; this advantage is, however, offset by the lack of specificity in the hfs of the spin adduct.^{4,5,12–16} Spin trapping by 5,5-dimethylpyrroline-*N*-oxide (DMPO, below) has emerged as an important method for the identification of unstable intermediates, because the splittings from the H_β protons are sensitive to the nature of the trapped radical; moreover, in some cases hfs's from H_γ protons can also be resolved. The use of DMPO as a spin trap has been described in many studies,^{4–10,17–26} for the detection of oxygen radicals such as $\cdot\text{OH}$, $\cdot\text{OOH}$, and $\text{O}_2^{\cdot-}$ as well as carbon-centered radicals. The clear distinction between these two groups of free radical adducts is based on the large difference in the hfs's of the H_β protons: typically 10–15 G and >20 G for oxygen and carbon-centered radical adducts, respectively. Spin trapping by DMPO is not always without problems, however. Numerous studies have discussed the presence of two types of radicals directly derived from the spin trap: the product of DMPO oxidation, 5,5-dimethyl-2-oxopyrroline-1-oxyl (DMPOX, see below), with hfs's $a_{\text{N}} = 7.1$ G and $a_{\text{H}_\gamma} = 4.2$ G (two protons);^{5,24} and the radical formed by cleavage of the N–C bond and ring opening, with $a_{\text{N}} \approx 15$ G.¹⁹ In the presence of more than one radical, the selectivity of the spin trap is also an important issue. In a recent paper the kinetics of trapping ethanol-derived radicals and $\cdot\text{OH}$ radicals has been compared.²³

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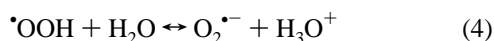
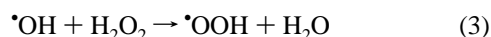
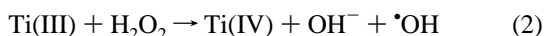
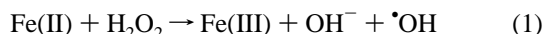
In this paper we describe the use of DMPO for the study of radicals present in proton exchange membranes (PEMs) used in fuel cells exposed to oxygen radicals produced in the Fenton reaction based on Fe(II) or Ti(III).²⁷ Most of our experiments have focused on Nafion perfluorinated membranes as a reference. The equivalent weight (EW) is the weight of polymer containing 1 mol of sulfonic groups.



The goal of the spin trapping experiments reported here is to assess the potential of DMPO as a spin trap in Fenton media in the absence and presence of Nafion ionomer. In a previous study we have used direct electron spin resonance (ESR) for the detection of $\cdot\text{OOH}$, $\text{TiOO}\cdot$, and $\text{O}_2^{\cdot-}$ radicals in Nafion treated by the Fenton reagent based on Ti(III).²⁸ Identification of the radicals listed above was possible by variation of sample preparation methods, temperature used for spectra acquisition, and annealing conditions. Electron spin resonance spectra were measured in the temperature range 77–360 K. The hydroxyl radical, $\cdot\text{OH}$, was not detected. In addition, a broad signal (peak-to-peak width ≈ 84 G) appeared after Fenton treatment of Nafion for three months at ambient temperature and was assigned to a fluorinated radical fragment from Nafion; the identity of the radical was not established, however. Several Nafion-derived fluorinated fragments were detected recently by UV irradiation of Nafion neutralized by Fe(II), Fe(III), and Cu(II) as counterions.²⁹

Fuel cells operate at ≈ 360 K. The use of DMPO as a spin trap is an attempt to determine the identity of radicals in various media at and above ambient temperatures, when direct ESR detection is difficult, if not impossible.

In Fenton reagents based on Fe(II) and Ti(III) radicals, $\cdot\text{OH}$ are produced and can react further to produce the hydroperoxy radical $\cdot\text{OOH}$ and the superoxide radical $\text{O}_2^{\cdot-}$:



The literature suggests that reactions in the presence of Ti(III) are faster compared to the case of Fe(II).²⁷

The nature of the spin adducts in Fenton media has been discussed in several papers,^{5,7,21,22,30} and the consensus seems to be that the $\cdot\text{OH}$ radical is the primary species formed. Additional spin adducts were also identified, and their nature and relative intensity appear to be sensitive to the details of the experiments, in particular the nature of the solvent and the presence and concentration of Fe(II) and Fe(III). For this reason, we performed two series of spin trapping experiments. The DMPO adducts in Fenton media were studied in the presence

of ethanol, methanol, and water as solvents, both in the absence and in the presence of Nafion.

Experimental Section

Materials. DMPO from Aldrich was used as received. The Nafion 117 membranes, with an equivalent weight of 1100 and a thickness of 0.178 mm, were obtained from DuPont. In the formula for the polymer given above $m = 6.5$; the mean repeat unit of the backbone consists therefore of 14 CF_2 groups and 1 CF group. The membranes were treated as reported²⁸ and stored in deionized water. For ESR measurements, the membranes were dried in a vacuum (10^{-4} Torr) for 24 h at ambient temperature and for 1 h at 373 K.

TiCl_3 , supplied by Aldrich and shipped in a pouch under nitrogen, was opened and stored in a glovebox under nitrogen. Aqueous solutions of H_2O_2 (30% and 3% w/v) were from Fisher Scientific. Methanol and ethanol (Fisher) were dehydrated using molecular sieves type 3A (Fisher) and deaerated in a glovebox by a stream of nitrogen. Fe(II) and Fe(III) salts from Aldrich were used as received.

Sample Preparation. In experiments with the Fenton reagent based on Fe(II) and Fe(III), the membranes were partially neutralized (10–20%) with ferrous sulfate heptahydrate $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ or ferric chloride hexahydrate $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ solutions in water; the exchange by TiCl_3 solutions in methanol was done in the glovebox.²⁸ The membranes were then vacuum-dried as described above. Samples were prepared using 1 mM solutions of $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ and $\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$, 100 mM DMPO solutions in the appropriate solvent, and 3% (w/v) aqueous solutions of H_2O_2 . Detailed information on the composition of each sample reported here is given in the Supporting Information. The ratio $[\text{H}_2\text{O}_2]/[\text{Fe(II)}]$ in the samples studied is given in Tables 1 and 2, and the corresponding concentrations of reactants and spin trap are given in the Supporting Information. When samples prepared with ethanol and methanol as solvents contained H_2O_2 , the solvent was a mixture containing $\approx 20\%$ v/v water.

Spectra of the Fenton solutions (no Nafion) were measured in capillary tubes prepared by flame sealing of disposable pipets. Samples containing Nafion were measured in regular ESR quartz sample tubes, which could be tuned only below ambient temperature.

ESR Measurements. X-band ESR spectra were recorded using a Bruker X-band EMX spectrometer operating at 9.7 GHz with 100 kHz magnetic field modulation and equipped with the Acquisit 32 bit WINEPR data system, version 3.01, for acquisition and manipulation and the ER 4111 VT variable temperature units. The microwave frequency was measured with a Hewlett-Packard 5350B microwave frequency counter.

The ESR settings for experiments with a Fenton solution in different solvents (no Nafion) were as follows: sweep width 75 G, microwave power typically 2 mW, time constant 40.96 ms, conversion time 81.92 ms, 4–10 scans, 1024–2048 points, modulation amplitude 0.5–0.8 G, and receiver gain 5.0×10^4 ; typical centerfield and microwave frequency were 3330 G and 9.352–9.364 GHz, respectively. In experiments with a Fenton system in different solvents and with Nafion as a source of counterions, the sweep width was increased to 140 G and the modulation amplitude to 1–3 G. The ESR absorption of Fe(III) was detected around $g = 4$ (centerfield 1565 G, peak-to-peak width ≈ 200 G at 77 K). The temperature was 300 K unless indicated otherwise.

Simulations. Electron spin resonance spectra in the rigid limit were simulated using SimFonia (Bruker) with manual parameter optimization. Solution spectra were simulated with the program

TABLE 1: Isotropic Hyperfine Splittings of DMPO Adducts in Solution

system	adduct	hyperfine splittings, G			relative intensity
		a_N	$a_{H\beta}$	$a_{H\gamma}$	
Fe(III)/EtOH/DMPO Figure 1	DMPO/*OC ₂ H ₅	13.6	7.8	1.6	87.7
	DMPO/*OOH	14.1	11.7	0.9	10.8
	DMPO/*CH(OH)CH ₃	15.2	21.9		1.5
Fe(II)/EtOH/DMPO/H ₂ O ₂ [H ₂ O ₂]/[Fe(II)] = 882	DMPO/*OC ₂ H ₅	14.1	8.4	1.6	100
Fe(II)/EtOH/DMPO/H ₂ O ₂ [H ₂ O ₂]/[Fe(II)] = 411 Figure 2	DMPO/*O ₂ ⁻	13.5	10.7	1.3	44.1
	DMPO/*CH(OH)CH ₃	15.3	22.1		25.7
	DMPO/*OC ₂ H ₅	13.9	8.1	1.6	16.5
	DMPO/*OOH	14.4	12.4	0.8	10.9
	DMPO Degrad			14.1	2.8
Fe(III)/EtOH/DMPO/H ₂ O ₂ [H ₂ O ₂]/[Fe(III)] = 411 Figure 3	DMPO/*O ₂ ⁻	13.6	10.8	1.3	43.38
	DMPO/*CH(OH)CH ₃	15.5	22.2		18.1
	DMPO/*OC ₂ H ₅	14.0	8.4	1.9	24.3
	DMPO/*OOH	14.6	12.8	0.8	7.2
	DMPO Degrad	14.6			7.0
Fe(II)/MeOH/DMPO/H ₂ O ₂ [H ₂ O ₂]/[Fe(II)] = 500 Figure 4	DMPO/*CH ₃	15.4	21.6		34.4
	DMPO/*OCH ₃	13.9	8.7	1.5	32.4
	DMPO/*O ₂ ⁻	13.7	10.7	1.4	23.1
	DMPO/*OOH	14.7	11.9	1.4	10.1
Fe(II)/H ₂ O/DMPO/H ₂ O ₂ [H ₂ O ₂]/[Fe(II)] = 500 Figure 5	DMPO/*OH	15.0	14.7		100

TABLE 2: Hyperfine and g-Tensor Components of DMPO Adducts

system	adduct	g	hyperfine splittings, G		
			¹⁴ N	H _β	H _γ
Nafion/Ti(III)10%/DMPO/MeOH/H ₂ O ₂ [H ₂ O ₂]/[Ti(III)] = 1455 Figure 6	DMPO/*OH	2.0089	4.9	14.7	4.5
		2.0057	4.9	14.7	4.5
		2.0024	33.7	6.8	14.7
		$g_{iso} = 2.0057$	$a_{iso} = 14.5^a$	$a_{iso} = 14.7$	$a_{iso} = 5.3$
Nafion/Fe(II)20%/H ₂ O ₂ /DMPO/H ₂ O at 160 K [H ₂ O ₂]/[Fe(II)] = 550 Figure 7	DMPO/*OH	2.0089	5.0	14.7	5.8
		2.0057	5.0	14.7	5.8
		2.0024	33.5	14.7	7.0
		$g_{iso} = 2.0057$	$a_{iso} = 14.5$	$a_{iso} = 14.7$	$a_{iso} = 6.2$
Nafion/Fe(II)20%/H ₂ O ₂ /DMPO/H ₂ O at 240 K [H ₂ O ₂]/[Fe(II)] = 550 Figure 7	DMPO/*R	2.0090	4.5	21.0	
		2.0060	4.5	21.0	
		2.0029	36.0	20.0	
		$g_{iso} = 2.0060$	$a_{iso} = 15.0$	$a_{iso} = 20.7$	

^a The hfs's for the DMPO/*OH adduct are $a_N = 14.8$ G and $a_{H\beta} = 14.8$ G in ref 25.

WinSim (NIEHS/NIH); the software provided an automatic fit and determined the relative intensity of each component in the case of a superposition of several components. The margin of error estimated for the hfs is on the order of 2–3%.

Results

To examine the effect of solvents on the nature and amount of the spin adducts, we first performed spin trapping experiments in the Fenton reagent in liquid solutions of ethanol, methanol, and water, in the absence of the polymer. The second set of experiments was performed in the presence of Nafion. The results for the Fenton solutions in the absence and presence of Nafion are summarized in Tables 1 and 2, respectively; the magnetic parameters for the spin adducts determined in this study are compared with literature values in Table 3.

Spin Trapping in Liquid Solutions. *Ethanol.* A strong ESR signal was obtained when mixing Fe(III) (as FeCl₃) and DMPO solutions in ethanol (EtOH), as seen in Figure 1. The signal was stable for at least 30 min after the addition of Fe(III)/EtOH to the DMPO/EtOH solution. The spectrum was simulated by a sum of three components: The major component, 87.7% of

the total intensity, was assigned to the DMPO/*OC₂H₅ adduct, with parameters in good agreement with the literature.^{8,10} The two additional minor species were assigned to DMPO/*OOH, 10.8%,^{6a,18,22} and DMPO/*CH(OH)CH₃, 1.5%.^{9,17,23}

The relative intensity of the DMPO/*OC₂H₅ adduct in the Fenton reagent based on Fe(II) (as FeCl₂) depends on the [H₂O₂]/[Fe(II)] molar ratio. For [H₂O₂]/[Fe(II)] = 882 the DMPO/*OC₂H₅ spin adduct was the only species present. For [H₂O₂]/[Fe(II)] = 411, a mixture of adducts was detected, as shown in Figure 2. The ESR spectrum measured immediately after sample preparation is spectrum A. The DMPO/*O₂⁻ adduct dominates the spectrum,^{6b} with hyperfine splittings: $a_N = 13.5$, $a_{H\beta} = 10.7$, and $a_{H\gamma} = 1.3$ G. The ESR signal after 2 h, spectrum B, was simulated by a superposition of five components, as shown in Table 1: DMPO/*O₂⁻ (the major component, 44.1%), DMPO/*CH(OH)CH₃,^{9,17,23} DMPO/*OC₂H₅, DMPO/*OOH, and the degradation product of DMPO.¹⁹

The same spin adducts were also identified when Fe(II) was replaced by Fe(III), as seen in Figure 3. As shown in Table 1, the relative intensities of the spin adducts are slightly different

TABLE 3: Summary of Isotropic Hyperfine Splittings of DMPO Adducts in Solution

spin adduct	hyperfine splittings, G			reference
	a_N	$a_{H\beta}$	$a_{H\gamma}$	
DMPO/ OC_2H_5	13.6	7.8	1.6	this work (EtOH)
	13.9	8.1	1.6	this work (EtOH 80%)
	14.1	8.4	1.9	this work (EtOH 80%)
	13.50	7.4	1.7	ref 10
	13.22	6.96	1.89	ref 8
	14.6	11.09	1.26	ref 22 (EtOH 1%)
DMPO/ $\text{CH}(\text{OH})\text{CH}_3$	15.2	21.9		this work
	15.3	22.1		this work
	15.5	22.2		this work
	15.9	23.1		ref 17
	15.0	21.7		ref 9
DMPO/ CH_3	15.4	21.6		this work
	16.4	23.6		ref 20
	16.3	23.4		ref 30
DMPO/ OCH_3	13.9	8.7	1.5	this work
	14.53	10.75	1.33	ref 21
DMPO/ OOH	14.1	11.7	0.9	this work
	14.4	12.4	0.8	this work
	14.6	12.8	0.8	this work
	14.7	11.9	1.4	this work
	14.2	11.34		ref 21
	14.3	11.70	1.25	refs 6a and 22
	14.3	11.4	1.25	ref 4
DMPO/ O_2^-	13.5	10.7	1.3	this work (EtOH 80%)
	10.8	1.3		this work (EtOH 80%)
	13.7		1.4	this work (MeOH 80%)
	14.1	11.3		ref 6b (water)
	13.3	10.4		ref 6b (MeOH)
	13.1	10.3		ref 6b (EtOH)
DMPO degradation	14.1 and 14.6 ≈ 15			this work ref 1

from those obtained in the case of the Fe(II) system, Figure 2. The OOH adduct can be formed in two ways: as a result of the decomposition of H_2O_2 by the ferric ion,²⁷ $\text{Fe(III)} + \text{H}_2\text{O}_2 \rightarrow \text{Fe(II)} + \text{H}^+ + \text{OOH}$, and by direct attack of Fe(III) on the solvent, as described in the Discussion.

The hfs's for the DMPO/ OC_2H_5 adduct are slightly different in the spectra shown in Figures 1–3; a_N varies in the range 13.6–14.1 G, and $a_{H\beta}$ in the range 7.8–8.4 G. Such hfs

variations are assigned to the different local polarity, as amply demonstrated in numerous studies of nitroxide radicals.^{31,32} To further check this aspect, we have repeated some experiments in the presence of various concentrations of NaCl instead of H_2O_2 , as suggested by Yoshimura et al.¹⁸ In these experiments the lines were broader. In addition, the hfs's of the various adducts were slightly higher; for adduct DMPO/ OC_2H_5 , for example, a_N increased from 13.75 to 13.9 G, and $a_{H\beta}$ increased

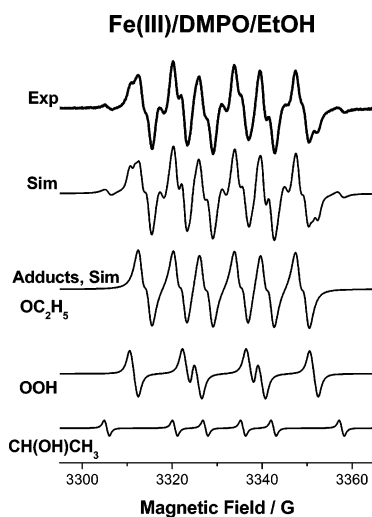


Figure 1. Electron spin resonance spectrum of DMPO adducts obtained in the system Fe(III)/EtOH–DMPO/EtOH. Top: Experimental spectrum at 300 K. Bottom: Simulated spectrum. Simulated spectra of each adduct are also shown. The corresponding hfs and relative intensities are given in Table 1.

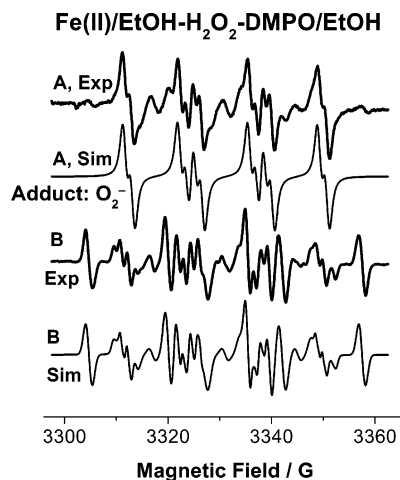


Figure 2. Electron spin resonance spectra at 300 K of DMPO adducts obtained in the system Fe(II)/EtOH– H_2O_2 –DMPO/EtOH ($[\text{H}_2\text{O}_2]/[\text{Fe(II)}] = 411$). (A) Experimental spectrum measured immediately after mixing of components and corresponding simulated spectrum. (B) Same as part A, except measured 2 h after mixing. The nature of the adducts and corresponding hfs and relative intensities are given in Table 1.

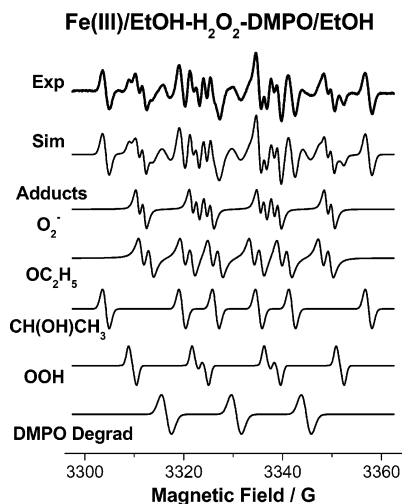


Figure 3. Electron spin resonance spectrum at 300 K of DMPO adducts obtained in the system Fe(III)/EtOH–H₂O₂–DMPO/EtOH. The top two traces are experimental and corresponding simulated spectra. Simulated spectra of the five DMPO adducts are also shown. The corresponding hfs and relative intensities are given in Table 1.

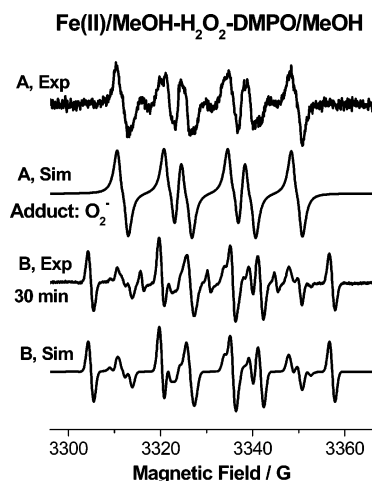


Figure 4. Electron spin resonance spectra at 300 K of DMPO adducts obtained in the system Fe(II)/MeOH–H₂O₂–DMPO/MeOH ([H₂O₂]/[Fe(II)] = 500). (A) Experimental spectrum measured immediately after mixing of components and corresponding simulated spectrum. (B) Same as part A, except measured 30 min after mixing. The nature of the adducts and corresponding hfs and relative intensities are given in Table 1.

from 7.87 to 8.50 G when the concentration of salt increased from 10 to 20 mM. The corresponding ESR spectra are not shown.

Methanol. The Fenton reagent based on (Fe(II)) in methanol (MeOH) generated several radicals that were trapped by DMPO. Figure 4 shows the ESR spectra measured in the Fenton reagent in methanol ([H₂O₂]/[Fe(II)] = 500). The dominant adduct in the ESR (spectrum A, measured immediately after sample preparation) is assigned to the DMPO•O₂[•] adduct,^{6a} with simulated parameters: $a_N = 13.8$ G, $a_{H\beta} = 10.2$ G, and $a_{H\gamma} = 1.2$ G. After 30 min the other adducts appeared, as seen in spectrum B in Figure 4. Computer simulation indicated the presence of four radical adducts: DMPO•Me,^{9,10,20,30} DMPO•OMe, DMPO•O₂[•], and DMPO•OOH. The relative intensities are shown in Table 1. In comparison to the literature,²¹ the splittings of ¹⁴N and H_β for DMPO•OMe are slightly lower in this work (13.9 G compared to 14.5 G, and 8.7 G compared to 10.7 G), again due to the different local polarity, which can change, depending on the concentration of the cation and other

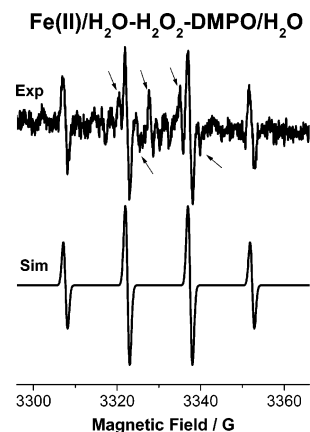


Figure 5. Experimental and corresponding simulated ESR spectra at 300 K of DMPO adducts obtained in the system Fe(II)/H₂O–H₂O₂–DMPO/H₂O ([H₂O₂]/[Fe(II)] = 500). See Table 1.

ionic species and on solvent composition. The higher values given in ref 21 were measured in a solvent that contained only 5% methanol and the rest water. In our samples the solvent was either 100% ethanol or methanol (in the absence of H₂O₂) or contained up to 20% v/v water (in the presence of H₂O₂). Slight variations are also possible due to the simulation procedure. Much closer parameters were usually obtained when one spin adduct only was present and the simulation was not complicated by superposition of several components.

Water. The DMPO•OH radical adduct²⁵ was the only species obtained in the Fenton reagent based on Fe(II) in water, Figure 5. The hyperfine splittings obtained by computer simulation are $a_N = 15.0$ G and $a_{H\beta} = 14.7$ G.

Spin Trapping in the Fenton Reagent in the Presence of Nafion. The spin adducts detected in the presence of Nafion with ethanol as a solvent were similar to the situation in the absence of Nafion (spectra not shown): DMPO•OC₂H₅, DMPO•OOH, DMPO•CH(OH)CH₃, the degradation product of DMPO in the absence of H₂O₂, and the additional DMPO•O₂[•] adduct in the presence of H₂O₂.

For methanol and water as solvents, ESR signals were obtained only after addition of H₂O₂. Figure 6A presents ESR spectra at 200 K in the system Nafion/Ti(III)(10%)/H₂O₂/DMPO/MeOH ([H₂O₂]/[Ti(III)] = 1455). The top spectrum was recorded 3 min after sample preparation and was simulated with the hyperfine tensor components given in Table 2. The signal was assigned to the DMPO•OH adduct. A similar spectrum was obtained after drying the membrane, as shown in the two bottom traces in Figure 6A. To prove this assignment, the sample temperature was raised from 200 to 280 K, lowered to 200 K to obtain the “rigid limit” spectrum, and raised again to 300 K; corresponding ESR spectra are shown in Figure 6B. The spectra prove the stability of the adduct and the fact that only one species is present, and this species is the DMPO•OH adduct.

In the Fenton reagent with water as solvent the nature of the adducts was studied after treatment of the membrane at a temperature typical of fuel cell operation, 358 K. Membranes were exchanged with FeSO₄ (20%) and left to react with 30% H₂O₂ at 358 K for 3 h ([H₂O₂]/[Fe(II)] = 550), followed by addition of the aqueous solution of DMPO. The temperature variation of the ESR spectra is shown in Figure 7A. Spectra below 230 K show the presence of the DMPO•OH adduct (near the rigid limit), while the spectra at and above 230 K are typical of a DMPO/carbon-centered adduct. ESR spectra at 240 and 160 K and corresponding simulated spectra are shown in Figure

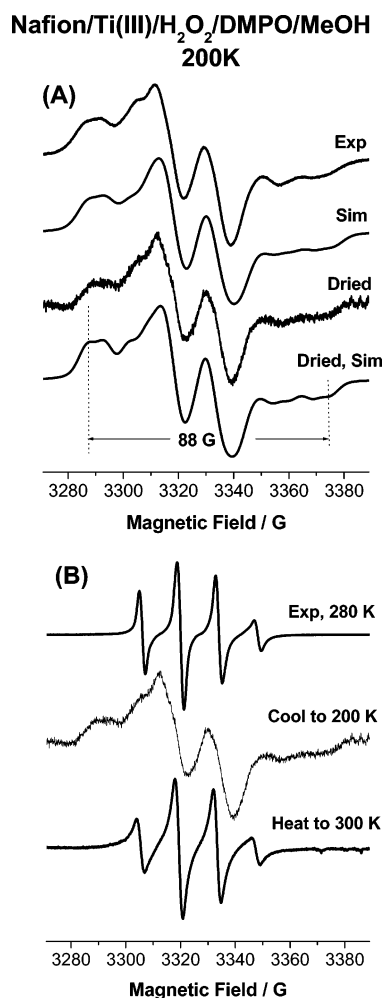


Figure 6. Electron spin resonance spectra of DMPO adducts obtained in the system Nafion/Ti(III)(10%)/DMPO/MeOH/H₂O₂ at 200 K. (A) Top two traces are the experimental and corresponding simulated spectra. Bottom two traces are from the same preparation, but the solution containing DMPO, MeOH, and H₂O₂ was removed, and the membrane was dried in a vacuum for 30 min. (B) Evolution of the ESR spectra of the dried membrane initially at 280 K, then cooled to 200 K and heated to 300 K. See Table 2.

7B. The calculated ESR parameters for the two adducts are given in Table 2. The DMPO/carbon-centered spin adduct is assigned to a Nafion-derived fragment, *vide infra*.

Discussion

Hyperfine Splittings for DMPO Adducts with Oxygen Radicals. The presence and hfs for the DMPO/•OH adduct are well established; in most cases $a_N \approx a_{H\beta} \approx 15$ G,^{11,25} as also measured in this study (Table 2). It is interesting to note, however, that the presence of this adduct was detected only with water as a solvent of the Fenton reagent (Figure 5) and for water and methanol as solvents for the Fenton reagent in the presence of Nafion. Two conclusions can be derived from this result. First, •OH radicals react with methanol, probably by hydrogen abstraction, to produce •OCH₃ and •CH₃ radicals and with H₂O₂ to produce •OOH and O₂•⁻ radicals and the corresponding adducts. Even with water as the solvent, when the DMPO/•OH adduct is dominant, the intensity of this adduct is low, and the presence of additional adducts, even less intense, is suggested and indicated by arrows in Figure 5. Second, •OH radicals are more stable in the presence of Nafion; for both water and methanol as solvents the DMPO/•OH adduct is present. This

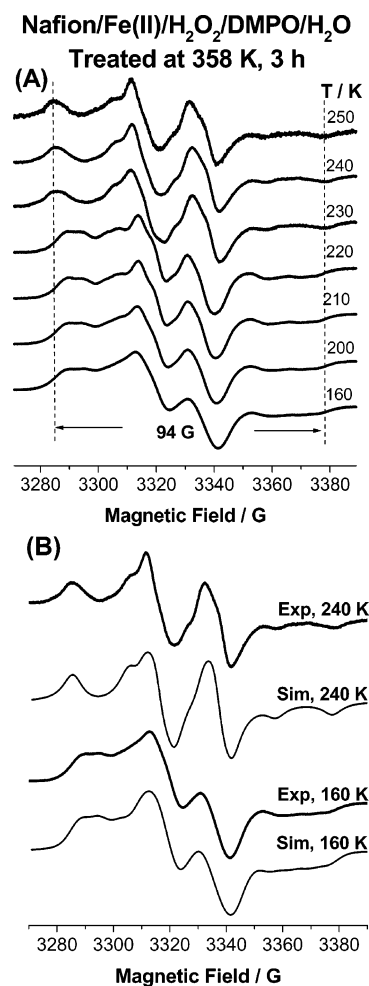
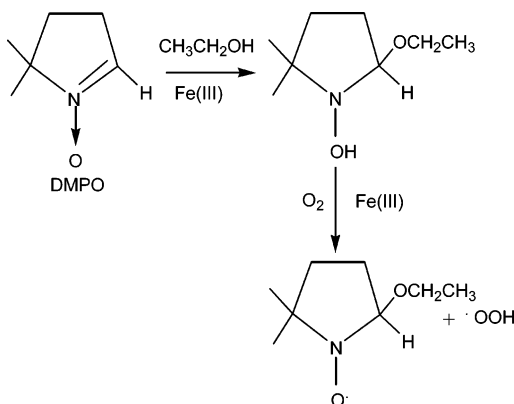


Figure 7. Electron spin resonance spectra of DMPO adducts obtained in the system Nafion/Fe(II)(20%)/H₂O₂/DMPO/H₂O. (A) Temperature variation of ESR spectra. (B) Experimental and corresponding simulated spectra at 240 and 160 K. The corresponding magnetic parameters for the •OH and carbon-centered radical •R adducts are given in Table 2.

effect may be due to the formation of •OH radicals close to the polymer chains and to a reduced mobility of these radicals. It is important to mention that signals from molecular oxygen were detected in dry Nafion, probably trapped and stabilized inside small pores formed in the self-assembled ionomer chains during removal of water.³³ The different behavior of •OH radicals when ethanol is the solvent may be due to the fact that ethanol is a better swelling solvent for Nafion, as determined by the degree of swell³⁴ and by the variation of ¹⁹F NMR spectra of Nafion swollen by water, methanol, and ethanol.³⁵

The assignment of signals from both the DMPO/•OOH and the DMPO/O₂•⁻ adducts requires an explanation. In much of the spin trapping literature these two adducts are given the same parameters, as seen, for example, in ref 4, because in aqueous systems the hfs's are about the same: $a_N \approx 14$ G and $a_{H\beta} \approx 11$ G. The concentrations of the •OOH and O₂•⁻ radicals are expected to depend on the pH, as seen in eq 4 in the Introduction. The rate constants for the reaction of DMPO with •OOH and with O₂•⁻ radicals were determined by fitting the variation of K_{app} with pH; K_{app} is the apparent rate constant for the reaction of DMPO with •OOH/O₂•⁻.³⁶ According to this model, trapping of •OOH predominates below pH = 7.7, and trapping of O₂•⁻ above this pH value.

We found a convincing precedent for the assignment of different hfs's for the DMPO/O₂•⁻ and DMPO/•OOH adducts in refs 6a and 6b: The hfs's for the DMPO/O₂•⁻ adduct vary

SCHEME 1: Formation of a DMPO Adduct by Fe(III)-Catalyzed Nucleophilic Addition of Ethanol to DMPO

with the solvent^{6b} and are in good agreement with the values assigned in different solvents in this study, as shown in Table 3. The slightly higher for a_N in methanol, 13.7, compared with 13.5 G in ethanol, is also in agreement with measured data.^{6b} The assignment of the DMPO/ $\bullet\text{OOH}$ adduct followed most data given in the literature.^{6a,21,24} These assignments mean that in the same sample we detected *separately* DMPO/ $\bullet\text{O}_2^-$ and DMPO/ $\bullet\text{OOH}$ adducts. To the best of our knowledge, this is the first time that the two adducts were detected simultaneously in a given system.

The DMPO/ $\bullet\text{O}_2^-$ and DMPO/ $\bullet\text{OOH}$ adducts were detected only when the DMPO/ $\bullet\text{OH}$ adduct was absent, suggesting that the $\bullet\text{OH}$ radical is formed first, and reacts further to form the other radicals.^{6a}

The nature of the DMPO adducts detected in this study can be compared with the direct detection of radicals in Nafion exposed to the Fenton reagent in water. The $\bullet\text{OH}$ adduct was detected by spin trapping and not by direct ESR. In both types of studies the $\bullet\text{OOH}$ and $\text{O}_2^{\bullet-}$ radicals were detected. The peroxy radical $\text{TiOO}\bullet$ was detected by direct ESR, but not by spin trapping, in line with the expected instability of peroxy radical DMPO adducts.²¹ The major difference is in the detection of the $\text{O}_2^{\bullet-}$ adduct. In direct ESR experiments this radical was detected only in the dried membrane. The signal is absent in direct ESR measurements of the swollen membrane because the ground state of $\text{O}_2^{\bullet-}$ is degenerate, with $g_{xx} = g_{yy} = 0$ and $g_{zz} = 4$ (z is the direction of the O—O bond).³⁷ Electron spin resonance signals are detected when the degeneracy is removed, for instance, by attachment of the radical to a surface. In solid matrixes all g values are close to 2; in dry Nafion the values were $g_1 = 2.0190$, $g_2 = 2.0092$, and $g_3 = 2.0036$ ($g_{\text{iso}} = 2.0106$). The results presented here suggest that the full picture on the type of radicals present in Nafion exposed to the Fenton reagent can be obtained by a combination of spin trapping and direct ESR.

Oxygen- and Carbon-Centered Radical Adducts in Fenton Solutions. The ESR spectra in the Fenton reagent based on Fe(II)/ H_2O_2 and for Fe(III) with ethanol as a solvent and in the absence of H_2O_2 , Figure 1, show the complicated coexistence of several DMPO adducts. In the simplest system, Fe(III)/EtOH/DMPO, the presence of three spin adducts has been determined by simulation, as seen in Table 1. The major component is the DMPO/ $\bullet\text{OCH}_2\text{CH}_3$ adduct (87.7%). In some papers it was suggested that this adduct can be formed via hydrogen abstraction by $\bullet\text{OH}$ radicals. As $\bullet\text{OH}$ radicals are not expected in the absence of H_2O_2 , we propose the formation of the adduct DMPO/ $\bullet\text{OCH}_2\text{CH}_3$ by Fe(III)-catalyzed nucleophilic addition of ethanol to DMPO, as illustrated in Scheme 1.²² The oxygen

solubility in ethanol is higher than that in water,³⁸ leading to the formation of the $\bullet\text{OOH}$ radical and of the corresponding spin adduct DMPO/ $\bullet\text{CH}(\text{OH})\text{CH}_3$ was also detected, exhibiting the typical large H_β splitting, 21.9 G.

The Fe(III)-catalyzed nucleophilic addition of ethanol to DMPO is expected to occur also in the case of the Fenton reagent Fe(II)/ H_2O_2 , where the presence of Fe(III) was detected by the strong ESR signal at $g \approx 4$. For these reasons the nature of the adducts depends on the $[\text{Fe(II)}]/[\text{H}_2\text{O}_2]$ molar ratio. When this ratio was high (882), the only adduct formed was DMPO/ $\bullet\text{OCH}_2\text{CH}_3$. For a lower ratio, 411, a mixture of radical adducts was formed, Figure 2, including the DMPO degradation product. A similar mixture was obtained when Fe(II) was replaced by Fe(III). These results indicate that in the Fenton reagent there are several reactions that lead to the formation of adducts. The Fenton reagent based on ethanol is complicated and should be avoided when the main goal is to detect the oxygen and membrane-derived fragments.

In the Fenton reagent based on methanol with $[\text{Fe(II)}]/[\text{H}_2\text{O}_2] = 500$, the DMPO/ $\bullet\text{O}_2^-$ adduct was dominant immediately after sample preparation, as seen in the top spectrum of Figure 4. After 30 min a mixture of adducts was detected, consisting of oxygen adducts and the carbon-centered DMPO/ $\bullet\text{CH}_3$ adduct. The relative intensity of the DMPO/ $\bullet\text{O}_2^-$ adduct was considerably lowered, from 100% immediately after mixing of the components to 23.1% after 30 min. The composition of the adduct mixture suggests involvement of the $\bullet\text{OH}$ radicals in the formation of methanol-derived radicals that become trapped by DMPO, and further reaction that leads to the formation of DMPO/ $\bullet\text{OOH}$ and DMPO/ $\bullet\text{O}_2^-$ adducts.

Electron Spin Resonance Spectrum of the Hydroxyl Radical Adduct, DMPO/ $\bullet\text{OH}$, in the Rigid Limit. In the presence of Nafion, most experiments were conducted at lower than ambient temperatures, because of the need for sample tubes with larger diameters that could be tuned in the ESR resonator only as frozen samples. For the Fenton reagent based on Ti(III) with methanol as solvent, the ESR spectrum was a “rigid limit” type that extended over ≈ 88 G at 200 K; a similar spectrum, though weaker, was obtained when the membrane was dried, as seen in Figure 6A. Heating of the dry membrane to 300 K indicated clearly that the adduct is DMPO/ $\bullet\text{OH}$, Figure 6B. It seems that the presence of Nafion reduces even further the reactivity of the $\bullet\text{OH}$ radicals with methanol, and the corresponding DMPO/ $\bullet\text{OH}$ adduct is the only species detected. The full tensor components of this adduct were deduced by simulation and are shown in Table 2. Quite unusual is the large value of the tensor components for H_γ , with $a_{H_\gamma} = 5.3$ or 6.2 G depending on the solvent (Table 2). Such a large hfs should be detected in the ESR spectrum of the DMPO/ $\bullet\text{OH}$ adduct at 300 K; the fact that it is not detected (see, for instance, Figure 6) may indicate that the conformation of the radical at low temperatures is different compared to that at 300 K.

Evidence for a DMPO Adduct of a Membrane-Derived Fluorinated Fragment. The temperature variation of the ESR spectra in the Fenton reagent based on water is shown in Figure 7A. The DMPO aqueous solution was added after Nafion was treated at 358 K in the Fenton reagent for 3 h. This experiment was an attempt to mimic the temperature conditions in a fuel cell. The ESR spectrum at 160 K extends over 88 G, as in the case of the methanol system described above (Figure 6), indicating the presence of the DMPO/ $\bullet\text{OH}$ adduct in a similar environment. With increasing temperature the spectrum changes, and at 240 K the full sweep increases to 94 G, indicating a

different type of adduct. Simulations of the spectra at 160 and 240 K are shown in Figure 7B, and the corresponding parameters in Table 2. The magnetic parameters deduced by simulating the spectrum at 160 K are almost the same as those used in the simulation of the DMPO/•OH adduct in the system with methanol as solvent. The spectrum at 240 K indicates clearly a carbon-centered adduct. The only possible source for this adduct is Nafion, which is the only source of carbon radicals. The signal intensity at 240 K is very high, indicating a high radical concentration; this is an additional reason for assigning this adduct to a DMPO/perfluorinated radical adduct. The identity of the radical was not determined, however. In general the hfs from the H_β proton in carbon-centered radical adducts is larger than that from the nitrogen. For the DMPO/ CH_3 adduct, for example, the hfs's are $a_N = 14.31$ G and $a_{H_\beta} = 20.52$ G; for the DMPO/ CF_3 adduct, however, the hfs's are $a_N = 13.22$ G, $a_{H_\beta} = 15.54$ G, and $a_F = 1.01$ G (3F) in benzene as solvent.³⁹ It is clear that the fluorinated adduct measured in this study, with $a_{H_\beta} = 20.7$ G, is not a CF_3 adduct. We are currently performing spin trapping of radicals formed in model fluorinated compounds as a way to fully identify the carbon-centered radical adduct.⁴⁰

Conclusions

Spin trapping by DMPO in Fenton media with ethanol, methanol, and water as solvents was studied in the presence and absence of Nafion. Ethanol-derived adducts, DMPO/•OC₂H₅ and DMPO/•CH(OH)CH₃, were identified, and their presence was rationalized by Fe(III)-catalyzed nucleophilic addition of ethanol to the spin trap and hydrogen abstraction by •OH radicals. The oxygen radical adducts DMPO/•O₂⁻ and DMPO/•OOH were also detected. The relative intensity of the various spin adducts depends on the [Fe(II)]/[H₂O₂] molar ratio, and under certain conditions, only the DMPO/•OC₂H₅ adduct was present. The same types of adducts were detected in the presence or absence of Nafion.

In Fenton media with methanol as solvent (and no Nafion) the DMPO/•O₂⁻ adduct dominated immediately after sample preparation, and a mixture consisting of DMPO/•OCH₃, DMPO/•CH₃, DMPO/•O₂⁻, and DMPO/•OOH adducts was detected after 30 min. In the presence of Nafion only the DMPO/•OH adduct was detected. To the best of our knowledge this study detected for the first time the simultaneous presence of both DMPO/•O₂⁻ and DMPO/•OOH adducts in the same system.

For water as solvent only the DMPO/•OH adduct was detected, both in the absence and in the presence of Nafion. The full hyperfine tensor components of this adduct near the rigid limit were determined in Fenton media in the presence of Nafion with water and methanol as solvents. A significant H_γ splitting in the DMPO/•OH adduct was measured: The largest hyperfine tensor component, obtained by simulation, was 6.8 and 7.0 G, and a_{H_γ} was 5.3 and 6.2 G, depending on the solvent. This large hfs should be detected in the spectra measured in the motionally narrowed regime, near 300 K. The fact that it was not detected suggests different conformations of the DMPO/•OH adduct at different temperatures.

In Nafion exposed to the Fenton reagent based on water at 358 K for 3 h, a DMPO adduct of a carbon-centered radical was also identified and assigned to a Nafion-derived fragment. The exact nature of the fragment is under investigation.

Data obtained by spin trapping were compared with the results of a direct ESR study of the Fenton reagent/Nafion. This comparison led to the conclusion that direct ESR and spin trapping provide complementary information on the nature of the radicals present.

The major limitation of DMPO as a spin trap for radical identification in Fenton media is the formation and trapping of solvent-derived free radicals in ethanol and, to some extent, in methanol; for water as solvent, the method appears set, however, to become useful when treatment of the polymer is carried out at higher than ambient temperatures.

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Supporting Information Available: Experimental details. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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