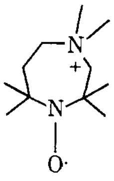
Communications to the Editor

Spin Labeling of Ion Exchange Membranes

Use of spin labeled organic ions is a convenient way to apply the spin label method to ion exchanger gels and membranes. A study concerning a spin labeled anion in ion exchange resins has recently been published.[1] These relatively small ions, stable at all pH in aqueous solutions, can be used to study the mechanism of electrostatic interactions in ion-exchange membranes.

In the present work, the interaction between a spin labeled monovalent cation (I) and the carboxylic sites in a cation exchange membrane obtained by cross-linking of carboxymethylcellulose (CMC) is studied.



I

Experimental. The preparation of the spin labeled cation under its iodide form (I) had been described previously.[2] The membranes have been prepared by cross-linking of CMC with degree of substitution 0.7 using formol as a reagent and special experimental conditions as established by one of us.[3]

Different degrees of cross-linking are obtained for different reaction times (from 20 min to 3 hr); the membranes' thicknesses are 50 μ and their capacity 3.2 × 10^{-3} equiv/g. The membranes have been stabilized under various ionic forms (H, Li, Na, K, Ca) before labeling by contact with a solution of counterions I (0.1 mg/ml or 3.3 × 10^{-7} mol/ml) followed by repeated washing, with distilled water.

The electron spin resonance (ESR) spectra are obtained at 20°C with a Varian V 4501 spectrometer (X band, modulation 100 kcycles/s) using Scanco S 808 flat cells.

To test the influence of the degree of swelling, 10 mg of each membrane under Na form are stabilized with 5 ml of I and washed. The labeled membranes are characterized by their ESR spectra (Table I).

The unbound nitroxides are determined directly by ESR on the supernatant to deduce the fraction of counterions I bound on carboxylic sites; the fraction of I bound is also obtained by extraction of the labeled membrane with 2 ml of a $CaCl_2$ 0.1 N solution and titration of I (Table II).

The various counterions affinity order for the membrane ionic sites is established by two different methods. Concentration of I in the supernatant is determined by ESR in one case after exclusion by the different counterions (chlorides) on the prelabeled membrane or after stabilization of membranes under different ionic forms with equal quantity of I. Following the second process, 10 mg of membrane had been stabilized with 1 ml of I and washed; the fraction of I bound is determined on the supernatant or after extraction by 2 ml $CaCl_2$ 0.1 N (Table III).

Results and Discussion. (1) The ESR spectra of the counterions I in aqueous solutions (a_N = 15.6 ± 0.2 G), of a spin labeled membrane in water (a_N = 15.3 ± 0.2 G), of the labeled dried membrane (Total width of the spectra L = 65 G), and of the labeled membrane in a solvent acetone–water (90:10, w/w) are respectively given in Figure 1 (spectra A to D). The usual three-line spectra (Figure 1, A and B) is characteristic of a nonimmobilized radical; the only difference between the two spectra is the relative height of the three lines (I_{+1},I_0,I_{-1} from lower to higher field). The spectrum C (Figure 1) on the contrary is characteristic of an immobilized nitroxide free radical. The spectrum D can be attributed to a mixture of free and immobilized forms.

(2) The high field and central lines heights ratio I_{-1}/I_0 is directly related to the radical rotational correlation time τ_c. This ratio is always lower than 1 and lower values of I_{-1}/I_0 correspond to larger τ_c values.

In Table I, the I_{-1}/I_0 ratios are given for membranes in water as a function of the degree of swelling which decreases as the degree of cross-linking increases. All the spectra display the same general shape as in Figure 1B but the radical immobilization increases with the degree of

Table I
Variation of the Ratio I_{-1}/I_0 as a Function of the Degree of Swelling of the Membrane[a]

H[b]	I_{-1}/I_0[c]
5	0.863
3	0.837
1.5	0.745
0.93	0.668
0.55	0.611
Free radical	0.940

[a] Results corresponding to 10 mg of Na form membrane labeled with 5 ml of 3.3 × 10^{-7} mol/ml of solution I. [b] H = weight of water by weight of dry membrane under Na form (g/g). [c] I_{-1}/I_0 = high field and central lines heights ratio (spectra A and B).

Table II
Degree of Association of Nitroxide as a Function of the Membrane Swelling[a]

H	Degree of association[b] 1	2
5	2.5 × 10^{-2}	1.88 × 10^{-2}
3	2.84 × 10^{-2}	1.94 × 10^{-2}
1.5	1.96 × 10^{-2}	1.40 × 10^{-2}
1	2.06 × 10^{-2}	1.26 × 10^{-2}
0.93	1.72 × 10^{-2}	
0.55	1.69 × 10^{-2}	0.98 × 10^{-2}

[a] Results corresponding to 10 mg of Na form membrane labeled with 5 ml of 3.3 × 10^{-7} mol/ml of solution I. [b] Degree of association = number of nitroxide I bonded per membrane ionic site obtained: (1) by determination in the supernatant; (2) by exclusion with a fixed excess of $CaCl_2$ on labeled membrane.

Table III
Affinity of Counterions for Membrane Ionic Site (H = 0.85)[a]

Ionic form of the membrane	Degree of association[b] 1	2
Li^+	4.81 × 10^{-3}	3.64 × 10^{-3}
K^+	3.42 × 10^{-3}	2.47 × 10^{-3}
H^+	2.87 × 10^{-3}	1.27 × 10^{-3}
Ca^{2+}	0.61 × 10^{-3}	~0

[a] Results corresponding to 10 mg of Na form membrane labeled with 1 ml of 3.3 × 10^{-7} mol/ml of solution I. [b] See Table II.

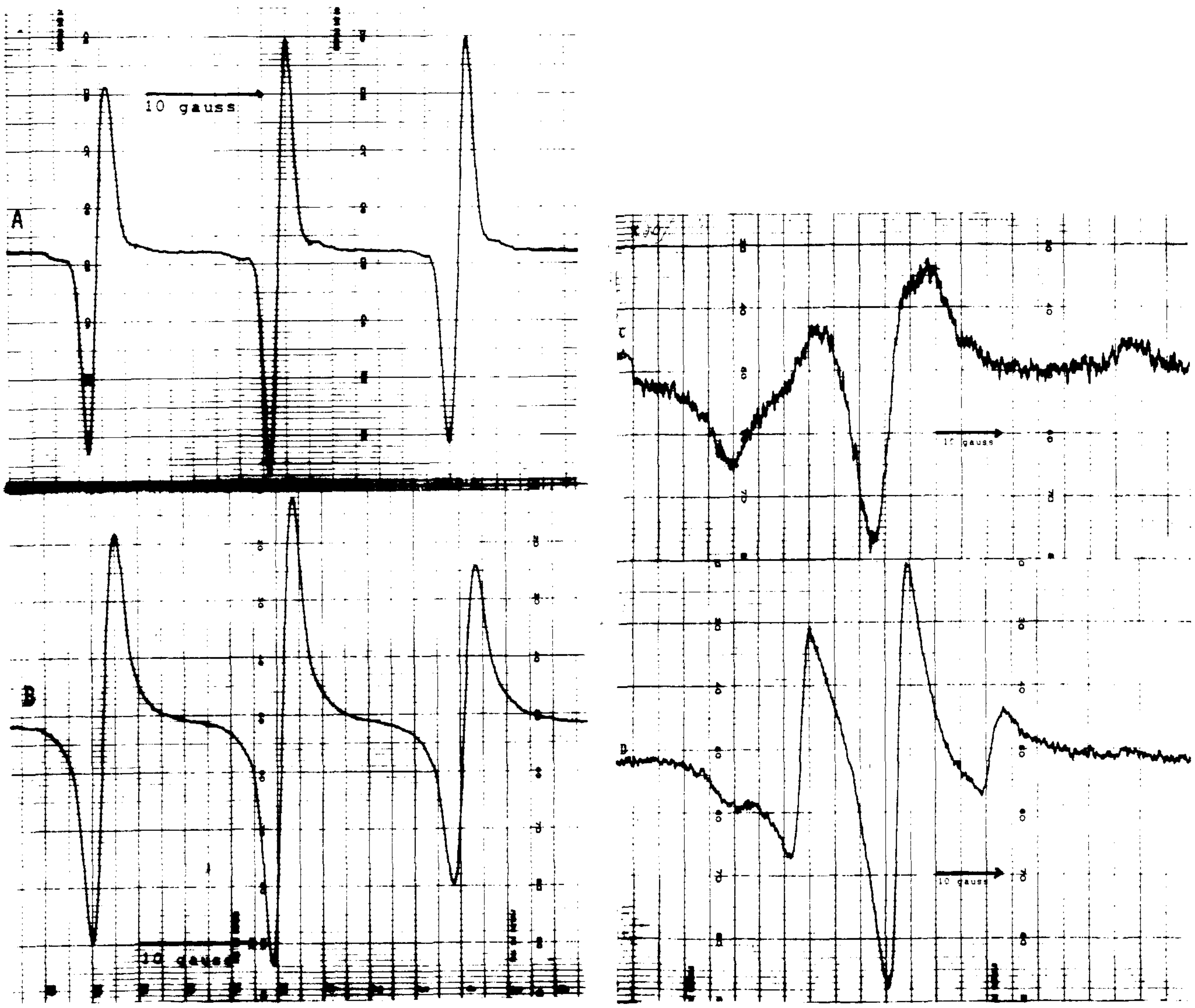


Figure 1. EPR spectra of: (A) radical I in aqueous solution. Membrane labeled with I: (B) in water; (C) in the dry state; (D) in a mixture of acetone–H_2O (90:10)

cross-linking. Two processes can explain this effect: a mechanical effect resulting from the porous material size (the pore radii are larger than 9 Å[4]), or, more validly, by lowering of the activity coefficient of the counterions with cross-linking. This latter process can be due to the ion pairs formation as the electrostatic interactions increase. The second hypothesis is also consistent with results obtained using formamide as a solvent (Table Va) and with the quantity of I fixed in the membrane as a function of swelling (Table II). An increase of the dielectric constant without larger swelling than in water increases the mobility of the radical; the quantity of counterions I in the membrane decreases as the degree of cross-linking increases by Donnan effect.[5]

(3) To interpret the solvent effect, the membrane has been stabilized in different water–acetone mixtures (Table IV) and in different pure solvents (Table V).

(a) For water–acetone mixtures, the spectra of I on the dried membrane and in the membrane in acetone are characteristic of immobilized radicals; when the water content and as a consequence the degree of swelling increase the spectra change from C to D and at last to B (Table IVa). The radical mobility increases with the water content. If the water-swollen membrane is progressively dehydrated with acetone (Table IVb), τ_c increases and the spectra show

Table IV
I_{-1}/I_0 Ratios[a] and L Values[b] as a Function of the Composition of Acetone–Water Mixtures

Water in acetone (% w/w)	I_{-1}/I_0	L	Type of spectrum (Figure 1)
(a) Addition of Water in Acetone (Membrane H = 0.930)			
Dry membrane		62	C
0		64.4	C
10		62[c]	D
25	0.384	33	B
50	0.531	33	B
80	0.631	33	B
100	0.668	32.6	B
(b) Addition of Acetone in Water (Membrane H = 0.55)			
Acetone in water (% w/w)			
0	0.668	32.6	B
20	0.593	32.8	B
50	0.477	32.8	B
90	0.3	32.6	B
100		66	C
Free radical	0.940	32.8	A

[a] I_{-1}/I_0 defined as previously (Table I). [b] L total width of the spectra. [c] Superposition of immobilized and nonimmobilized spectra.

Table V
I_{-1}/I_0 Ratios and L Values in Various Solvents

Solvent	Dielectric constant[8]	I_{-1}/I_0	L	Type of spectrum (Figure 1)
(a) Addition of Solvent to Dry Labeled Membrane (H = 0.930)				
Ethanol	24		61.4	C
Methanol	32.6		62	C
Formamide	109	0.918	33	B
(b) Dry Labeled Membrane (H = 0.930) Successively Stabilized in the Following Solvents				
Acetone	19		64.4	C
Water	78	0.668	32.6	B
Formamide	109	0.840	31.4	B
Water	78	0.678	32.8	B
Methanol	32.6	0.416	32	B
Ethanol	24.3		62[a]	D
2-Methyl-2-butanol	5.82		62	D

[a] Superposition of immobilized and nonimmobilized spectra.

the progressive radical immobilization. The only difference between the two sets of results is obtained with the 90:10 acetone–water mixture and is attributed to a slow solvent exchange.

(b) The dry labeled membranes have been stabilized in pure solvents (Table Va). A kinetical effect is observed with formamide; the dried membrane immobilized spectrum changes immediately to a superposition of free and immobilized spectra on addition of formamide. This spectrum slowly becomes a three-line spectrum with a high ratio I_{-1}/I_0 compared to the value obtained with H_2O whose swelling is equal. This phenomena can be due to the kinetics of swelling in formamide.

From the comparison of results with ethanol and methanol (Table Va) whose dielectric constants are respectively equal to the calculated values for acetone–water mixtures 90:10, 75:25 (Table IVa), the influence of solvation is clear; in aqueous solution, the degree of swelling of the membrane is larger than in organic solvent and the τ_c observed decrease.

The same conclusion is obtained if the sequence of results of Table Vb is considered; the dehydration by solvent exchange is not complete. Radical immobilization is only obtained with low polarity solvent. In conclusion, when the solvent dielectric constant and its water content (or the degree of swelling) decrease, the fraction of tight binding increases with a corresponding total width of the spectra characteristic of a low polarity medium.[6]

(4) The ESR spectra for the labeled membrane parallel and perpendicular to the magnetic field are similar; the I_{-1}/I_0 ratios are the same and it seems that there is no anisotropy of the system.

(5) Using the spin labeled counterion, the order of affinity of various counterions has been established. The affinity decreases from Ca > H > K > Li (Table III) on the CMC membranes in accord with previously obtained results;[4] this sequence is opposite to the sequence obtained on CMC in aqueous solution[7] but inversions had been previously established in ion exchangers.

Conclusion. From a general point of view, the labeling with counterions like I is convenient to investigate properties of relatively loose membranes and ion exchange gels and resins. We have discussed the evidence of site binding of these counterions as a function of the degree of swelling of the membrane and of the solvent polarity. The mobility of the counterions decreases when the degree of cross-linking increases corresponding to an increase of the electrostatic potential and when the polarity of the solvent decreases. At last the labeled counterion I is useful to test rapidly and accurately the affinity order of different counterions for the membranes ionic sites.

References and Notes

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