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Multibore sample cell increases EPR sensitivity for aqueous samples

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Abstract

We have performed calculations, verified by experiment, to explain why the sensitivity of biological EPR can be dramatically increased by dividing the aqueous sample into separate compartments. In biological EPR, the major factor affecting sensitivity is the number of spins in the sample. For an aqueous sample at ambient temperature, this is limited by the requirement for a small volume, due to strong non-resonant absorption of microwaves by water. However, recent empirical studies have shown that this volume limitation can be greatly relieved by dividing the aqueous sample into separate volumes, which allows much more aqueous sample to be loaded into a resonant cavity without significant degradation of the cavity quality factor. Calculations, based on the Bruggeman mixing rule, show quantitatively that the composite aqueous sample has a permittivity much less than that of bulk water, depending on the aqueous volume fraction f. Analysis for X-band EPR spectroscopy shows that the optimal volume fraction of an aqueous composite sample, producing maximum sensitivity, is $f = 0.15$, increasing the sensitivity by a factor of 8.7, compared with an aqueous sample in a single tube.

Keywords: EPR; ESR; Composite; Water; Nitroxide

1. Introduction

The major factor limiting the sensitivity of biological EPR is the non-resonant absorption of water in the GHz range. The present study seeks to maximize the sensitivity of EPR for an aqueous sample at a fixed concentration of spins. EPR signal intensity S is proportional to the filling factor η and the unloaded quality factor Q'_U of the resonant cavity $S \propto \chi'' \eta Q'_U P^{1/2}$ [1]. At values of the microwave power P low enough to ensure the absence of saturation, the magnetic susceptibility χ'' is simply proportional to the concentration of spins N/V_s, where N is the number of spins and V_s is the sample volume, so at fixed incident power and sample concentration in the absence of saturation, this becomes

$$S \propto \eta Q'_U. \tag{1}$$

However, most EPR experiments are performed under conditions of moderate saturation, at a value of the microwave field amplitude H_1 that gives approximately the maximum signal (usually around half saturation). Under these conditions

$$S \propto (\omega \mu_0 V_s \eta Q'_U)^{1/2}, \tag{2}$$

where ω is the angular resonance frequency, and μ_0 is the permeability of free space [2,3]. Eqs. (1 and 2) characterize the signal intensity of non-saturated and saturated aqueous samples, respectively. The filling factor reflects the volume of the aqueous sample and the distribution of microwave field H_1 in the cavity [4]

$$\eta = \int_s H_1^2 \, \mathrm{Sin}^2 \varphi \, \mathrm{d}V \Big/ \int_c H_1^2 \, \mathrm{d}V, \tag{3}$$

where indices s and c reflect integration over the sample volume and the cavity volume, respectively, and φ is the angle between the DC polarizing magnetic field and H_1 (it is 90° for all experiments considered in this work, so the angle dependence can be eliminated).

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The quality factor reflects the non-resonant absorption of water (loss), which also depends on the volume of the aqueous sample [3,5]:

$$1/Q'_{\mathrm{U}} = 1/Q_{\mathrm{U}} + 1/Q_{\mathrm{E}}, \tag{4}$$

$$Q_{\mathrm{E}} = \int_{\mathrm{c}} E^2 \,\mathrm{d}V \Big/ ((1/2)\varepsilon'' \int_{\mathrm{s}} E^2 \,\mathrm{d}V), \tag{5}$$

where $1/Q_{\mathrm{U}}$ is proportional to loss in the cavity walls of the empty cavity (Q_{U} is the quality factor of empty cavity) and $1/Q_{\mathrm{E}}$ reflects non-resonant absorption by the sample (ratio of the energy, stored in a cavity to the energy, dissipated in the sample, Eq. (5)).

Increased volume of an aqueous sample in an EPR cavity, at constant concentration, leads to a competition between signal increase due to a larger number of spins and signal decrease because of increased losses. For an aqueous sample at a fixed H_1, maximum sensitivity (2) occurs at a sample size that depends on the properties of the cavity (quality factor, microwave field distribution) and the sample's permittivity [2,5].

Once the sample volume has been optimized, another opportunity is to increase the filling factor through the redistribution of H_1 in the cavity; for example, H_1 can be concentrated on the sample with a cavity insert such as a folded half-wave resonator [6] or a dielectric hollow cylinder [7]. In both cases, the redistribution of the microwave magnetic field leads to a concentration of the electric field at the sample, thus increasing losses.

It has been found empirically that dividing the volume of an aqueous sample into separate vessels can increase the volume without substantial degradation of Q, thus increasing EPR sensitivity for a sample at constant concentration [8–12]. For example, three stacked flat cells increased signal intensity 4.4 [12] and 3.8 [11] times, relative to a single capillary. Recent finite-element modeling of a stacked flat cell in a rectangular cavity predicted that the signal intensity can be further improved, by at least a factor of two, through careful optimization of flat cell size and intercell distance in a standard rectangular cavity [13,14]. The Bruker AquaX cell (19-bore aqueous sample) [10] gives 4.5-fold signal improvement for an aqueous sample, compared to a single bore aqueous sample; finite-element analysis of a similar structure predicted that this is the maximum possible enhancement for a multibore sample configuration [15].

Why does Q increase when an aqueous sample is divided into separate volumes? It is not due to elimination of conductivity, since the conductivity of water is low (2 μS/cm), and increased conductivity due to salts does not change signal intensity much [2]. On the other hand, increasing the sample permittivity (e.g., due to a temperature change) causes a large degradation of Q [2]. Therefore, we hypothesized that the division of an aqueous sample into separate volumes decreases its permittivity. A divided sample can be treated as a composite material with dielectric properties that are different from the dielectric properties of its components. Several methods have been developed to determine the permittivity of a composite material [16], as discussed below. In the present study, the effective permittivity of a multibore aqueous sample was determined with the Bruggeman mixing rule [16–18]. This method relies on the average field concept and treats the environment of every inclusion (e.g., a bore in a multibore assembly) as an infinite uniform medium of permittivity $\varepsilon_{\mathrm{eff}}$. The applied electric field is considered as the average electric field that exists far away from the inclusion. There is a charge at the surface of every inclusion and an associated dipole moment, and the total polarization is the sum of the individual dipole moments of the inclusions. In the Bruggeman mixing rule, the same consideration is valid for the host medium, which also can be treated as an inclusion. Surface charges of both components are equal, and the polarizations of two components must be compensated. This requirement introduces the concept of effective permittivity for a composite material, which is determined by the permittivities of sample components and their volume fractions. From the effective permittivity of a multibore aqueous sample, we calculated EPR parameters such as resonant frequency and signal intensity, and compared this prediction with experimental data. We used this approach to optimize the multibore configuration of an aqueous sample for maximum performance of biological EPR.

2. Methods

2.1. Experimental

EPR experiments were performed with a Bruker EleXsys E500 spectrometer (Bruker Instruments, Billerica, MA), using the Bruker SHQ cavity with a quartz dewar (Wilmad, Buena, NJ). The temperature was maintained at 25 °C, using a nitrogen gas-flow temperature controller, and monitored with a digital thermometer using a Sensortek (Clifton, NJ) IT-21 thermocouple microprobe inserted into the top of the sample tube, such that it did not affect the EPR signal. All measurements were done at critical coupling. The sample was an aqueous solution of 100 μM TEMPO spin label (Aldrich, Milwaukee, WI), prepared with doubly distilled water (Millipore) with DC conductivity 2 μS/cm. This solution was loaded into round fused quartz capillaries, OD/ID = 0.55/0.4 mm (VitroCom, Mt. Lakes, NJ) or Teflon capillaries GA30, OD/ID = 0.62/0.35 mm (Small Parts, Miami Lakes, FL). Filled and empty capillaries were packed into a sample holder, which was either a 4 mm OD NMR tube (in the case of quartz capillaries) or a 5 mm OD NMR tube (for Teflon capillaries) (Wilmad, Buena, NJ) with both ends opened (Fig. 1).Twenty-five quartz capillaries fit into the 4 mm NMR tube, and 26 Teflon capillaries fit into the 5 mm NMR tube. The ratio of filled and empty capillaries was changed to produce different volume fraction of multibore composite sample. Filled and empty capillaries were mixed randomly before

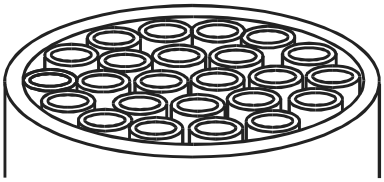

Fig. 1. Multibore sample tube used. Small quartz or Teflon tubes, filled with the sample, packed into a larger quartz tube.

loading into the holder tube; capillaries were taken out of the holder tube and remixed between experiments. Resonant frequency and EPR signal intensity of non-saturated samples were measured at subsaturating incident power $P = 2$ μW. Signal intensity was also measured at half-saturation, as determined for each sample from the power saturation curve [7,19]. Spectra were acquired using 100 kHz field modulation with 0.1 G peak-to-peak modulation amplitude. The quality factor of the empty cavity $Q_U = 2Q_L$ (Q_L is the quality factor of loaded cavity), was determined with the HP8510C network analyzer, $Q_U = 28{,}100$ [2]. All EPR measurements, as well as Q measurements with network analyzer were done at critical coupling, $0.5/Q_L = 1/Q'_U = 1/Q_r$, where Q_r is the radiation quality factor, reflecting the energy lost through the cavity iris. According to the Q factor measurements with the network analyzer, the minimal radiation quality factor $Q_{rmin} = 4200$. This value was used to determine the condition of critical coupling for calculated results, $Q'_U = Q_r$, $Q'_U \geqslant Q_{rmin}$.

3. Methods

3.1. Theoretical

The value of the complex permittivity of water ε was found from the Debye function [20]:

$$\varepsilon(\nu) = \varepsilon(\infty) + (\varepsilon(0) - \varepsilon(\infty))/(1 + i2\pi\nu\tau), \quad (6)$$

for $T = 25$ °C $\varepsilon(0) = 78.36$, $\varepsilon(\infty) = 5.16$, and $\tau = 8.27$ ps, which gives $\varepsilon = 64.26 - i28.87$ at $\nu = 9.4$ GHz. Permittivity of aqueous composite sample was determined from Bruggeman mixing rule [16–18]:

$$\sum_{i=1}^{3} f_i \frac{\varepsilon_i - \varepsilon_{eff}}{\varepsilon_i + (n-1)\varepsilon_{eff}} = 0, \quad (7)$$

where ε_{eff} is permittivity of composite sample, ε_i are permittivities of composite sample components, n is system dimension ($n = 2$ for cylinders in electric field, perpendicular to cylinder axis) and f_i are volume fractions of composite sample components. Air ($\varepsilon = 1$), water and quartz ($\varepsilon = 3.78$) or Teflon ($\varepsilon = 2.2$) (sample tubes material) were treated as components of composite sample. The resonant frequency and distribution of microwave field in the TE_{011} cavity with dewar and cylindrical composite sample were calculated with the radial mode matching (RMM) method, as described in our previous papers [2,21]. The RMM method is based on the solution of Maxwell equations, with boundary conditions determined by the cavity walls, sample geometry, and other objects (e.g., dewar) inside the cavity. Calculated distributions of E and H_1 fields in the cavity were used to calculate the cavity Q and filling factor η (Eqs. (3)–(5)), based on the measured value $Q_U = 28100$, see Section 2.1). The signal intensity for non-saturated and half-saturated aqueous samples was calculated based on Eqs. (1,2):

$$S \propto f\eta Q'_U, \quad (8)$$

for non-saturated sample and

$$S \propto f(\omega\mu_0 V_s Q'_U \eta)^{1/2}, \quad (9)$$

for half-saturated sample, where f is the volume fraction of bulk aqueous sample in a composite. The sample volume fraction in the last two equations reflects the change of number of spins in the composite sample with f.

4. Results

Non-saturated and half-saturated EPR signal intensities were measured for two aqueous multibore composite samples, containing aqueous TEMPO in a bundle of quartz or Teflon capillaries. The outside diameter of each multibore sample was constant in diameter; with volume fraction changed in the range of $0.014 < f < 0.180$ (Teflon tubes in 5 mm NMR tube) and $0.030 < f < 0.274$ (quartz tubes in 4 mm NMR tube), made by variation of filled tubes number. Experiments were repeated 2–4 times. Permittivity of sample was calculated with the Bruggeman mixing rule, Eq. (7). The signal intensity of composite multibore samples, as well as frequency of resonance, were calculated and shown on Figs. 2 and 3, together with experimental data. The data in Figs. 2A, B and 3A, B are normalized to the maximal signal intensity of a single-bore aqueous sample under the same conditions (dewar in the cavity, the same temperature and incident microwave power). Figs. 2A, B and 3A, B show the dependence of EPR signal intensity on aqueous sample volume (or volume fraction) for constant diameter of the multibore sample tube. Figs. 2 and 3 show that the optimal volume fraction depends on the multibore sample tube diameter, this will be discussed below.

To optimize the geometry of a multibore aqueous sample for EPR measurements, signal intensities of non-saturated and half-saturated samples were calculated for the range of sample volume fractions $0.05 < f < 1$ ($f = 1$ corresponds to bulk water) at $T = 25$ °C, and different diameters of the multibore sample, 0.1 mm < ID < 11 mm (11 mm is a diameter of the sample stack of the cavity). Results at critical coupling condition are shown on Fig. 4. Multibore aqueous sample has an optimal diameter to achieve the best sensitivity at no saturation, the same as a single bore aqueous sample [2]. For a half-saturated sample, the larger the sample the better sensitivity (at critical coupling). The calculations show (Fig. 4) that the maximum sensitivity

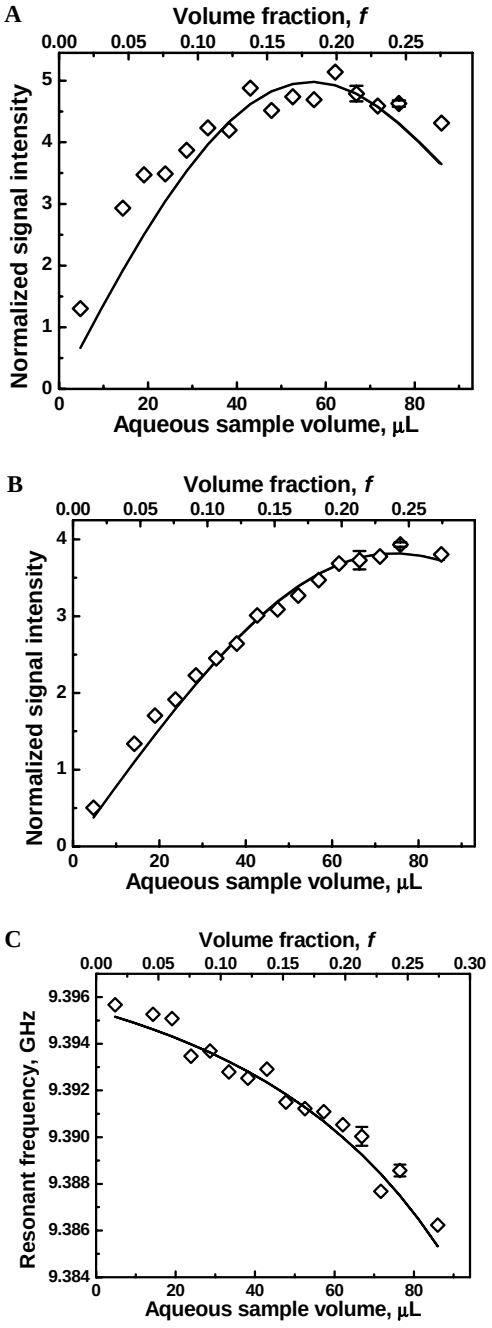

Fig. 2. Signal intensity and resonant frequency dependence on multibore aqueous sample volume at constant sample diameter (ID = 3.2 mm, 4 mm NMR tube). Critical coupling. Aqueous sample in quartz tubes. (A) Non-saturated sample, (B) half-saturated sample, (C) frequency of resonance. Open diamonds: experiment, line: theory. $T = 25$ °C. Signal intensity is normalized on the maximal signal intensity of single bore aqueous sample at the same conditions.

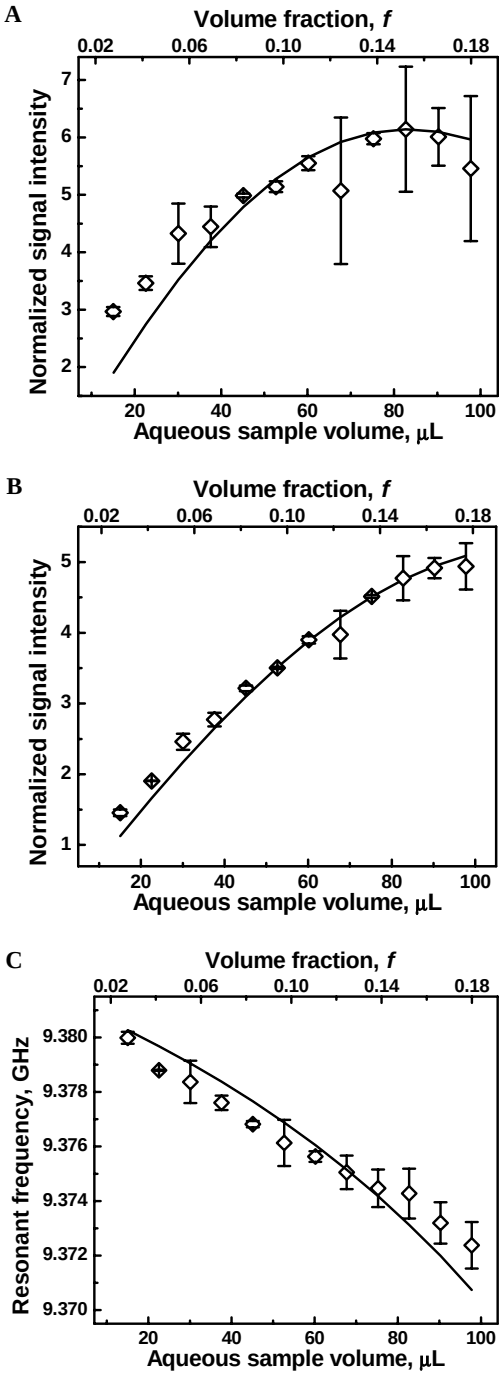

Fig. 3. Signal intensity and resonant frequency dependence on multibore aqueous sample volume at constant sample diameter (ID = 4.2 mm, 5 mm NMR tube). Critical coupling. Aqueous sample in Teflon tubes. (A) Non-saturated sample, (B) half-saturated sample, (C) frequency of resonance. Open diamonds: experiment, line: theory. $T = 25$ °C. Signal intensity is normalized on the maximal signal intensity of single bore aqueous sample at the same conditions.

of EPR measurement can be achieved for an aqueous composite sample with volume fraction of water $f = 0.15$. Increase of signal intensity is 8.7 ± 0.2 for both non-saturated and half-saturated sample, compared with a single bore aqueous sample of optimal size [2]. This sensitivity increase corresponds to the increase of aqueous sample volume in multibore sample. Calculations for Figs. 4–6 were made for a multibore aqueous sample in the cavity without a dewar, but at constant temperature of the sample $T = 25$ °C, to explore the possible range of sample diameters. Effect of temperature and cavity quality factor variation is presented in Figs. 5 and 6.

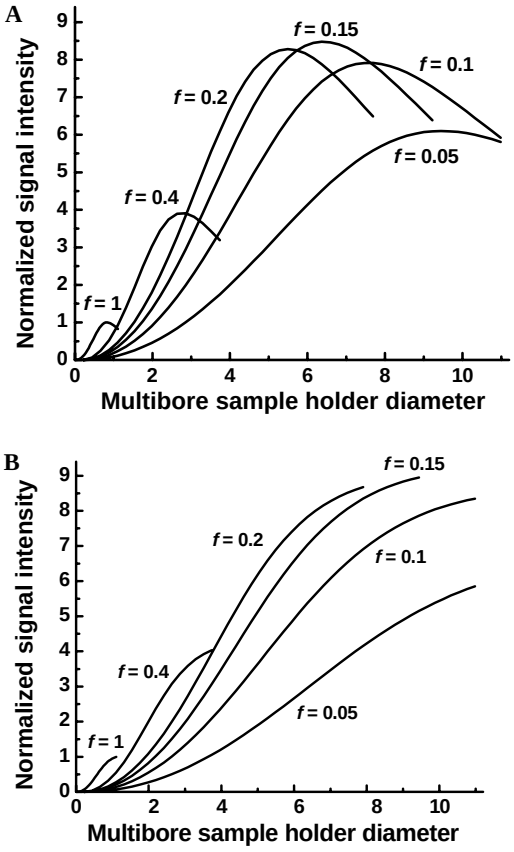

Fig. 4. Calculated signal intensity dependence on multibore aqueous sample diameter for different volume fraction f of water in the sample. (A) Non-saturated sample, (B) half-saturated sample. $T = 25$ °C. Critical coupling. Signal intensity is normalized on the maximal signal intensity of single bore aqueous sample ($f = 1$) at the same conditions (temperature and saturation).

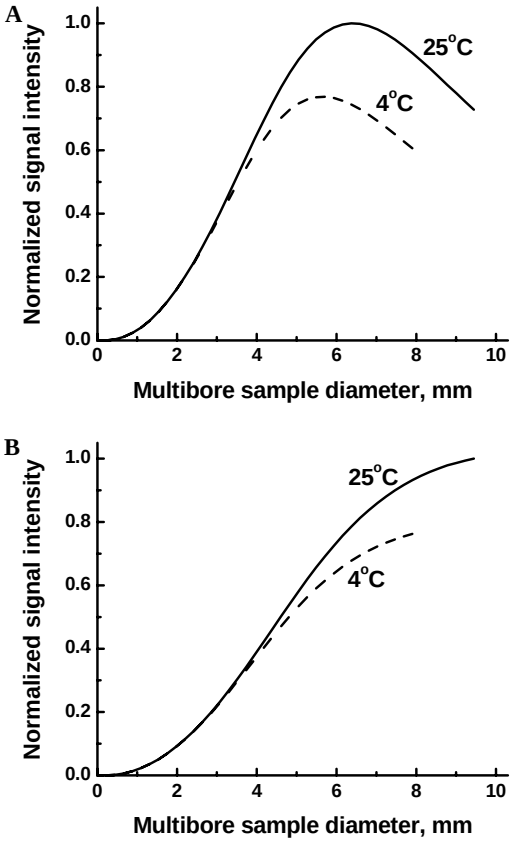

Fig. 5. Calculated effect of aqueous sample temperature on signal intensity of non-saturated (A) and half-saturated (B) multibore aqueous sample. $T = 4$ and 25 °C. Critical coupling. $f = 0.15$.

5. Discussion

5.1. Mixing rules

Available mixing rules are divided into two groups: based on volume averaging, where permittivity of a composite is proportional to volumes of its components, and on the medium field concept, where permittivity is related to the average electromagnetic field in a composite. The first group of mixing rules can be expressed by equation

$$\varepsilon_{\text{eff}}^{\beta} = f\varepsilon_1^{\beta} + (1-f)\varepsilon_2^{\beta}. \tag{10}$$

$\beta = 1$ gives a simple intuitive volume average formula; there are rules with $\beta = 1/2$ or $\beta = 1/3$ [16]. Comparison of our experimental data with predictions of these rules did not give satisfactory result. The Maxwell Garnett rule and the Bruggeman rule form the second group; the Maxwell Garnett rule [22] works best for diluted systems, where the volume fraction of inclusion media is low. It follows immediately from the Clausius–Mossotti relation that the effective permittivity for cylindrical inclusions in the Maxwell Garnett rule can be found using equation [16]:

$$\frac{\varepsilon_{\text{eff}} - \varepsilon}{\varepsilon_{\text{eff}} + \varepsilon} = f_1 \frac{\varepsilon_1 - \varepsilon}{\varepsilon_1 + \varepsilon}, \tag{11}$$

where ε is the permittivity of host media, ε_1 is the permittivity of inclusions, and ε_{eff} is the effective permittivity of composite media.

The Bruggeman rule treats all components of the composite equally, there is no difference between host and guest media, and therefore it works for high volume fractions of inclusion. At low inclusion volume fractions, the Bruggeman rule gives the same result as the Maxwell Garnett rule, at higher volume fractions ($f > 0.03$) only the Bruggeman rule (Eq. (7)) gives the result, which is in agreement with our experimental data.

5.2. How general are the results?

Experiments and calculations of this paper were done for a spherical Bruker SHQ cavity with TE_{011} symmetry of microwave field. The same calculations were performed for a cylindrical cavity, Bruker 4122 SHQE, for the same Q_U; the conclusions remain the same and valid for both types of a cavity. The optimal diameter of a multibore aqueous sample, producing maximum sensitivity for a non-saturated sample depends on a cavity Q_U (Fig. 6). Decrease of a cavity quality factor decreases signal intensity, but does not affect optimal the volume fraction. Tem-

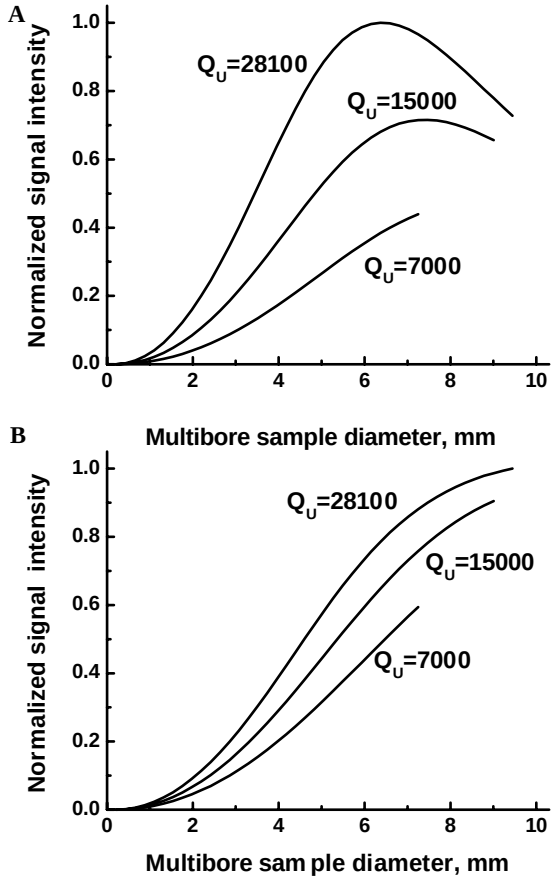

Fig. 6. Calculated effect of cavity Qu on signal intensity of non-saturated (A) and half-saturated (B) multibore aqueous sample. Critical coupling. $f = 0.15$.

perature changes water permittivity dramatically [2] and thus affects signal intensity of multibore aqueous sample. Fig. 5 shows signal intensity of non-saturated sample at $T = 25$ and 4 °C for $f = 0.15$; maximal signal intensity decrease is ~24%, when a multibore aqueous sample cools down from 25 to 4 °C. We found that temperature change does not affect the optimal volume fraction of the multibore aqueous sample.

5.3. Multibore vs. single bore aqueous sample

The pattern of dependence of signal intensity on sample diameter is the same for single bore and multibore aqueous samples. Indeed, the multibore sample configuration affects only the effective permittivity of the sample, remaining all dependencies of signal intensity on EPR parameters the same. The lower permittivity of the composite sample allows increase of aqueous sample volume at critical coupling; signal intensity increases due to increased amount of spins at constant sample concentration.

5.4. Size of a capillary in multibore configuration

The Bruggeman mixing rule was developed for inclusions with the size, less than a wavelength. We have tested capillaries of two sizes and experimental data agreed with the results of calculations. To analyze the dependence of effective permittivity of composite media on inclusion size, more rigorous analysis, based on solution of Maxwell's equations in certain boundary conditions should be done [23]. Important to notice, that Bruggeman rule was developed in mean field approximation that supposes constant field over a sample. In an EPR cavity, the microwave field distribution is not constant and obviously, increase of multibore sample diameter will require decrease of capillary diameter.

Figs. 2A and 3A show that there is a deviation between experimental and calculated results at low volume fractions of water in the sample at non-saturated incident power, suggesting that Q is underestimated by the calculations. However, at high aqueous volume fraction, where the results are most useful to the experimentalist, the calculations are quite accurate.

5.5. Relation to other work

The reported performance of the Bruker AquaX is 4.5 times better than a single-bore aqueous sample. The water volume fraction of AquaX is $f = 0.24$ (18 μL/cm version). At this volume fraction, our calculations predict that the improvement should be a factor of 8. The difference in results is probably due to differences in the permittivity of host media (plastic in AquaX and air/quartz in our case). The performance of an aqueous multibore improves as the permittivity of the host medium decreases. A similar effect was found by Sidabras et al. [14] for a flat cell assembly in a rectangular cavity. Optimized flat cell assembly in a rectangular cavity with finite-element method [14] shows maximal sensitivity of EPR measurement at volume fraction of water $f = 0.25$. The optimal volume fraction of aqueous sample depends on the type of cavity and microwave field distribution; it is not surprising that optimal volume fractions are different for cylindrical and rectangular EPR cavities. It is important to mention that the principle remains the same: distributed aqueous sample produces fewer losses due to decreased permittivity.

6. Conclusions

An aqueous EPR sample, divided into separate volumes (multibore aqueous sample), is a composite sample whose permittivity depends on the permittivities of its components (water, air, material of a sample vessel) and their volume fractions. The permittivity of a composite aqueous sample is less than that of a bulk aqueous sample, and it can be determined accurately with the Bruggeman mixing rule. The decreased permittivity of a composite aqueous sample allows the increase of aqueous sample volume in an EPR cavity without a proportional degradation of Q'_U, thus increasing the EPR signal intensity at constant concentration (Eq. (2)). The optimum value of the volume fraction of water in a composite aqueous sample, produc-

ing a maximum signal intensity at constant concentration, is quite low ($f = 0.15$). This optimal value of the volume fraction does not depend on the cavity Q and is valid for ambient temperatures $T = 4$ and 25 °C. The increase of signal intensity has a factor of 8.7, compared with a single bore aqueous sample at critical coupling, for both non-saturated and half-saturated samples.

Acknowledgments

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References

[1] G. Feher, Sensitivity considerations in microwave paramagnetic resonance absorption techniques, Bell Syst. Tech. J. 36 (1957) 449–484.
[2] Y.E. Nesmelov, A. Gopinath, D.D. Thomas, Aqueous sample in an EPR cavity: sensitivity considerations, J. Magn. Reson. 167 (2004) 138–146.
[3] D.P. Dalal, S.S. Eaton, G.R. Eaton, The effect of lossy solvents on quantitative EPR studies, J. Magn. Reson. 44 (1981) 415–428.
[4] J.S. Hyde, W. Froncisz, Loop gap resonators, in: A.J. Hoff (Ed.), Advanced EPR: Applications in biology and biochemistry, Elsevier, Amsterdam, 1989, pp. 277–306.
[5] L.G. Stoodley, The sensitivity of microwave electron spin resonance spectrometers for use with aqueous solutions, J. Electron. Contr. 14 (1963) 531–546.
[6] C.P. Lin, M.K. Bowman, J.R. Norris, A folded half-wave resonator for ESR spectroscopy, J. Magn. Reson 65 (1985) 369–374.
[7] Y.E. Nesmelov, J.T. Surek, D.D. Thomas, Enhanced EPR sensitivity from a ferroelectric cavity insert, J. Magn. Reson. 153 (1) (2001) 7–14.
[8] J.W. Stoner, G.R. Eaton, S.S. Eaton, Comparison of signal-to-noise for unlimited volume aqueous samples at L-band and X-band, in: 44th Rocky Mountain Conference on Analytical Chemistry, Abstract Book Program Supplement, 2002.
[9] W.D. Nelson et al., Site-directed spin labeling reveals a conformational switch in the phosphorylation domain of smooth muscle myosin, Proc. Natl. Acad Sci. USA 102 (11) (2005) 4000–4005.
[10] J.J. Jiang, Using Bruker's high sensitivity AquaX sample cell, Bruker EPR: Experimental techniques, 7.
[11] G.R. Eaton, S.S. Eaton, Electron paramagnetic resonance sample cell for lossy samples, Anal. Chem. 49 (8) (1977) 1277–1278.
[12] J.S. Hyde, A new principle for aqueous sample cells for EPR, Rev. Sci. Instrum. 43 (1972) 629–631.
[13] R.R. Mett, J.S. Hyde, Aqueous flat cells perpendicular to the electric field for use in electron paramagnetic resonance spectroscopy, J. Magn. Reson. 165 (2003) 137–152.
[14] J.W. Sidabras, R.R. Mett, J.S. Hyde, Aqueous flat-cells perpendicular to the electric field for use in electron paramagnetic resonance spectroscopy, II: design, J. Magn. Reson. 172 (2) (2005) 333–341.
[15] Sidabras JW, Hyde JS, Mett RR. Optimization of close-packed capillary assemblies for EPR spectroscopy of aqueous samples, in: 46th Rocky Mountain Conference on Analytical Chemistry, Abstract Book, Denver, CO, 2004, p. 72.
[16] A. Sihvola, Electromagnetic mixing formulas and applications, in: P.J.B. Clarricoats, E.V. Jull (Eds.), IEE Electromagnetic Waves, vol. 47, The Institution of Electrical Engineers, 1999, p. 284.
[17] R. Landauer, The electrical resistance of binary metallic mixtures, J. Appl. Phys. 23 (7) (1952) 779–784.
[18] D.A.G. Bruggeman, Berechnung verschiedener physikalischer Konstanten von heterogenen substanzen, I. Dielectrizitaetsskonstanten und Lietfaehigkeiten der Mischkoerper aus isotropen substanzen, Annalen der Physik 24 (Ser.5) (1935) 636–664.
[19] D.A. Haas, C. Mailer, B.H. Robinson, Using nitroxide spin labels. How to obtain T1e from continuous wave electron paramagnetic resonance spectra at all rotational rates, Biophys. J. 64 (1993) 594–604.
[20] U. Kaatze, V. Uhlendorf, The dielectric properties of water at microwave frequencies, Z. Phys. Chem. 126 (1981) 151–165.
[21] D. Kaifez, P. Guillon, Dielectric Resonators, Noble, Atlanta, GA, 1998.
[22] J.C. Maxwell Garnett, Colours in metal glasses and metal films, Trans. Royal Soc. (London) CCIII (1904) 385–420.
[23] D.J. Bergman, Calculation of bounds for some average bulk properties of composite materials, Phys. Rev. B 14 (10) (1976) 4304–4312.