Light scattering determination of magnetic moments of magnetotactic bacteria (invited)

Charles Rosenblatt, F. Flavio Torres de Araujo,* and Richard B. Frankel

Francis Bitter National Magnet Laboratory, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139

Light scattering is used to determine the average lengths and magnetic moments of magnetotactic bacteria in culture. The results are consistent with estimates made from electron micrographs.

INTRODUCTION

Magnetotactic bacteria from aquatic sediments orient and swim along magnetic field lines.[1] Each cell contains single magnetic domain Fe_3O_4 particles that impart a magnetic dipole moment to the cell, parallel to the axis of motility.[2,3] The cells migrate in the field direction (North-seeking) or opposite to the field direction (South-seeking) depending on the orientation of the moment in the cell. The vertical component of the geomagnetic field selects the predominant cell polarity in natural environments.[4,5]

In Aquaspirillum magnetotacticum, a freshwater spirillum grown in pure culture[6] the Fe_3O_4 particles are typically cuboidal of 400 - 500 Å dimension.[7] The average number of particles per bacterium can vary from zero to as many as forty depending on the conditions such as the age of the culture, the number of bacteria and the concentration of nutrients and dissolved oxygen The single-magnetic-domain particles are arranged in chains and, because interparticle interactions cause their moments to orient parallel along the chains, the total magnetic moment per bacterium is equal to the sum of the individual particle moments. We report the use of light scattering to determine the average lengths and magnetic moments of bacteria in culture. The results are consistent with estimates of the volume of Fe_3O_4 per bacterium from electron micrographs.

EXPERIMENTAL RESULTS

An electron micrograph of a magnetotactic spirillum is shown in Fig. 1. The electron-opaque particles in the bacterium is the Fe_3O_4 that constitutes its biomagnetic compass. The length of the spirillum is 3.4 μm. The estimated volume of Fe_3O_4 is 9.1×10^{-16} cc, with a remanent magnetic moment of 4.4×10^{-13} emu. Average lengths and magnetic moments were calculated from thirty micrographs of bacteria from each of two cultures S1 and S2; values are given in Table 1.
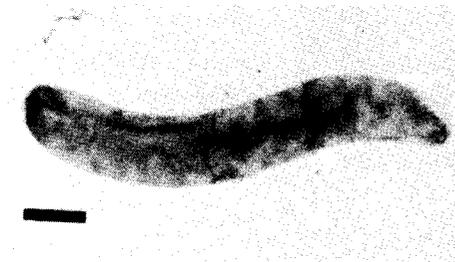

Fig. 1. Electron micrograph of typical bacterium. Dark bar represents 0.5 μm.

Light scattering was used to determine both the average length[8] and magnetic moment of the bacteria in cultures S1 and S2. Since the volume of Fe_3O_4 is only about 1/1000 of the volume of a bacterium, we assume the scattering strength is uniform over the entire bacterium. Thus, modeling the bacteria as optically isotropic cylinders of length L and radius R, one can show that the single particle static structure factor is given by

$$S_{sp}(QL,QR,\vec{\phi}) = \left| \frac{\sin\left(\frac{QL\cos\phi}{2}\right)}{\frac{QL}{2}\cos\phi} \frac{2J_1(QR\sin\phi)}{QR\sin\phi} \right|^2 \quad (1)$$

where J_1 is the Bessel function of order 1 and ϕ is the angle between $\vec{Q}$ and the long axis of the bacterium. $\vec{Q}$ is the momentum transfer of light ($\vec{Q} = \vec{K}_i - \vec{K}_f$). For small diameter bacteria, QR << 1 and the term $2J_1(QR\sin\phi)/(QR\sin\phi) \simeq 1$; thus the corrections caused by the spiral nature of the bacterium are restricted to a small rescaling of L. The assumption of orientational optical isotropy is valid because it was determined that the birefringence of a fully aligned sample in vitro is about 10^{-7}, and thus should cause only insignificant depolarization effects.

If a magnetic field $\vec{H}$ is applied parallel to $\vec{Q}$, bacteria of magnetic moment μ orient parallel to both $\vec{Q}$ and $\vec{H}$ with an angular distribution function $P(\phi,\alpha)$ given by

$$P(\phi,\alpha) = \frac{\alpha e^{\alpha\cos\phi}\sin\phi}{2\sinh\alpha} \quad (2)$$

where $\alpha = \mu H/k_B T$, k_B is Boltzmann's constant, T is temperature, and ϕ is defined as before. The total static structure factor $S(QL,QR,\alpha)$ is therefore the single particle structure factor, weighted by the angular distribution function, integrated over all angles:

$$S(QL,QR,\alpha) = N\int P(\phi,\alpha) S_{sp}(QL,QR,\phi) d\phi \quad (3)$$

where N is the number of scatterers and $\vec{Q}$ is parallel to $\vec{H}$. The scattered light intensity is thus proportional to $S(QL,QR,\alpha)$.

Three Helmholtz coil pairs oriented at right angles to each other were used to cancel the ambient magnetic field to less than 0.01 Oe at the sample position. A fourth Helmholtz coil pair produced magnetic fields from 0 to 40 Oe parallel to $\vec{Q}$ at the sample.

To perform the experiment, the wavevector Q was chosen to be 9898 cm^{-1}, with an acceptance width δQ of 500 cm^{-1} (Fig. 2). The average scattering intensities, measured with a phototube in conjunction with a low pass filter (time constant t = 13.5 sec), were cor-
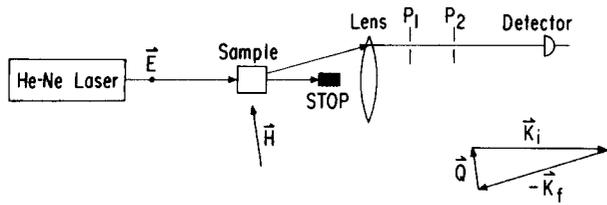
Fig. 2. Schematic representation of scattering experiment (top view). Sample cuvette is 1 cm in length. The lens has a focal length f = 320 mm and is placed 320 mm from the sample. Pinholes P_1 and P_2 specify $\vec{Q}$ and are of diameter d = 1.58 mm, spaced 75 mm apart.

rected for background by first measuring the scattering of water in a cuvette. The water was then removed and the sample was added. The scattering intensities from the bacteria, corrected for background and proportional to $S(QL,QR,\alpha)$, were measured in fields H ranging from zero up to 40 Oe. Since $Q \simeq 10^4$ cm^{-1}, $S(QL,QR,\alpha)$ is virtually independent of R, i.e., $S(QL,QR,\alpha) = S(QL,\alpha)$. $S(QL,\alpha)$ as calculated from Eq. (3), is shown in Fig. 3 for various values of α. As can
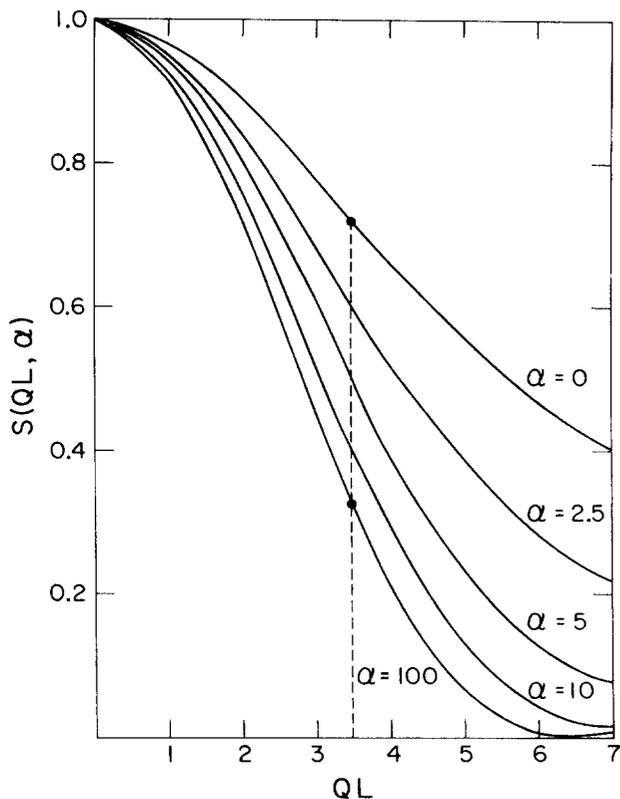

Fig. 3. Structure factor $S(QL,\alpha)$ vs QL for various values of α, where $\alpha = \mu H/k_B T$. The quantity $R(QL,\alpha) \equiv S(QL,\alpha)/S(QL,0)$ for very large α (saturation) was measured to determine QL; dashed line at QL = 3.48 represents one measured culture. Since Q was established by the scattering geometry (Q = 9898 cm^{-1}), a value <L> = 3.52 μm was obtained.

be seen in Fig. 3, if the distribution of lengths is symmetric about some average <L> in the vicinity of 2.5 to 5.5 x 10^{-4} cm, $S(QL,\alpha)$ is independent of both the acceptance width δQ and the distribution width δL as long as $\delta Q/Q$ and $\delta L/<L> \leq 0.5$.

In zero field the alignment is random and in high field (α > 100) it is saturated, with all bacteria aligned parallel to $\vec{H}$. <L> was obtained by measuring $S(OL,\alpha)$ in zero field and in a saturation field of 40 Oe, for which it was later determined that $\alpha \simeq 200$. The quantity $R(QL,\alpha) \equiv S(QL,\alpha)/S(QL,0)$, which rapidly approaches a limiting value for α > 100, is a strong function of QL, as can be seen in Fig. 3 by comparing the curves for α = 0 and α = 100. Thus a measurement of the intensity ratio $R(QL,\alpha)$ for α > 100 accurately determines QL, and since Q is known from the scattering geometry, <L> is determined. In Fig. 3 we show the measured value of OL for culture S2. Values of <L> for S1 and S2 are given in Table I.

Having determined QL, and therefore <L>, using the saturation field, <μ> is obtained by measuring $R(QL,\alpha)$ at many different fields over the range zero to 40 Oe. n Fig. 4, R(QL = 3.48,α), corresponding to the dashed ine in Fig. 3, is shown. For a given field H, the
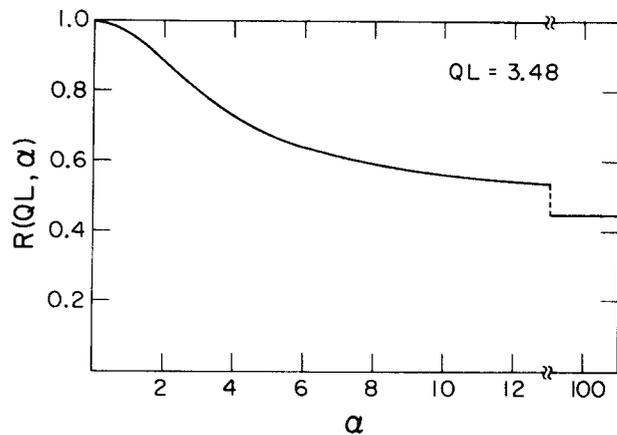

Fig. 4. Theoretical plot of $R(QL,\alpha)$ vs α for a particular value of QL(QL = 3.48) where $R(QL,\alpha) \equiv S(QL,\alpha)/S(QL,0)$. For a given applied field, $R(QL,\alpha)$ was measured, thus establishing α. <μ> = $\alpha k_B T/H$ was obtained.

value of α corresponding to the measured value of $R(QL,\alpha)$ is determined from Fig. 4. Since $\alpha = \mu H/k_B T$, and H and T are known, <μ> is obtained. This measurement was performed for approximately ten values of H for each culture and results were averaged to obtain <μ>. Values of <μ> are listed in Table I. We note in passing, however, that since $R(QL,\alpha)$ is a strong function of α, the distribution width $\delta\mu/<\mu>$ must be less than 0.2 to obtain an accurate measurement of <μ>.

In addition to the light scattering measurement, a bifrefringence technique, similar to that of Scholten,[9] was used to measure <μ>. Here the bacteria are assumed to possess an optical polarizability anisotropy Δp, so that the sample birefringence is

$$\Delta n = N\Delta p <3/2 \cos^2\phi - 1/2> \qquad (4)$$

where N is the number of bacteria and ϕ is the angle between H and the long axis of the bacterium. This technique is insensitive to L and preliminary results for <μ> are consistent with those of the light scattering method. Details will be published elsewhere.

TABLE I. Parameters of Magnetotactic Bacteria Determined by Light Scattering (L.S.) and Electron Microscopy (E.M.)

		Sample 1	Sample 2
Average Length (Microns)	L.S.	2.93	3.52
	E.M.	3.2 ± 0.6	3.4 ± 0.8
Average Magnetic Moment (emu)	L.S.	$(2.2\pm0.2)\times10^{-13}$	$(4.3\pm0.5)\times10^{-13}$
	E.M.	$2.7\pm0.5)\times10^{-13}$	$(5.0\pm0.8)\times10^{-13}$

CONCLUSIONS

Considering the uncertainties in estimating the magnetic moments from the electron micrographs, the agreement of the moments obtained by light scattering with those estimates is quite good. Especially important is the fact that both methods give the same ratio for the average moments in the two samples. Thus static light scattering is a reliable method for rapidly determining the average lengths and magnetic moments of magnetotactic bacteria in culture, and can be used to assay these physical characteristics for bacteria grown under differing culture conditions.

ACKNOWLEDGEMENTS

REFERENCES

*Permanent Address:Department de Fisica, Universidade Federal do Ceara, Caixa Postal, 1262, 60000 Fortaleza, Ceara, Brasil.

1. R.P. Blakemore, Science 190, 377 (1973).
2. R.B. Frankel, R.P.Blakemore and R.S. Wolfe, Science 203, 1355 (1979).
3. R.B. Frankel and R.P.Blakemore, J. Magn. and Mag. Mater. 15-18, 1562 (1980).
4. R.P. Blakemore, R.B. Frankel, and A.J. Kalmijn, Nature 286, 384 (1980).
5. R.B. Frankel, R.P. Blakemore, F.F. Torres de Araujo, D.M.S. Esquivel and J. Danon, Science 212, 1269 (1981).
6. R.P. Blakemore, D. Maratea and R.S. Wolfe, J. Bacteriol. 140, 720 (1979).
7. D.L. Balkwill, D. Maratea, and R.P. Blakemore, J. Bacteriol. 141, 1399 (1980).
8. B. Berne and R. Pecora, Dynamic Light Scattering John Wiley,N.Y.,1976).
9. P.C. Sholten, IEEE Trans. Magn. MAG-11, 1400 (1975).

2729   J. Appl. Phys. Vol. 53, No. 3, March 1982

Magnetism & Magnetic Materials–1981   2729