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Alignable biomimetic membranes

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Abstract

Using neutron diffraction, we have recently shown that lipid bilayers doped with paramagnetic lanthanide ions are highly alignable ($\leq 1.0^\circ$ mosaic) in a magnetic field and give rise to distinct Bragg reflections ($h = 5$) indicative of a smectic mesophase. In addition, using rigid substrates, we have succeeded for the first time in aligning ($\leq 0.5^\circ$ mosaic) membrane systems under conditions of excess water. These two methods of aligning biomolecules will enable a variety of systems to be studied under physiologically relevant conditions. © 1998 Elsevier Science B.V. All rights reserved.

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1. Introduction

When diffraction is used, the structural information is greatly enhanced if the data collected is obtained from an aligned sample [1–3]. Since biologically relevant materials are anisotropic systems, many of their properties are best studied using aligned samples.

Although lipid membrane systems are routinely aligned (e.g., Refs. [1–4]), aligning them under physiological conditions (i.e., physiological pH, ionic strength and excess water) has proven to be a difficult task. Here we present two methods of aligning lipid bilayers under physiologically relevant conditions. The first, is a magnetically alignable system doped with paramagnetic ions (e.g.,

Tm^{3+}) [4,5]. In this system the bilayers are in the physiologically relevant L_α phase while remaining stable over a wide range of temperature [4], pH and ionic strength [6,7]. Most recently, we have also produced mechanically aligned samples under physiologically relevant conditions having the same physical properties as liposomes in contact with water [8]. Due to the relatively complex sample environments required by both samples, neutron diffraction has proven to be an excellent technique in characterizing both systems.

2. Experimental

Details for the preparation of the Tm^{3+} -doped, magnetically alignable dimyristoylphosphatidylcholine (DMPC)/dihexanoylphosphatidylcholine (DHPC)/water system can be found in Ref. [4]. Design of the sample holder and methods for

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preparation of the mechanically aligned samples in the excess water condition are given in Ref. [8].

The experiments were carried out using monoenergetic neutrons having a wavelength of $2.37 \pm 0.005 \text{ \AA}$ selected using the (0 0 2) reflection of a pyrolytic-graphite monochromator (mosaic $\approx 0.4^\circ$) while higher-order neutrons were eliminated using a graphite filter. The wavelength was determined using a powder sample of aluminum.

3. Results and discussion

In the absence of a magnetic field and at a temperature of $315 \pm 1 \text{ K}$ the DMPC/DHPC/Tm³⁺ system forms a poorly aligned (mosaic $\approx 90^\circ$) smectic phase having a repeat distance of 120 \AA and giving rise to only two Bragg maxima (Fig. 1a). However, at a magnetic field of $\approx 1 \text{ T}$ the system began to rapidly align giving rise to the diffraction

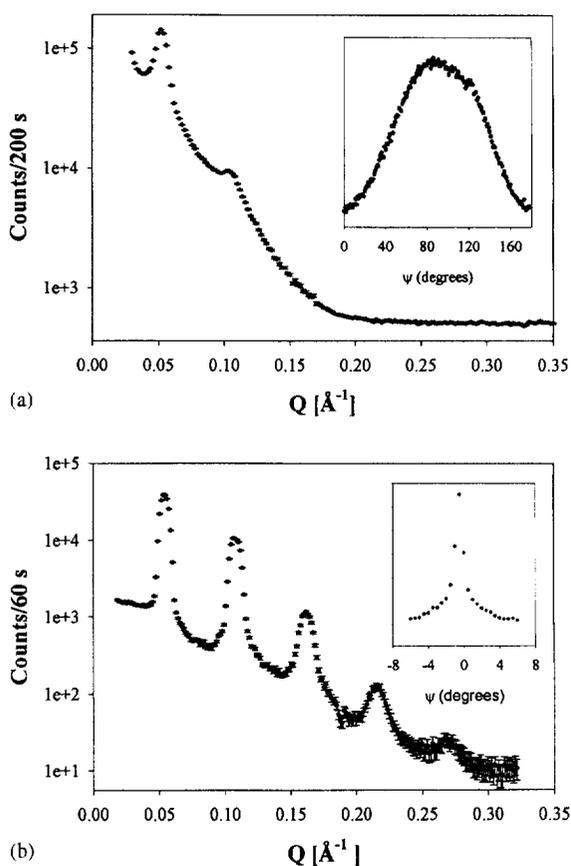


Fig. 1. (a) Q scan at a temperature of $315 \pm 1 \text{ K}$ and no applied magnetic field of the bicelle system containing Tm³⁺ ions. The inset contains the rocking scan of the first order Bragg peak which corresponds to a mosaic of $\approx 90^\circ$. (b) Diffraction pattern from the DMPC/DHPC unilamellar bilayer stack system containing Tm³⁺ at $315 \pm 1 \text{ K}$ and a magnetic field of 2.6 T and exhibiting a mosaic of $\leq 1.0^\circ$ (inset). The diffraction geometry was such that at $2\theta_B = 0$ the incident beam was perpendicular to B .

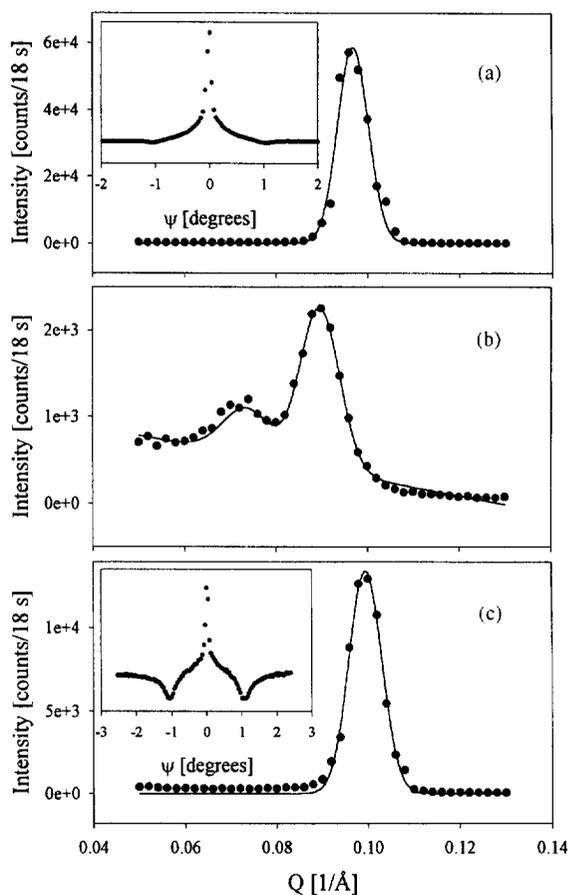


Fig. 2. Q scans of highly aligned ($\leq 0.5^\circ$ mosaic) DPPC multibilayers on a silicon substrate in the (a), L_α phase at 322.2 K having a lamellar repeat spacing of 65.4 \AA , (b), 70.5 \AA multibilayers in the $P_{\beta'}$ phase at a temperature of 313.5 K and (c), L_β multibilayers exhibiting a repeat spacing of 63.5 \AA at a temperature of 292.9 K . The minima seen in the rocking curves (inset) are the result of increased absorption when either the incident or diffracted beam is aligned parallel to the silicon substrate as the sample is rotated.

pattern shown in Fig. 1b ($B = 2.6$ T) with the bilayers having a repeat spacing of 116 Å. It is interesting to note that in the absence of Tm^{3+} but the presence of the magnetic field (2.6 T), the system is characterized by a single broad peak resulting from bilayers lacking positional order and similar to that observed in an analogous lipid/detergent system [9].

Membranes aligned on substrates are hydrated using water vapor at 100% humidity. It is well known that these bilayers have a reduced level of hydration compared to bilayers in contact with water [10]. This so-called “vapor pressure paradox” is widely recognized and results in multi-bilayers having reduced repeat spacings [11], elevated transition temperatures and different mesophases [12].

Fig. 2 shows the first-order Bragg maxima for the three different phases of DPPC obtained using a highly aligned sample (mosaic spread of $\leq 0.5^\circ$) in excess water (Fig. 2, inset). For all three mesophases the repeat spacings and the transition temperatures exhibited by the aligned sample are in agreement with the data found in the literature for DPPC liposomes in excess water [13–17]. In addition, when dealing with samples of limited availability, the specially designed sample holder allows for the buffer's conditions (pH and ionic strength) to be altered without any effect on the sample's alignment.

References

- [1] G.S. Smith, E.B. Sirota, C.R. Safinya, N.A. Clark, *Phys. Rev. Lett.* 60 (1988) 813.
- [2] V.A. Raghunathan, J. Katsaras, *Phys. Rev. Lett.* 74 (1995) 4456.
- [3] J. Katsaras, V.A. Raghunathan, E.J. Dufourc, J. Dufourcq, *Biochemistry* 34 (1995) 4684.
- [4] J. Katsaras, R.L. Donaberger, I.P. Swainson, D.C. Tennant, Z. Tun, R.R. Vold, R.S. Prosser, *Phys. Rev. Lett.* 78 (1997) 899.
- [5] R.S. Prosser, S.A. Hunt, J.A. DiNatale, R.R. Vold, *J. Am. Chem. Soc.* 118 (1996) 269.
- [6] C.R. Sanders, J.P. Schwonek, *Biochemistry* 31 (1992) 8898.
- [7] C.R. Sanders, G.C. Landis, *J. Am. Chem. Soc.* 116 (1994) 6470.
- [8] J. Katsaras, *Biophys. J.* 73 (1997) 2924.
- [9] B.J. Hare, J.H. Prestegard, D.M. Engleman, *Biophys. J.* 69 (1995) 1891.
- [10] G.L. Jendrasiak, J.H. Hasty, *Biochim. Biophys. Acta* 337 (1974) 79.
- [11] J. Torbet, M.H.F. Wilkins, *J. Theoret. Biol.* 62 (1976) 447.
- [12] E. Sackmann, *Physical foundations of the molecular organization and dynamics of membranes*, in: W. Hoppe (Ed.), *Biophysics*, Springer, New York, 1983, p. 425.
- [13] M. Janiak, D.M. Small, G.G. Shipley, *Biochemistry* 15 (1976) 4575.
- [14] M.J. Ruocco, G.G. Shipley, *Biochim. Biophys. Acta* 684 (1982) 59.
- [15] J. Stümpel, H. Eibl, A. Nicksch, *Biochim. Biophys. Acta* 727 (1983) 246.
- [16] R. Zhang, S. Tristram-Nagle, W. Sun, R.L. Headrick, T.C. Irving, R.M. Suter, J.F. Nagle, *Biophys. J.* 70 (1996) 349.
- [17] J.F. Nagle, R. Zhang, S. Tristram-Nagle, W. Sun, H.I. Petrache, R.M. Suter, *Biophys. J.* 70 (1996) 1419.