Title
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Publication Date
1973-06-01

Published in Journal of
Supramolecular Structure
(1973), pp. 281-284

LBL-2058
c.1

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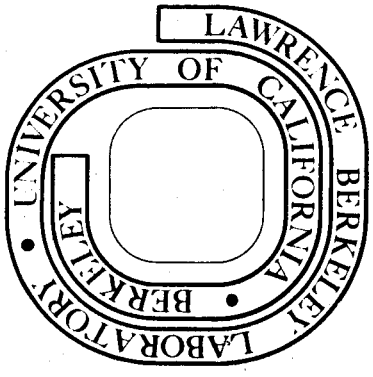
June 1973

Prepared for the U. S. Atomic Energy Commission
under Contract W-7405-ENG-48



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THE FLUID STATE OF LECITHIN BILAYERS

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Current evidence strongly suggests that a substantial fraction of the phospholipids in many membranes are arranged into fluid bilayers (1). Although the word "bilayer" is self-explanatory, the term "fluid" is not. This paper summarizes recent data on the fatty acid configurations, dynamics, and conformations that result in the fluidity of sonicated lecithin bilayers.

In n-alkanes the configurations about each C–C bond are termed trans and gauche isomers. In the gaseous and liquid states of most alkanes the trans isomer is 500–800 cal/mole more stable than either of the two gauche states (2). Therefore, a Boltzmann distribution predicts 10–12 gauche configurations for each pair of 16 carbon n-alkane chains. Several lines of evidence suggest that for sonicated lecithin above its transition temperature this value is 8–10 (3, 6). From measurements of the volume and entropy changes during the endothermic transition of dipalmitoyl-lecithin, and from estimates of the bilayer thickness and surface area per molecule, Träuble and Haynes (3) concluded that there are 3–8 gauche configurations per molecule. Nagle (4) has also treated the calorimetric data from DPL and suggests a value of 7–10 as most reasonable. The laser Raman studies of Lippert and Peticolas (5, 6) provide a more direct estimate. The Raman spectra, which are sensitive to the presence of gauche configurations, of sonicated dipalmitoyl and dioleolyllecithin are similar to those of liquid n-alkanes. For dioleolyllecithin this is true on both sides of the double bond (6). Thus there are at least two gauche configurations above and at least two to three below the double bond, or a total of 8–10 per molecule.

The dynamics of fatty acid segmental motions can be inferred from the nuclear relaxation times of the methylene protons and carbons. The values of T_1 for these resonances are less than an order of magnitude shorter than those for the same molecules in organic solvents (7–9) but are similar to those for neat n-decanol (10). The value of the correlation time estimated from the T_1 data is about an order of magnitude longer than that calculated from an Arrhenius equation with an activation energy of 2.4–3.0 kcal/mole and a preexponential factor of 3.3×10^{12} sec^{-1} (9, 11). These values can be construed to yield an effective Stokes–Einstein viscosity which is within an order of magnitude of that of a liquid.

The activation energy for the T_1 process is ~3.0 kcal/mole (12), a value which is

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similar to the barrier for trans-gauche isomerizations in n-alkanes. T_1 may also contain contributions from rotations of the whole molecule, rotational oscillations within an isomer, and diffusion interactions with neighboring chains. The distribution of proton and carbon T_1 values and activation energies eliminate the first of these as a major source of relaxation (9, 12, 13); similarly, the second can be eliminated by noting that rotational oscillations usually occur at high frequencies, are not random and are thus inefficient in causing nuclear relaxation. The proton T_1 values and their activation energies also contain a component resulting from diffusional interactions between neighboring chains; the available evidence shows that this contribution is less than 50% (14). Thus the fatty acid methylene T_1 values arise in large part from rapid trans-gauche isomerizations, implying therefore that all of the methylene protons are interconverting between their trans and gauche isomers with a correlation time of less than 10^{-9} sec. (15).

Although the fatty acid configurations and configurational dynamics are similar to those of a liquid, the conformations of the fatty acid chains are somewhat ordered. The evidence for this comes from X-ray diffraction measurements which show spacings of ~5 Å (16), the X-ray electron density profiles, the bilayer thickenss and surface area per molecule (3, 17, 18), and the configurational entropy for lipids in a bilayer (3, 30).

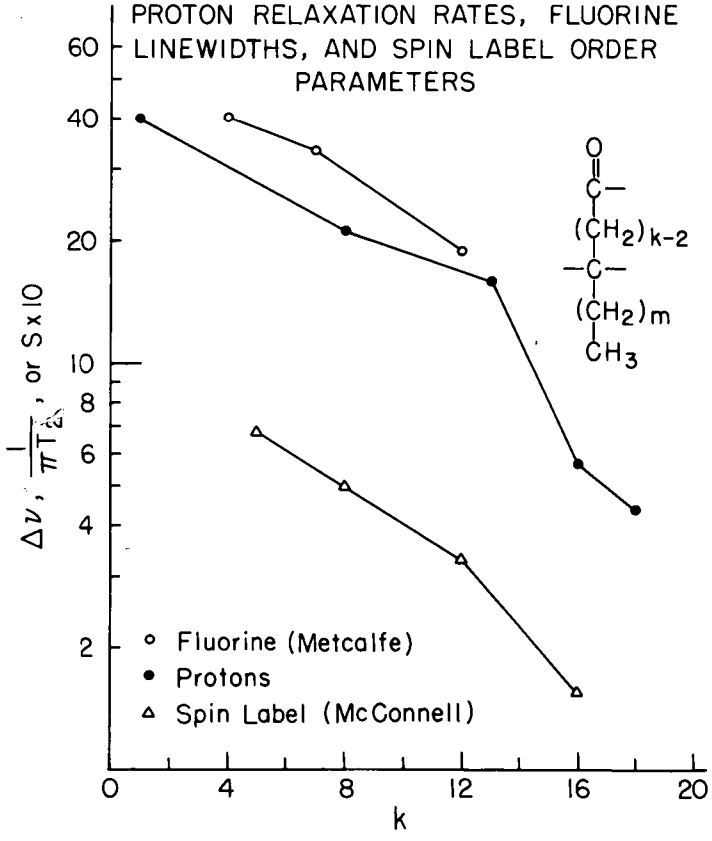

Fig. 1. NMR linewidths, transverse relaxation rates, and ESR order parameters are related entities (15), and, as shown here, they imply similar fatty acid motions. The fluorine data are from Birdsall et al., (27) the proton data from Horwitz et al. (12), and the spin label data from McFarland and McConnell (21).

This order is also reflected in the ESR and NMR data. These measurements show that the motion of most of the methylene protons is highly anisotropic; the motion is fastest and most isotropic at the methyl terminus and the adjacent 2–3 carbons, decreases in rate and isotropy abruptly at this point, and decreases further by only a factor of 2–3 more over the entire length of the fatty acid chain (Fig. 1) (9, 12, 15, 21–23). The data of Chan and Seiter (24) on unsonicated egg yolk lecithin are best fit with a model postulating a component of rapid axial motion and a component of rapid restricted transverse motion (a deviation of 60–70° from the long axis of the fatty acid chain). On the other hand, an unsonicated lecithin from yeast, which is more unsaturated than that from egg, gives NMR spectra which are motionally narrowed (25), i.e., all of the expected intensity is found in a relatively narrow resonance. This shows that the interproton vector on each methylene group moves through the entire solid angle frequently on the NMR time scale. This is also true for sonicated lecithin from egg, which yields narrow NMR lines (15).

We suggest that the fatty acid chains maintain a degree of order by means of β-coupled gauche configurations (3, 9, 12, 15). The ratio of β-coupled configurations to others is a characteristic of each different lecithin bilayer (9). For example, the unsonicated EYL NMR data are consistent with a high proportion of β-coupled configurations (24) while the unsonicated yeast lecithin and sonicated EYL spectra reflect a relatively lower proportion (15, 24). Direct evidence supporting this concept of β-coupled configurations in maintaining straight chains comes from the work of Batchelor et al (26). They have found that the carbon atom β to a cis unsaturate has a relatively high probability of a gauche configuration.

In summary, the fatty acids in some bilayers are best described as configurationally mobile yet relatively ordered (15).

ACKNOWLEDGMENT

This work was supported by the U. S. Atomic Energy Commission.

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