Hydrocarbon chain conformation in the H_{II} phase

Dear Sir:

A recent article by Scherer (1989) on the phospholipid H_{II} phase contains statements indicating a misreading of the literature. In discussing our theory of the L_α–H_{II} phase transition (Gruner, 1985) Scherer states that "An underlying assumption of his (Gruner's) hypothesis is that the CH_2 chains of the lipids around a hydrophilic tube are not interdigitated . . ." Our theory is not predicated on assumptions of the state of interdigitation of the chains. Further, as explicitly indicated in the literature referenced by Scherer (e.g., Tate and Gruner, Fig. 1), we, in fact, expect that interdigitation does occur.

Sherer has apparently failed to grasp the essential physics of what we have termed hydrocarbon chain stress. It arises from the sixfold periodic anisotropy of the chain environment encountered as a function of angle around an H_{II} tube, leading to an energetically expensive variation in the conformation of the lipid chains. Examples of such variations include chains that extend to different mean radial lengths, chains in which the local average director is not purely radial (i.e., sideways deformation of the mean chain volume), and local variations in the hydrocarbon density. If one considers the ways in which chains may be distributed so as to fill the hydrophobic volume of the H_{II} phase, it is apparent that some distributions will reduce the overall relative contributions to the chain stress energy of one or another of the above variations, but typically at the expense of the others. The model presented by Scherer is an extreme case in which the radial chain length variation is reduced. This necessarily results in large variations of the local hydrocarbon density or in sideways deformation of the chains, as may be seen by considering the overlap volumes of adjacent H_{II} tubes in Fig. 4 of Scherer (1989). Contrary to Scherer's claims, interdigitation has not resulted in removal of the hydrocarbon chain stress; rather it has resulted in the substitution of one source of hydrocarbon stress for others. No evidence is presented that would lead one to believe that the overall stress is reduced.

In the absence of a suitable statistical description of the energy associated with different hydrocarbon conformations, one cannot determine which distributions minimize the overall free energy. For this reason we chose to leave the detailed chain conformations unspecified and simply noted that the azimuthal variation in chain environment will cause a rise in the free energy, relative to unconstrained chains, whenever there is variation of the distance from the hydrophilic-hydrophobic boundary—however one chooses to reasonably define it—to the edges of the bounding hexagonal Wigner-Seitz cell. This is the sense in which we mean that there is an energetic cost associated with the hydrocarbon packing. The essential feature of our model is that this energetic cost is a strong component of the free energy in the H_{II} phase but not in the L_α phase.

Explicit calculations require selection of a functional form for the energetic cost of the hydrocarbon packing. When explicit energy calculations have been done we have used forms that increase monotonically with the variation in the radial distance, $|l - l_0|$, from an assumed hydrophilic-hydrophobic boundary to the Wigner-Seitz cell (e.g., Anderson et al., 1988) and have termed this, for the sake of convenience, lipid chain length variation. Perhaps this choice of words was misleading. However, it is important to understand that the distance variation involved is over surfaces which divide unit cells, not over individual lipid chains. Moreover, the essential frustration of curvature vs. packing which we believe drives the L_α–H_{II} transition does not depend on a given explicit form. Rather, as indicated in Anderson et al. (1988), the picture is expected to be qualitatively unaltered for a variety of functional forms in which the chain free energy increases with $|l - l_0|$.

Scherer also states that ". . . the addition of small amounts of tetradecane to DOPE in the H_{II} phase (Gruner et al., 1986) has little effect on the observed dimensions at any level of hydration, which casts doubt on the importance of the presumed stress in controlling the structure." We have never claimed that the hydrocarbon stress is an important factor controlling the *size* of the H_{II} tubes. The spontaneous curvature and the amount of water available control the tube diameter. The frustration between chain stress and spontaneous curvature dominate the onset temperature of the L_α–H_{II} phase transition, as evidenced by the dramatic reduction in the phase transition temperature observed upon the addition of a few weight per cent of dodecane.

Finally, Scherer says that "The reduced strain hypothesis has been invoked to explain observed increases in tube radii from incorporation of relatively longer chain PC lipids . . ." (Tate and Gruner, 1987). Not so. The reduced strain of the longer chains was invoked to explain the change in the L_α–H_{II} transition temperature. Most of the change in the size of the H_{II} tubes was explained by chain length dependent changes in the spontaneous curvature. The essential point of these experiments was to emphasize the distinction between the spontaneous curvature and the energetics of packing the chains.

In closing I note that although there may be virtue in redefining the position of the polar–nonpolar interface, the advantages are not apparent from Scherer's article. Most of the paper is concerned with geometrical manipulations which follow tautologically. Two apparent conclusions of the paper are encompassed in Eq. 14 and in the prediction of a central vacuum void. Eq. 14 of Scherer (1989) follows from the geometry and the observation that the interstitial distance (Fig. 4 *b*, Scherer, 1989) of the fully hydrated H_{II} phase is about the same length as the distance between the hydrophilic-hydrophobic and bilayer midplane surfaces in the L_α phase; it does not depend on the exact definition of the polar–nonpolar interface. (The similarities between these distances in the H_{II} and L_α phases were noted in Table II of Kirk et al., 1984.) The prediction that there is a 5-Å central vacuum void in the anhydrous H_{II} phase follows from the assumption that the headgroups pack without radial disorder. This prediction is certainly provocative but is without experimental support. Microscopic vacuum voids are generally entropically expensive. No justification was provided as to why

the headgroups are not expected to exhibit radial disorder. In the end one must ask as to what experimentally observed data has been explained in a new and more insightful way?

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