Spin Label Saturation Transfer ESR Study of the Purple Membrane of Halobacterium halobium.

M. Renard and M. Delmelle

Institut de Physique, Université de Liège
4000 Sart-Tilman, Liège 1, Belgium

Bacteriorhodopsin is an immobile (1) membrane protein which is embedded within the purple membrane (PM) of Halobacterium halobium. The molecular dynamic of the lipid phase is accordingly severely restricted as shown with spin labels and the conventional electron spin resonance (ESR) technique(2).The saturation transfer method (ST-ESR) complements the conventional ESR spectroscopy since correlation times (τ) ranging from 10^{-3} to 10^{-7} sec become observable (3).
We used ESR and ST-ESR in order to monitor within the -15° to 60°C range, the motion of a stearic acid spin label (5SASL) embedded in the P.M.
The conventional technique allows only to draw one definite conclusion: below 60°C, the probe motion is in a slow regime ($\tau >> 10^{-9}$ sec).
The simplified methods which have been proposed to estimate the correlation times in the 10^{-7} to 10^{-9} sec range, give a value of 10^{-8} sec at around 40°C. These methods assume however an isotropic motion which is obviously unrealistic here.
The St-ESR spectra are strongly temperature dependent between -15 and 30°C. Above 30°C, they become progressively less temperature sensitive and the motion ceases to be observable on the ST-ESR time scale.
The C'/C parameter decreases from 0.1 at -15°C to -0.7 at 30°C. These values seem to demonstrate the presence of a permanent axial rotation of the probe at a rate which is increasing with temperature and which amounts to 10^{6} sec^{-1} around 0 °C. The L'/L and H'/H data indicate that the wobbling motion is slower than the axial rotation.
In the whole temperature range investigated with the two techniques, no evidence was found in favor of a phase transition.

1. Razi Naqvi, K., Gonzales-Rodrigues, J., Cherry, R.J. and Chapman, D. (1973) Nature New Biol. 245, 249 - 251.
2. Chignell, C.F. and Chignell, D.A. (1975) Biochem. Biophys. Res. Comm. 62, 136 - 143.
3. Thomas, D.D., Dalton, L.R. and Hyde, J.S. (1976) J. Chem. Phys. 65, 3006 - 3024.