Themal motions and function in bacteriorhodopsin in purple membranes, effects of temperature and hydration studied by neutron scattering by Ferrand, Dianou, Zaccai, Petry, PNAS 90, 9668 (1993)

communicated by Hans Frauenfelder

Abstract:

harmonically. The ability of the protein to functionally relax and complete the photocycle initiated by the absorption of a photon, however, is strongly correlated with the onset of low-frequency, large-amplitude anharmonic atomic motions in the membrane. For a normally hydrated sample, this occurs at about 230 K, where a dynamical transition from a low-temperature harmonic regime is observed. In moderately dry samples, on the other hand, in which the photocycle is slowed down by several orders of magnitude, no transition is observed and protein motions remain approximately harmonic up to room temperature. These results support the hypothesis, made from previous neutron diffraction studies, that the "softness" of the membrane modulates the function of bacteriorhodopsin by allowing or not allowing large-amplitude motions in the protein.

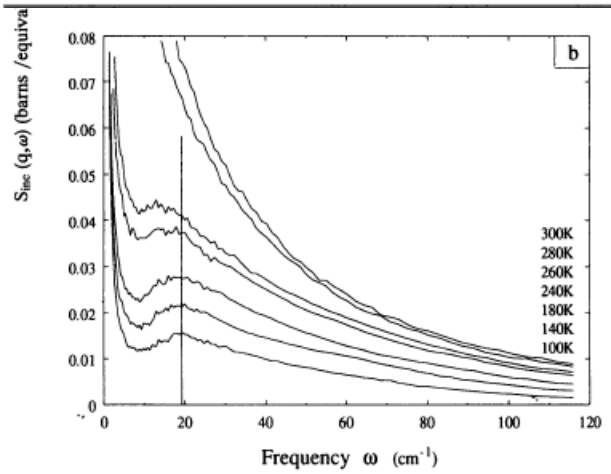


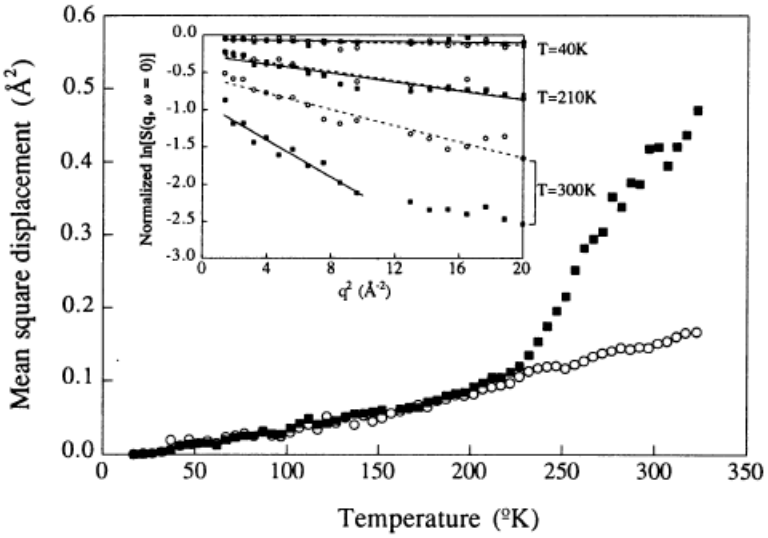
FIG. 2. Temperature dependence of the IINS observed on the IN6 time-of-flight spectrometer (energy resolution ≈ 100 μeV). Data shown are for q (elastic) = 1.76 Å^{-1}. Data were normalized by the scattering of a standard vanadium sample and by the mass of BR in each sample. "Dry" (*a*) and "wet" (*b*) samples are represented (see caption of Fig. 1).

Comment by Wolfgang Doster at bioneutron.de

It is important to note, that this questionable PNAS neutron scattering publication was communicated by Hans Frauenfelder.

This was the first dynamics paper of the small angle scientist G. Zaccai. The idea was to copy our Nature 89 paper (Doster et al. Nature 89) on a low level, but with improved biological relevance. Instead of hydrated myoglobin, they used the membrane protein bacteriorhodopsin (BR), which complicates the interpretation of the spectrum. In addition to protein, also the

membrane would contribute to the spectrum. In addition the authors had massive problems with the sample preparation, they were not aware of the crucial role of hydration at low temperature. Instead of what is said in the Abstract, the authors recorded the melting of bulk ice in the presence of BR and not a protein dynamical transition. There was too much bulk water in the sample, which is easily detected by the discontinuity of the spectra in fig. 2 at 273 K. When I saw the preprint, I explained this problem to the authors, which was ignored. This paper is still cited by Zaccai and his supporters as the seminal publication of the field.



This figure compares displacements of dry and hydrated PM including the elastic scattering function in the insert. In contrast to our paper (Nature 1989) they use qualitative straight lines to approximate the assumed Gaussian scattering function. Thus they miss the methyl group transition around 160-180 K, which is observed even in the dry state (Doster et al.Nature 1989) There is a strange kink around 260-270 K related to melting of bulk ice. The formation of ice involves freeze concentration of the sample, resulting in drastic changes of pH, hydration, ionic strength, which could lead to protein denaturation. All these complications were not considered by the authors. This was only the first of many highly questionable publications of the Zaccai group.