Dynamics of a photo-excited antenna protein

Phycocyanin (PC), a blue protein present in the light-harvesting system of cyanobacteria, plays a key role in the first steps of photosynthesis. For the isolated PC, one part of captured light energy is dissipated *via* emission of fluorescence and the other part is dissipated by fast and localized dynamics of the pigment-protein complex. The aim of this project is to investigate whether dissipation of excitation energy in PC leads to modifications of the protein internal dynamics on longer timescales and larger amplitudes than that of localized vibrations of the pigments.

We measured photo-induced dynamics of PC on MIBEMOL time-of-flight spectrometer (LLB) with a pulsed Nd:YAG laser (λ = 532 nm, 5 ns pulses) at different energy and frequency values (20 mJ and 14 Hz max.) to illuminate *via* optic fibres both sides of the sample (130 g/L of hydrogenated PC solubilized in 20 mM Na_2DPO_4). MIBEMOL data acquisition system has been successfully modified to synchronize the laser excitation flashes with the neutron pulses at sample position and get "double beam" relative measurements ("light" and "dark"). This "double beam" procedure is extremely novel and eliminates spurious effects that could occur in the sample during the experiment. We used an aluminium sample holder surrounding sapphire glasses, which exhibit a much lower neutron scattering than quartz glasses. In a preliminary experiment, a difference of 3% between PC illuminated and PC in the dark has been observed in the maximum of elastic scattering peak, with the "low energy" mode of the laser (pulses ~ 110 µs similar to the duration of the neutron pulses ~ 87 µs). Further experiments will take place very soon to improve this device (laser wavelength closer to the maximum absorption (620 nm) of PC, integrating sphere to illuminate the sample inside uniformly) and also to get reasonable statistics.

[Collaboration: S. Combet, J.-M. Zanotti, M.-C. Bellissent-Funel, LLB; J. Pieper, TU-Berlin].
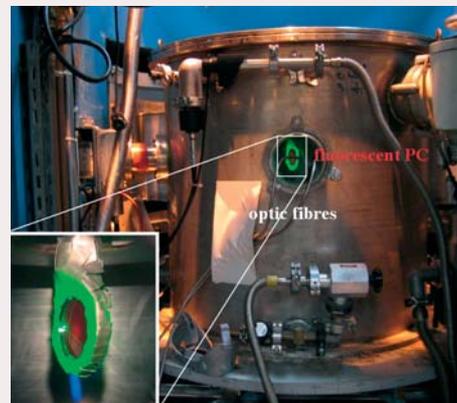
Side view of the sample environment on MIBEMOL time-of-flight spectrometer during illumination experiments on PC. Insert: fluorescence of the PC (red color) in the sample holder with the green laser beam reflected on the cadmium mask.

Glass transitions in cryoconcentrated sucrose solutions.

Since it drives the rate of diffusion limited bio-chemical reactions, the glass transition temperature is a key parameter for frozen food conservation. DSC signal of a maximally cryo-concentrated sucrose solution (50% sucrose mass fraction), considered here as a model system, shows a two steps baseline shift (Figure 1a).
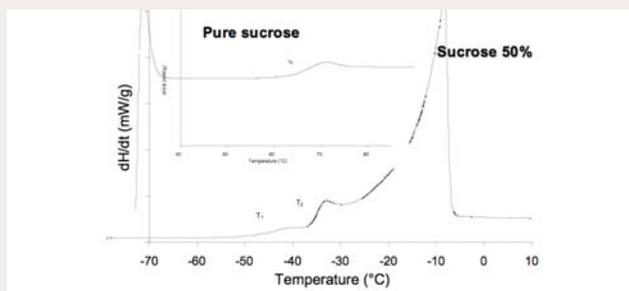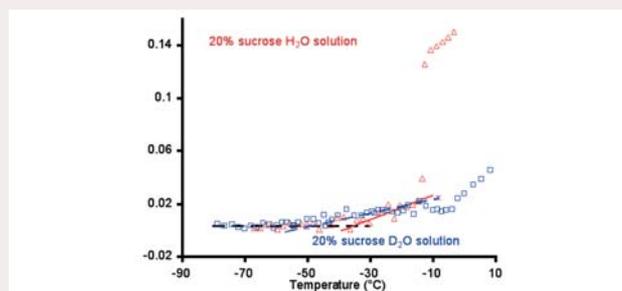
Figure 1 a): Comparison of the DSC results between a pure glassy sucrose and the cryoconcentrated sucrose solution.

b): Temperature dependence of the mean square displacements for 20% sucrose solution in H_2O and D_2O.

To identify the onset temperature of water mobility in a 20% sucrose solution, H/D isotopic labelling has been used to directly probe molecular motions of water (H_2O+sucrose) then sucrose (D_2O+sucrose). The mean square displacement ($<u^2>$) as measured on Mibémol shows a small slope change at -37°C and -48°C in 20% sucrose/H_2O and sucrose /D_2O mixtures respectively. These results are in fully agreement with DSC observations: the first transition at -48°C is to be correlated to a dynamic change of the sucrose molecule whereas the other one seems to be linked to a change of water dynamic. The sharp evolution of $<u^2>$ seen at higher temperature (around -10°C) is due to ice melting, which acts like the dilution of the liquid phase. In order to study if the water dynamical change around -35°C affects the ice structure during its formation, diffraction studies were also carried out with MIBEMOL. The Bragg peak intensity were analysed as a function temperature during cooling. The crystallization of heavy water started at temperatures around -7°C. This temperature is around the same as the temperature of $<u^2>$ evolution variation (Figure 1b). The intensity of both Q=1.61 Å^{-1} and Q=1.71 Å^{-1} peaks were followed during temperature sweep. The observed change in the evolution of the Q=1.71 Å^{-1} peak intensity at the temperature around T_2 on DSC (Figure 1a) may be the consequence of cubic ice formation. Indeed, confined water beyond the interfacial region created by the high cryo-concentration of sucrose may crystallize into a distorted form of cubic ice in contrast to bulk water which crystallizes into ordinary hexagonal ice.

[Collaboration : Champion D., Loupiac C., Simatos D., ENSBANA, Dijon and Zanotti JM (LLB]