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Report:

Temperature dependent phonon spectra of iron in myoglobin
using inelastic X-ray scattering of synchrotrons radiation

A recently developed method for determining the phonon spectra of samples containing a Mössbauer isotope /1,2/ was used to measure selectively the modes of motion which couple to the iron atom in myoglobin, a protein of 153 amino acids and with an iron atom in the active centre. The method is established at the beamline BL 11 using 14.4 keV X-rays from an undulator with 23 m period. With a system of a double Si(111) monochromator and a 'nested Si(422)/Si(975) monochromator one yields radiation of width 4.4 meV tuneable in the range of ± 100 meV. Using the 16 bunch mode of the synchrotrons, there is a X-ray flash of ~100 ps duration every 176 ns. This radiation is scattered by the sample and detected in a time resolving avalanche photo-diode (APD) which covers a solid angle of 2.5* 1.9 sterad. One part of the radiation is promptly Rayleigh scattered by the electrons of the sample,

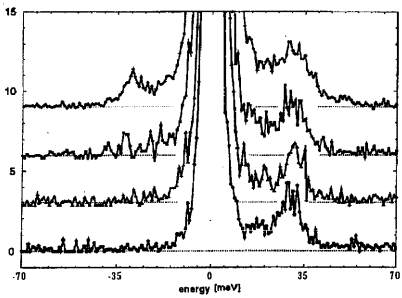

Fig. 1 Phonon spectra of iron in myoglobin. T=73, 124, 189 and 295 K from bottom to top.

whereas a second part is resonantly Mossbauer absorbed by the “Fe in the sample and re-emitted with a characteristic time of τ = 141 ns. The delayed intensity for a given incoming energy was integrated from 15 to 150 ns to obtain one intensity value in the phonon spectra as a function of the energy difference to the Mossbauer level (see Fig. 1). The sample was polycristalline deoxymyoglobin contained in a vacuum tight plastic holder with mylar windows. The natural iron in the heme plane of the molecule was exchanged by ^{57}Fe. The sample was mounted in a closed cycle refrigerator and measured at temperatures T = 73, 124, 189 and 295 K. The hydration degree of the sample (-40% for a myoglobin crystal) is identical to that of the amorphous sample of a preceding experiment where we measured the inelastic X-ray scattering at the electrons of the sample /3/. This scattering is mainly sensitive to the modes of motion coupling to the N-,C-,O-atoms of the protein, whereas we now measured selectively the modes coupling to the iron. This results in remarkably different phonon spectra with a narrower central line and a distinct maximum at -30 meV close to the energy of the iron-histidine vibration /4/. An additional inelastic X-ray scattering experiment on the polycristalline sample shows that the phonon spectra of the N-,C-,O-atoms is identical for both samples within the experimental errors /5/.

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