ESRF	**Experiment title:** Drugs-hemoglobin interaction studied by XANES	**Experiment number:** LS-1258 08-01-167
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Report: Nifedipine is a dihydropiridinic calcium channel blocker known to be a photolabile compound, that under exposure to UV-Vis light, is changed into a nitroso compound that seems to have spin trapping properties. We studied, at iron k edge, pH 7.2 and in presence of 10 mM nifedipine light-activated or not, the effect of the drug on the active site of human hemoglobin bound to different ligands: an oxygen molecule, water (aquomet form) and carbonmonoxyde. The effect of nifedipine, as it results comparing a single spectra, is shown in fig.1 on acquomet derivative. It results to be, in general, quite small but meaningful as it takes place on the rising edge and in spectral regions where falls the features of the ligand. However, at this stage of the analysis (that is still going on) a direct interaction between nifedipine and heme seems unlikely, whereas a possible interaction between the drug and aminoacids outside the heme pocket can't be excluded.

A protein damage due to production of radical by high flux of X-ray radiation has been reported in the past during many experiments. In our experiment we found evidence, in some cases, of sample's modifications due to prolungated exposure and an effect of nifedipine on this phenomenon has been carried out. In fig.2 we show the spectra of the oxy

derivative (without nifedipine) in which, at increasing exposure, a progressive release of oxygen results. A similar effect takes place also in presence of nifedipine, whereas in the case of activated nifedipine a strong dumping of the rate of oxygen release has been found. In fig.3 we report (upper part nifedipine-free sample, lower part hemoglobin plus activated nifedipine), for each sample, the difference spectra between the first acquired scan and the ones collected in the follow. This data show that a comparable amount of oxygen release is reached only after a 4 times longer exposure in presence of activated nifedipine. This "protective" effect of nifedipine could be interpreted as a trapping of the radical produced by X-rays that are responsible for the oxygen displacement in absence of the drug.

We further measured model compounds for the interaction of nifedipine with heme. They present different iron's oxydation state and different kind of ligands. Two of them, with large difference in the spectral features due to different iron oxydation state, are reported in fig.4 and consists in heme with two piridine occupying the fifth and sixth iron coordination.

FIG. 1

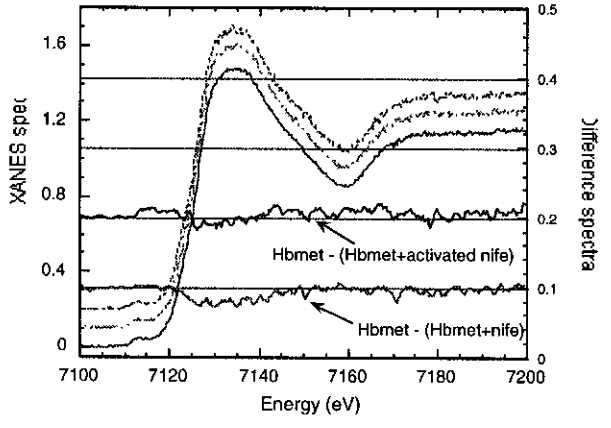

FIG. 2

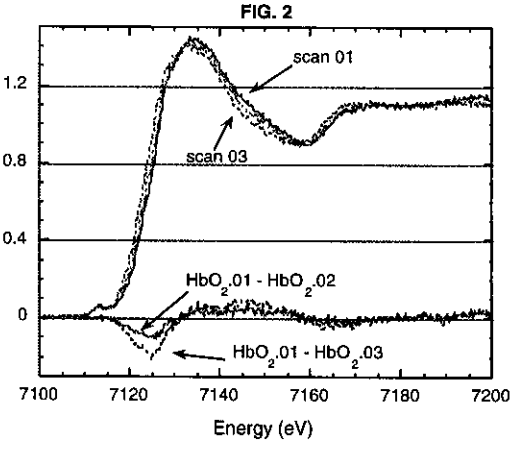

FIG. 3

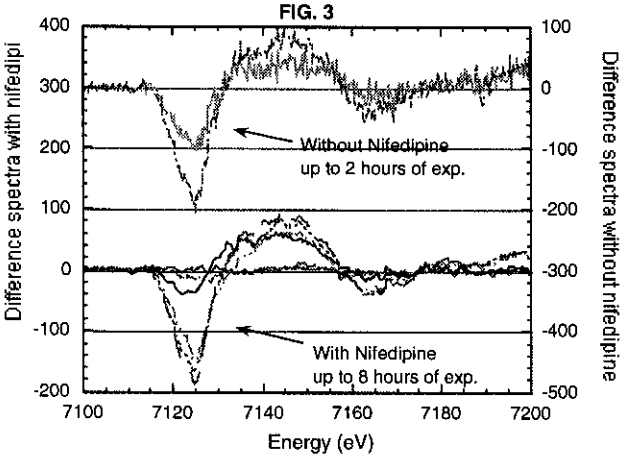

FIG. 4