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Concluding remarks

We organized the results of this thesis in three important parts. In the first one (Chapter 2), we studied the interaction solute-solvent of heme and PPIX in water. The structure and fluctuations of the solvation layers around porphyrins were determined. We found time-averaged local densities ranging between 0.6-4.0ρ_w. In some regions surrounding the solutes, local densities were found to take values several times the bulky ones, reflecting local immobilization of water molecules rather than tighter packing. The introduction of iron Fe^{2+} in PPIX changes the local organization of the surrounding water. There is a tendency that the center of the porphyrin ring of PPIX acts as a weak acceptor of water hydrogens. In the case of the heme, the first layer has a preferential axial occupation, moreover, the spatial distribution functions show that the interstitial C-C bond regions that form the porphyrin ring are penetrated by water. An analysis of the potential of mean force shows that the free energy of the water molecule interacting axially with the iron is approx. -1 kcal/mol.

In the second part (Chapter 3), we sampled the structures of HSA available in the Protein Data Bank to explore the conformations that HSA assumes in the presence of different ligands and under different experimental conditions. Through a clustering analysis, only two well defined structures were detected. They represent the apo and the HSA-myristate conformations, with a structural distance of 4.6 Å between them. One of the conclusions is that ligands like metal-porphyrins do not induce a conformation different from the HSA-myristate.

By keeping the domain II fixed, the conformational change X1 $\rightarrow$ X2 can be characterized by a twist motion with an angle of rotation of 24 degrees between domains I and II and a hinge motion with an angle of 15 degrees between domains III and II. The twist might have its origin in a reorganization of H-bonds around TYR-152, and residues LYS-195–GLN-196 act as bending residues.

On the other hand, the first principal component of the HSA-myristate represents a collective anticorrelated motion between the C-terminal and N-

terminal regions, motion which is not present in the apo conformation.

The HSA/heme interaction is approached in the last part of the thesis (Chapter 4, and also App. D). When the heme enters the hydrophobic pocket in HSA, it suffers a dramatical desolvation, conserving about two water molecules in the distal position. In this environment, the heme strongly interacts with the residues LEU-115, HIS-146, LYS-190, ARG-145, ARG-114, ARG-186 helping to fix the ligand in the hydrophobic pocket via either H-bonds or salt bridges representing molecular latches. The dynamics reveals that the heme induces anti-correlated fluctuations between domains IB and IIIB, consistently with the dominant mode observed in the HSA-myristate conformation.

The present thesis gives details about the mechanisms that rule the interaction HSA-heme and can be useful on future research to turn HSA an oxygen carrier.