Estimate the Time of Soak Simulating Small Molecule Diffusion in Protein Crystal

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For a biocrystallographer, incubating a protein crystal in a solution containing a diffusive compound is a recurring operation. The soaking technique is widely used for preparation of heavy-atom derivatives, cryoprotection of crystals and introduction of inhibitors or substrates in crystal structure of enzymes. However, very few experimental values of the diffusion times in protein crystals are reported in literature. The methods used to detect diffusion of substrates are mainly: X-ray diffraction (monitoring changes in X-ray intensities for some selected reflections as substrates are diffused into the crystal); birefringence of the crystal; and spectroscopy (monitoring specific spectroscopic properties of substrates). Due to experimental difficulties, a theoretical approach by simulations of the diffusion process of small molecules inside protein crystals are particularly important to plane and to understand soaking experiments. Starting from the Fick's second law, a simplified approach to the diffusion is proposed. The reliability of the simulation model has been validated on the basis of experimental data found in literature. These experiments cover a wide range of situations: from small to relatively large diffusing molecules; crystals with quite different size and with low or medium protein density, and more important, with diffusion times from few seconds to several hours.

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