

# Supplementary Online Material

## S<sub>1</sub>-State Model of the O<sub>2</sub>-Evolving Complex of Photosystem II

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## Methods and Models

### QM/MM model and structure optimization

The QM/MM model was built using the coordinates from the new crystal structure at the resolution of 1.9 Å [1]. All water molecules and chloride atoms within 15 Å distance as well as all amino-acid residues with the  $\alpha$ -carbon within 15 Å distance from the OEC of chain A were included and hydrogen atoms were added where appropriate. The amino acid residues were protonated in accordance with physiological pH. The QM layer includes the inorganic core of the OEC, the carboxylate groups and corresponding  $\alpha$ -carbons of the ligands D170, A344, E189, E354, E333, and D342, the imidazole rings of H332 and H337, the guanidium group of R357 and several water molecules. The structure optimization was performed using the ONIOM (our own N-layered integrated molecular orbital plus molecular mechanics) method [2] with the link-hydrogen atom scheme as implemented in GAUSSIAN 09 [3]. Unrestricted density functional theory with the BP86 [4, 5] or B3LYP [6, 7] density functionals and the LACVP\* basis set as implemented in the JAGUAR 7.7 [8] program were employed for the QM part. The initial-guess wavefunction for the QM part was generated with the JAGUAR 7.7 program using the same density functional and basis set as in the QM/MM calculation. The MM part was calculated using the AMBER force field [9]. The  $\alpha$ -carbons (except of the QM layer) were fixed during the optimization.

### Calculation of EXAFS spectra

The EXAFS spectra were calculated using the programs FEFF 8.30 and IFEFFIT v.1.2.11d in accordance to previous work [10–15]. For all calculations involving the DFT-QM/MM model, the atoms of the QM part model were taken. In general, all paths with less than nine scattering legs were considered and the negligibly small contribution of hydrogen atoms was not considered. In addition, we took into account all paths with mean amplitudes exceeding 4% and 2.5% of the largest path in a full curved wave and plane-wave calculation to the oscillatory part of the dipole transition matrix element (or EXAFS data)  $\chi$ , respectively. A value of 0.002 Å<sup>2</sup> for the Debye–Waller factors was employed in all calculations except for the isotropic spectra for the S1 models where the Debye–Waller factors were obtained by a least-square fit of the calculated data to the corresponding experimental one in reduced distance ( $R$ ) space and all paths were

considered. The energy ( $E$ ) axis was converted into photoelectron wave vector ( $k$ ) space by using the transformation  $k = \sqrt{(2m_e/(h/2\pi)^2)(E - E_0)}$  where  $m_e$  is the mass of the electron and  $h$  is Planck's constant. A value of  $E_0 = 6543.3$  eV for the Fermi energy has been employed for the calculations involving the QM/MM model and  $E_0 = 6568$  eV (for details, see below) for the OEC and surrounding ligands of chain A of the 3ARC crystal structure. A fractional cosine-square (Hanning) window with  $\Delta k = 1$  was applied to the  $k^3$ -weighted EXAFS data. The grid of  $k$  points, which are equally spaced at  $0.05 \text{ \AA}^{-1}$  was then used for the Fourier transformation (FT) to  $R$  space. A  $k$  range of  $3.5 - 11.5 \text{ \AA}^{-1}$  and  $1.8 - 11.8 \text{ \AA}^{-1}$  was taken for the FT for the polarized and isotropic EXAFS data, respectively. The FT magnitude and EXAFS  $\chi k^3$  values were appropriately scaled to match the experimental data. The polarized EXAFS calculations were computed with polarizations of the electric field of the X-ray beam parallel to the crystal axes  $a$ ,  $b$ , and  $c$ , respectively. The eight monomers which have to be considered for the polarized EXAFS calculations, were obtained by applying the local  $C_2$  axis rotation and the  $P2_12_12_1$  space group transformations derived from the 3ARC PSII crystal structure with the resolution of  $1.9 \text{ \AA}$ .

### **Fit of experimental EXAFS spectra to the spectrum of the $1.9 \text{ \AA}$ crystal structure**

In order to determine whether the 3ARC crystal structure corresponds to a mixture of S-state intermediates, along the catalytic cycle, in the absence of radiation damage, we performed a least-square fit of a linear combination of isotropic EXAFS spectra for the OEC in the  $S_0$ ,  $S_1$ ,  $S_2$ , and  $S_3$  states [16,17] in  $k$  space to the calculated  $k$ -space spectrum of the crystal structure (see Fig. 1).

[Figure 1 about here.]

We used Fermi energy values  $E_0$  from 6540 to 6580 eV (in steps of 1 eV) for the calculations in order to get the best overlap of the experimental data with the calculated one from the crystal structure in this energy range. The smallest error has been found for an  $E_0$  value of 6568 eV and we used this  $E_0$  value for the EXAFS calculations involving the 3ARC crystal structure. The highest contribution was obtained for the  $S_0$  experimental spectrum (about 60 %).

## Structural refinement using EXAFS calculations

The refinement procedure uses a scoring function defined in terms of the root mean square deviation of the calculated isotropic and polarized and the corresponding experimental EXAFS spectra. In order to minimize this scoring function, the refinement procedure iteratively adjusts the atomic coordinates of the molecular system under study by using a conjugate gradient optimization method [15]. The molecular structure is changed in such a way that the nuclear displacements required for a better agreement of calculated with the experimental EXAFS data are minimal compared to the starting structure. This is ensured by applying a quadratic penalty factor, which evaluates the root of the squared distances between the atomic coordinates of the actual and the starting structure, to the above-mentioned scoring function. We excluded the hydrogen atoms from the refinement procedure since their contribution to EXAFS spectra is very small.

### Cartesian Coordinates from the DFT-QM/MM Model (in Å)

Mn -27.328174 -35.175369 205.339330  
Mn -24.929089 -35.653121 203.980346  
Mn -27.220808 -33.274222 203.282474  
Mn -27.619399 -33.414781 200.566152  
O -26.533593 -36.480326 204.355112  
O -25.987041 -34.138352 204.615790  
O -26.724514 -34.332209 201.905640  
O -28.333312 -34.594725 203.944351  
O -28.379467 -32.630669 202.053159  
Ca -27.954967 -36.717887 202.430369  
C -29.960710 -35.520519 199.884851  
O -29.680127 -36.341734 200.833948  
O -29.462768 -34.324949 199.843910  
C -21.636836 -33.288768 202.485318  
N -21.266658 -33.640152 203.791804  
C -22.900900 -33.835727 202.354885

C -22.246091 -34.354168 204.387534  
N -23.265370 -34.502614 203.529767  
H -20.389934 -33.439398 204.268859  
H -23.569995 -33.823079 201.516321  
H -22.171263 -34.752790 205.385678  
C -25.517462 -31.372303 201.657171  
O -25.927591 -31.829710 202.806847  
O -25.962066 -31.777754 200.512267  
C -25.054337 -36.225262 207.001174  
O -26.283369 -35.817938 207.010205  
O -24.318916 -36.199520 205.928169  
O -29.540886 -37.273955 204.135436  
O -28.720833 -36.356156 206.045487  
C -24.323008 -38.251199 202.177134  
O -24.931790 -38.605244 201.117703  
O -24.556179 -37.045989 202.676995  
C -28.251589 -32.488359 206.095531  
O -28.068955 -33.709827 206.473723  
O -27.993082 -32.082348 204.883270  
C -29.373963 -37.286023 205.406251  
O -26.584716 -41.130967 206.680270  
O -29.216332 -29.894170 203.822770  
O -30.700603 -31.438022 202.217870  
O -26.304866 -39.245967 204.742661  
O -26.694951 -34.649889 199.195413  
O -28.394011 -32.095315 199.151825  
O -27.625685 -39.051827 202.614922  
O -26.772234 -37.024952 200.285768  
H -28.639902 -29.193628 203.459359  
H -28.651948 -30.606218 204.231616  
H -27.321661 -37.582808 199.698126

H -26.067877 -38.343014 205.026686  
H -26.258148 -39.936565 205.455928  
H -28.358380 -31.121896 199.208209  
H -26.559980 -35.573517 199.621263  
C -32.101350 -34.602836 203.762404  
N -31.899743 -35.918465 203.793158  
N -31.151372 -33.751387 203.439004  
H -32.659572 -36.585225 203.967721  
H -30.992202 -36.374065 203.646332  
H -31.232559 -32.733144 203.137020  
H -30.185879 -34.097286 203.343625  
H -26.417208 -40.604417 207.513108  
H -25.841216 -41.786426 206.663884  
H -30.358971 -30.695178 202.800658  
H -29.861466 -31.895284 201.927840  
H -29.243470 -32.419693 198.776723  
H -28.288290 -39.776272 202.506882  
H -27.071099 -39.262203 203.456712  
H -25.954026 -37.582012 200.593447  
H -27.117013 -34.811743 198.303505

**Cartesian Coordinates from the refined R-QM/MM Model (in Å)**

Mn -27.196 -35.180 205.358  
Mn -24.884 -35.642 203.926  
Mn -27.127 -33.369 203.342  
MN -27.364 -33.485 200.590  
O -28.375 -32.699 201.980  
O -25.951 -34.221 204.521  
O -26.552 -34.454 201.921  
O -26.465 -36.567 204.295

O -28.317 -34.650 203.942  
Ca -27.890 -36.639 202.385  
O -28.029 -33.770 206.538  
O -27.903 -32.090 205.027  
O -26.138 -35.791 206.882  
O -24.190 -36.240 205.808  
O -24.520 -38.279 201.048  
O -24.156 -36.718 202.616  
O -25.874 -31.639 203.146  
O -25.900 -31.553 200.879  
O -29.750 -35.919 200.986  
O -28.827 -34.783 199.279  
O -29.462 -37.225 204.086  
O -28.495 -36.635 206.040  
O -26.097 -33.995 199.024  
O -26.875 -37.266 200.251  
C -30.747 -36.141 198.806  
C -29.728 -35.573 199.781  
C -23.955 -37.898 202.102  
C -21.428 -34.214 202.558  
N -21.166 -34.409 203.903  
C -22.241 -34.994 204.474  
N -23.191 -35.184 203.549  
C -22.694 -34.702 202.347  
C -24.667 -29.875 201.990  
C -25.541 -31.122 202.014  
C -24.395 -36.871 208.116  
C -24.930 -36.245 206.839  
C -29.915 -38.591 206.006  
C -29.248 -37.411 205.315  
C -28.867 -31.600 207.236

C -28.222 -32.554 206.185  
C -32.159 -34.868 204.089  
N -32.149 -36.201 204.133  
N -31.061 -34.156 203.935  
O -27.255 -38.643 207.776  
O -27.121 -36.033 197.764  
O -29.127 -29.765 204.234  
O -30.641 -31.615 202.877  
O -27.277 -39.024 202.715  
O -26.270 -39.116 205.193  
O -28.313 -32.081 199.139  
C -23.261 -39.211 202.916  
C -33.473 -34.217 204.258

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## List of Figures

- 1 Calculated isotropic EXAFS spectra (left panel:  $k^3$ -weighted EXAFS spectra; right panel: FT spectra in  $R$  space) for the 3ARC crystal structure (black) and the fit based on experimental data for the  $S_0$ - $S_3$  states [16, 17]. . . . . 12

Figure 1: Calculated isotropic EXAFS spectra (left panel:  $k^3$ -weighted EXAFS spectra; right panel: FT spectra in  $R$  space) for the 3ARC crystal structure (black) and the fit based on experimental data for the  $S_0$ - $S_3$  states [16, 17].

