

Supplementary data.

Simulation of the F-Tyr and estimation of the incorporation yields.

The simulation of the 3F-Tyr EPR spectrum was done using Bruker SimFonia software, using starting EPR parameters obtained from a full DFT optimization of the F-Tyrosyl-Histidine pair at B3LYP/6-31+g* level and subsequent calculation of the magnetic observables using B3LYP functional combined with the EPR-II basis set of Barone. The g-tensor was calculated at the same level of theory as the optimization of the F-Tyrosyl-Histidine pair using GIAO approach as implemented in Gaussian 03 software. The parameters were subsequently refined by adjusting the simulation to the experimental spectrum. Although the proton hyperfine couplings for H3, H4 and H6 are less than the experimental line width, they were included in the simulation based on the computational results. However, their contribution to the overall spectral shape is very small. The following parameters for the ^{19}F and ^1H hyperfine coupling were used in the final simulation:

Table S1: g matrix and the hyperfine coupling parameters used for the simulation of the 3F-Tyr spectrum.

g matrix	$g_{xx} = 2.074$	$g_{yy} = 2.044$	$g_{zz} = 2.033$
Atom	A_{xx} (MHz)	A_{yy} (MHz)	A_{zz} (MHz)
F ₁	-15.5	-10.0	62.0
H ₂	-10.5	-3.2	-7.0
H ₃	1.7	2.7	0.4
H ₄	1.7	2.7	0.4
H ₅	8.3	7.7	7.6
H ₆	2.5	2.5	1.0

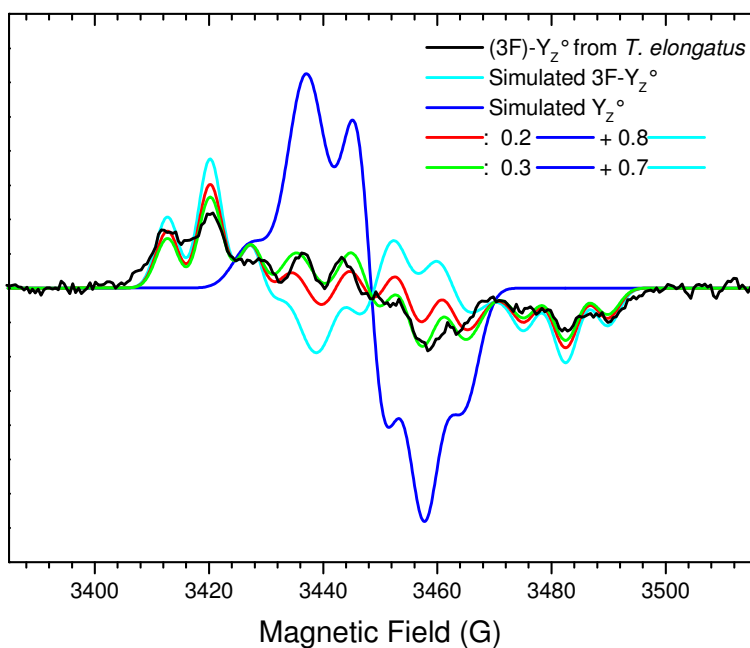


Figure S1. Simulated EPR spectrum of 3F-Tyr radical (cyan line), simulated spectrum of Tyr radical (blue) and the mixture of both with the ratio 8:2 (80% 3F-Tyr; 20% Tyr) and 7:3 (70% 3F-Tyr; 30% Tyr).

The estimate of the incorporation yield was done using the procedure outlined in the manuscript. The final simulated spectra of the incorporated 3F-Tyr were obtained by summing the appropriate fractions of the normalized 3F-Tyr and Tyr radical spectra. In the Figure S1 are shown the simulated 3F-Tyr spectrum (cyan), the simulated Tyr spectrum (blue) and the mixture of both with the ratio of 8:2 (red, 80% 3F Tyr; 20% Tyr) and 7:3 (green).

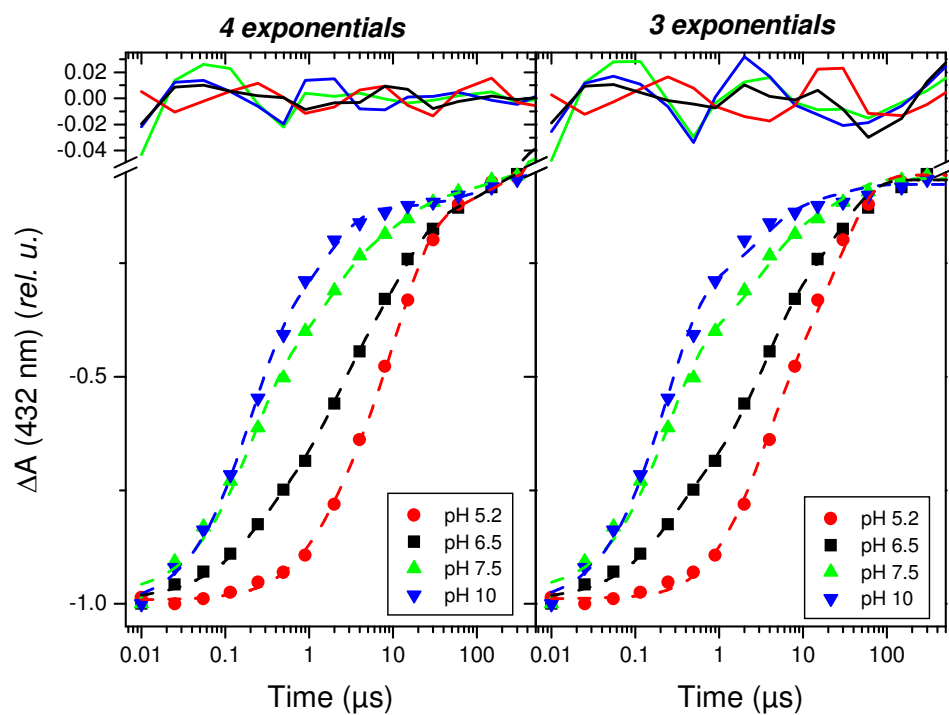


Figure S2: pH dependence of the reduction of $P_{680}^{+\bullet}$ in Mn-depleted PSII from *T. elongatus*. The transient absorption changes were globally fitted with four (left panel, dashed lines) and (right panel, dashed lines) three exponentials, imposing pH-independent time constants. In both panels the solid lines are the residuals of the fits with the corresponding color code.

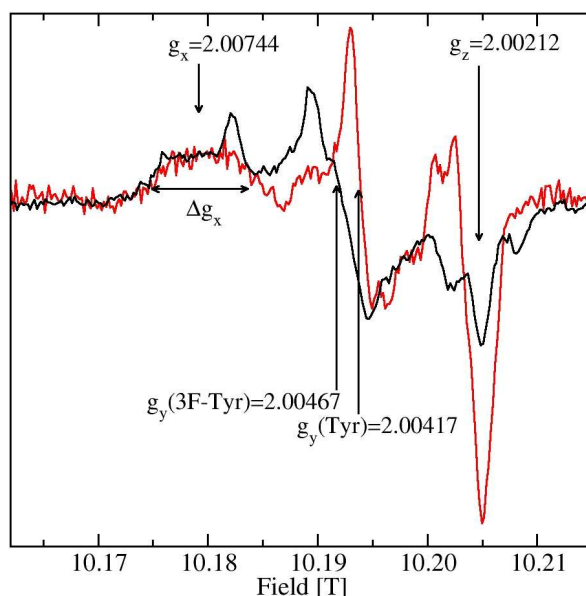


Figure S3: 285 GHz High-field EPR spectra of Y_ZO° (black) and $3F\text{-}Y_ZO^\circ$ (red) in PSII complexes from *T. elongatus*, at pH 9. The g_x and g_z values of both radicals were essentially identical. As discussed in Reference 1, the magnitude of g_x value is determined by the electrostatic environment around the radical. The broadness of the Y_ZO° and $3F\text{-}Y_ZO^\circ$ g_x regions (denoted by Δg_x) indicates that in both case the environment is highly distributed, but apparently to the same degree. Hence, the identical g_x values and distributions demonstrate that the electrostatic interaction between the Y_Z radical and the protein is unaffected by presence of the fluorine substituent.

Table S2. Summary of the geometries and hydrogen bonding energies of the 4-ethylphenolate/imidazole, 3-fluoro-4-ethylphenolate/imidazole hydrogen-bonded supercomplexes and their respective oxidized radical versions as determined from B3LYP/6-31+G(D,P) density functional theory calculations. Calculations were performed in dielectric with $\epsilon=20$ using the polarizable continuum model. g -value calculations for the radicals were carried at the same levels. The calculations were performed using the Gaussian 03 package. Reference g -values in the absence of hydrogen bonding are given parenthesis. (see Reference 3 for details).

X	Hydrogen Bonding Energies (kcal/mol)	Hydrogen Bonding Geometry		g-Values		
		Distance (O..N)	Angle (CO..N)	g_x	g_y	g_z
3-X-4-ethylphenolate/imidazole						
F	-5.9	2.69	131			
H	-6.4	2.64	125			
3-X-4-ethylphenoxy radical/imidazole						
H	-0.7	2.92	135	2.0077 (2.0087)	2.0045 (2.0047)	2.0022 (2.0021)
F	-1.1	2.88	136	2.0076 (2.0084)	2.0049 (2.0050)	2.0021 (2.0021)

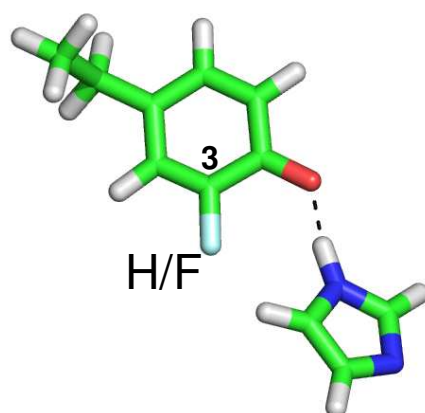


Figure S4. The B3LYP/6-31+G(D,P) geometry of the 3-Fluoro-4-ethylphenolate/imidazole supercomplex.

Like the high-field EPR measurements, these computational results show that the fluorine substitution has a negligible effect on energy and geometry of hydrogen bonding between ethylphenol and imidazole in reduced and oxidized radical state. Consistent with the experimental data, the calculations show that the only effect of the fluorine substituent is a small positive shift in the g_y (or shift to lower magnetic field) relative to hydrogen (see Figure S3).

References

1. Un, S., Tang, X. S. & Diner, B. A. (1996) 245 GHz high-field EPR study of tyrosine-D degrees and tyrosine- Z degrees in mutants of photosystem II *Biochemistry* 35: 679-684.
2. Un, S., Atta, M., Fontecave, M. & Rutherford, A. W. (1995) g-Values as a probe of the local protein environment: High-Field EPR of Tyrosyl radicals in Ribonucleotide Reductase and Photosystem II *J Am Chem Soc* 117 : 10713-10719
3. Un S. (2005) The g-values and hyperfine coupling of amino acid radicals in proteins: comparison of experimental measurements with ab initio calculations. *Magn Reson Chem.* 43, S229-S239.