

Supporting Information

Fluctuations of an Exposed π -Helix Involved in Lipoyxygenase Substrate Recognition.

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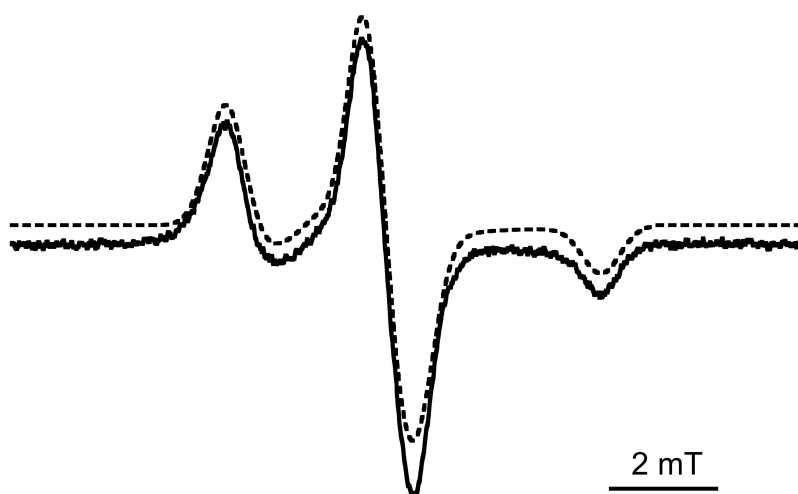


Figure S1. CW-EPR spectrum of Q267R₁ SBL1 at 60K (*solid*) with simulation (*dashed*). Spectrum recorded at 9.404 GHz microwave frequency, 0.2 μ W microwave power, 2 Gauss modulation amplitude, with 100 kHz modulation frequency. Spectrum was simulated with the program pepper of the EasySpin software package¹ (version 4.5.5) using a Gaussian line shape with an 8.4 G (FWHH) line width. The spectrum was simulated using the following g tensor values: g_{xx} : 2.0084, g_{yy} : 2.0061, g_{zz} : 2.0020; and the following A tensor values: A_{xx} : 3.5 G, A_{yy} : 5.5 G, A_{zz} : 35.4 G.

Table S1: Comparison of Steady State Kinetic Constants of Spin-Labeled Mutants of SBL1.

| Residue | k_{cat} (s^{-1}) | Km (μM) |
|---------------------------------|--------------------------------------|----------------------|
| WT ^a | 234 | 16 |
| NoCys ^a | 215, 178 ^b | 20, 18 ^b |
| I257R ₁ | 250 ± 9 ^c | 20 ± 4 |
| T259R ₁ | 174 ± 5 | 7.5 ± 0.6 |
| K260R ₁ | 149 ± 4 | 6.5 ± 0.5 |
| S261R ₁ | 186 ± 8 | 8.0 ± 0.9 |
| L262R ₁ | 173 ± 6 | 15 ± 1 |
| S263R ^d | 203 ± 4 | 26 ± 1 |
| Q264R ₁ | 126 ± 2 | 13.3 ± 0.5 |
| I265R ₁ | 218 ± 5 | 8.3 ± 0.5 |
| Q267R ₁ | 185 ± 4 | 12.4 ± 0.6 |
| F270R ₁ ^a | 96 | 13 |
| F274R ₁ | 230 ± 4 | 4.9 ± 0.3 |

^aKinetic constants of WT, cysteine-free (NoCys), and F270R₁ SBL1 from Gaffney *et al.* 2012. ^bKinetic constants for NoCys determined by detergent-free assay shown for comparison with S263R₁ kinetic constants. ^cUncertainties of the fit to the Michaelis-Menten equation are reported as mean-squared error. ^dS263R₁ displayed non steady state kinetics in the presence of the detergent TWEEN-20 and was determined by detergent-free assay.

References:

(1) Stoll, S., Schweiger, A. (2006). EasySpin, a comprehensive software package for spectral simulation and analysis in EPR. *J. Magn. Reson.* 178, 42-55.