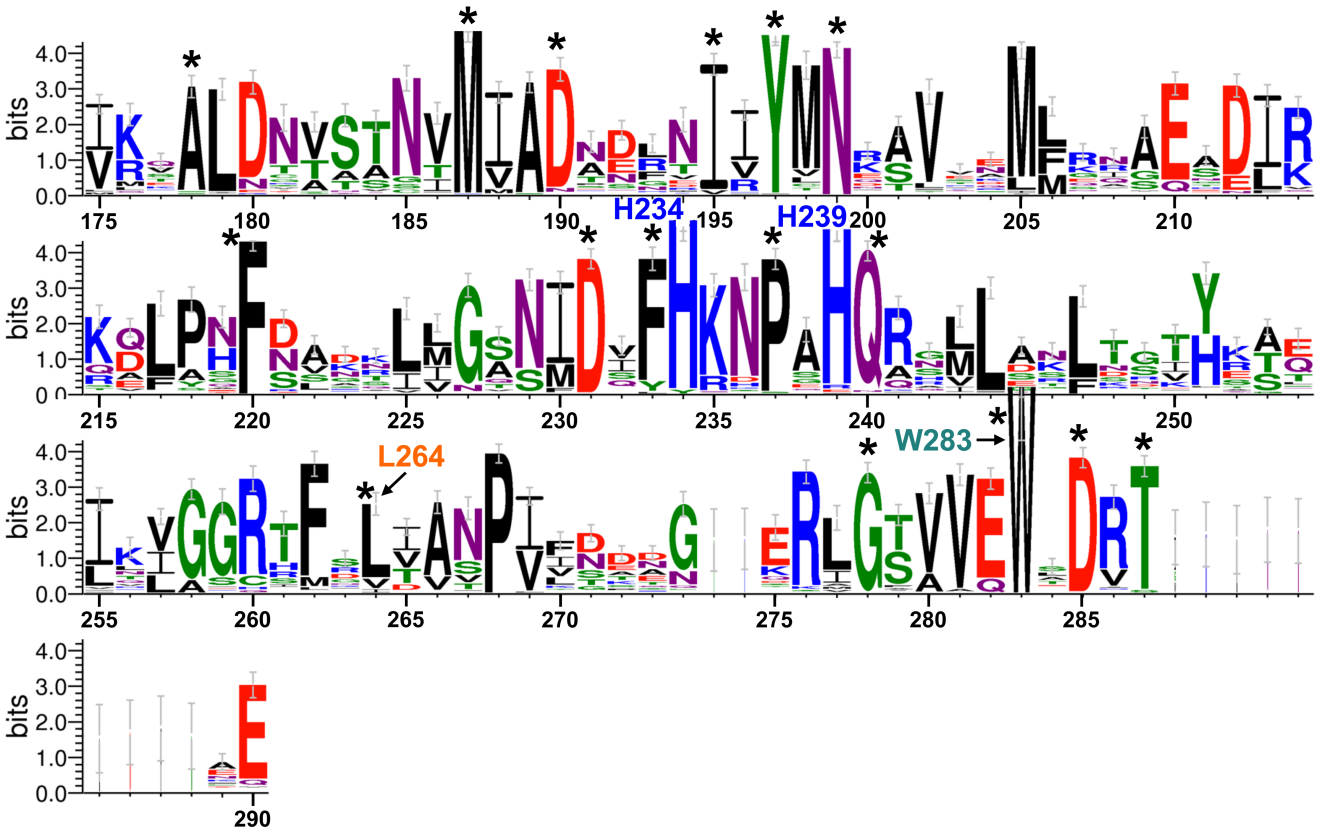

Fig. S1. WebLogo sequence alignment of 100 Aer2 PAS domain-like sequences. Sequence homologs were acquired by performing an NCBI protein BLAST search (http://blast.ncbi.nlm.nih.gov/Blast.cgi?PAGE=Proteins) against *P. aeruginosa* PAO1 Aer2 residues 175-290, and the top 100 sequences were aligned to create a WebLogo [http://weblogo.berkeley.edu (1)]. The overall height of each letter stack indicates the sequence conservation at that position (measured in bits), while the height of each letter within the stack indicates the relative frequency of each amino acid at that position. Error bars are provided at twice the height of sample correction for positions with limited sequence information. Asterisks indicate the 16 conserved residues that were selected for site-directed alanine mutagenesis.

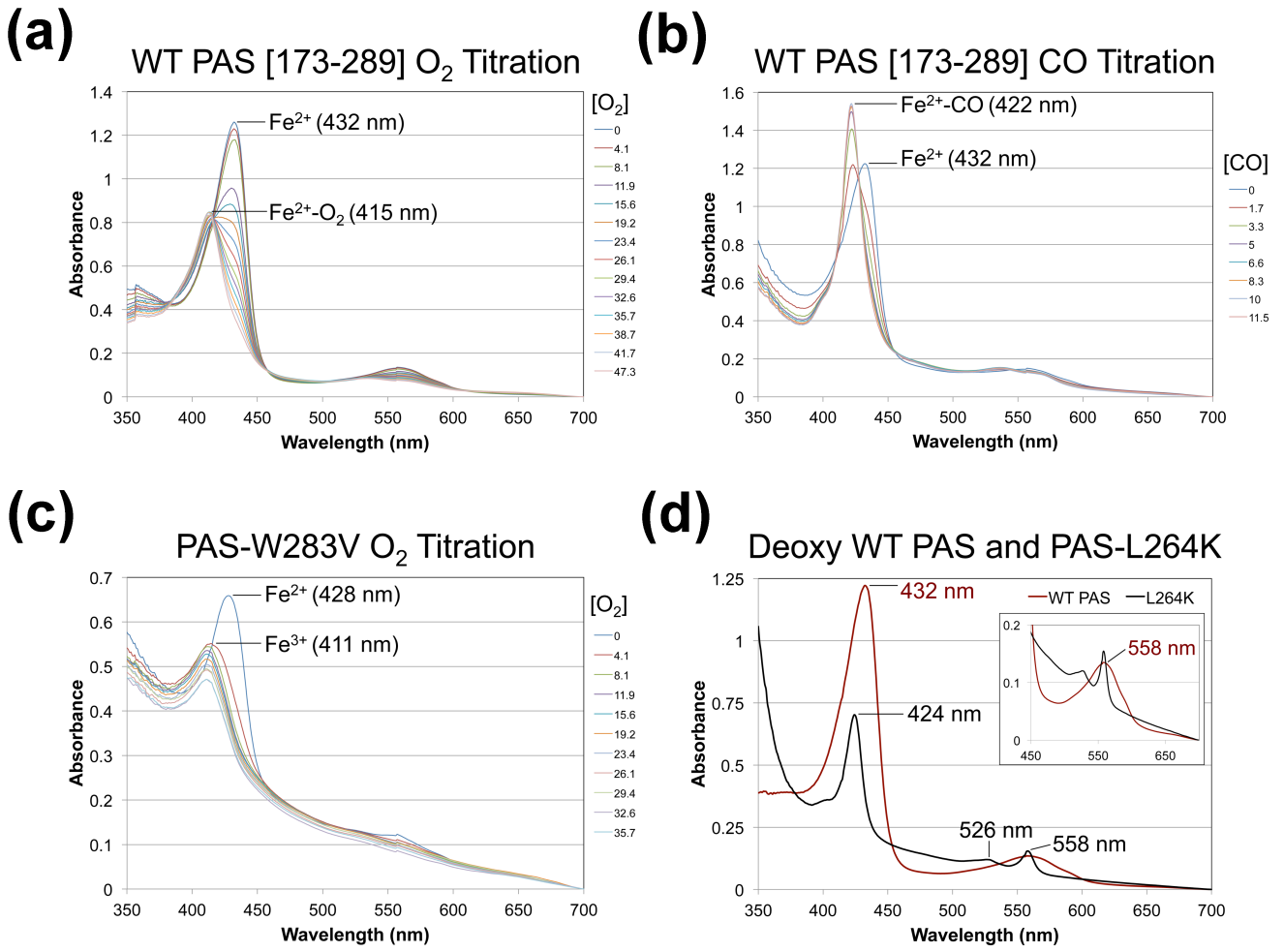

Fig. S2. Examples of gas titrations using 10 μM purified Aer2[173-289] PAS peptides. (a-b) WT deoxy-PAS peptide titrated with 5 μl aliquots of air-saturated buffer (a), and 0.5 μl aliquots of CO-saturated buffer (b). (c) Deoxy PAS-W283V peptide titrated with air-saturated buffer, showing rapid oxidation to met-heme (with a soret maximum of 411 nm). The designation of met-heme instead of oxy-heme was verified spectrophotometrically after oxidizing PAS peptides with potassium ferricyanide and comparing the spectra. (d) Deoxy spectra of WT Aer2-PAS and Aer2-L264K. Aer2-L264K exhibits β and α bands (526 and 558 nm, respectively), which is indicative of hexa-coordinate heme, whereas WT Aer2 has a single broad band with a 558 nm maxima (see enlarged inset), which is indicative of penta-

coordinate heme. The spectra in panels (a), (b) and (c) are raw scans and have not been adjusted for dilution.

Reference

1. **Crooks GE, Hon G, Chandonia JM, Brenner SE.** 2004. WebLogo: a sequence logo generator. Genome Res **14:**1188-1190.