Supplementary Material

Determination of the Open-Bundle Structure of Cytochrome *c*' as a Folding/Unfolding Intermediate by Small-Angle Neutron Scattering

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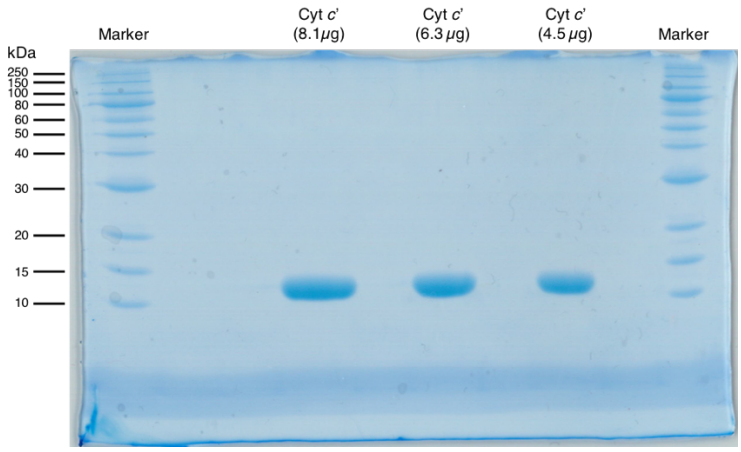


Figure S1. SDS-PAGE of purified Cyt *c*' on a 12.5% gel.

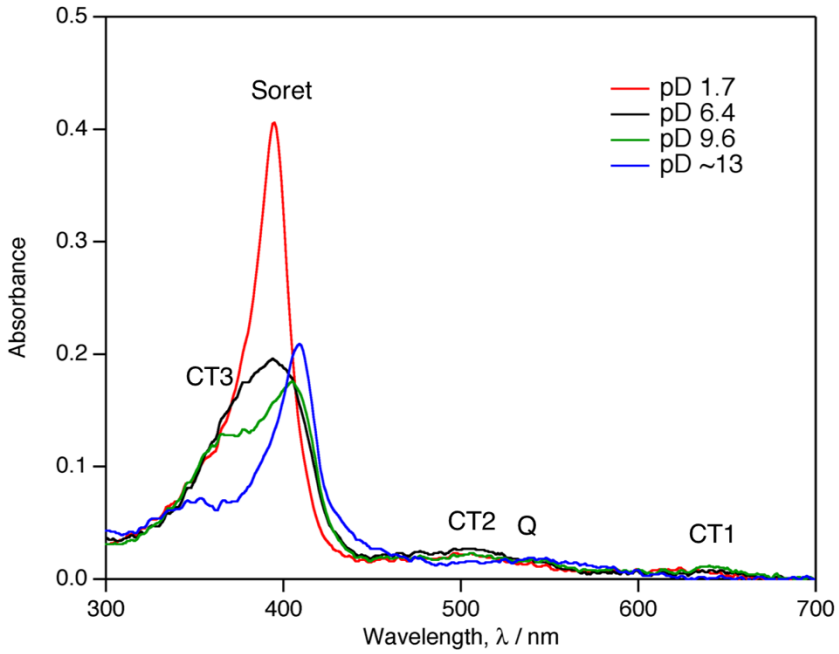


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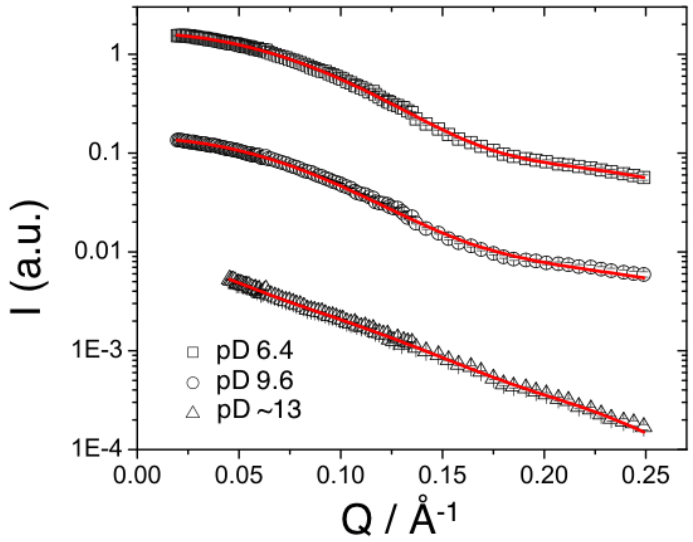


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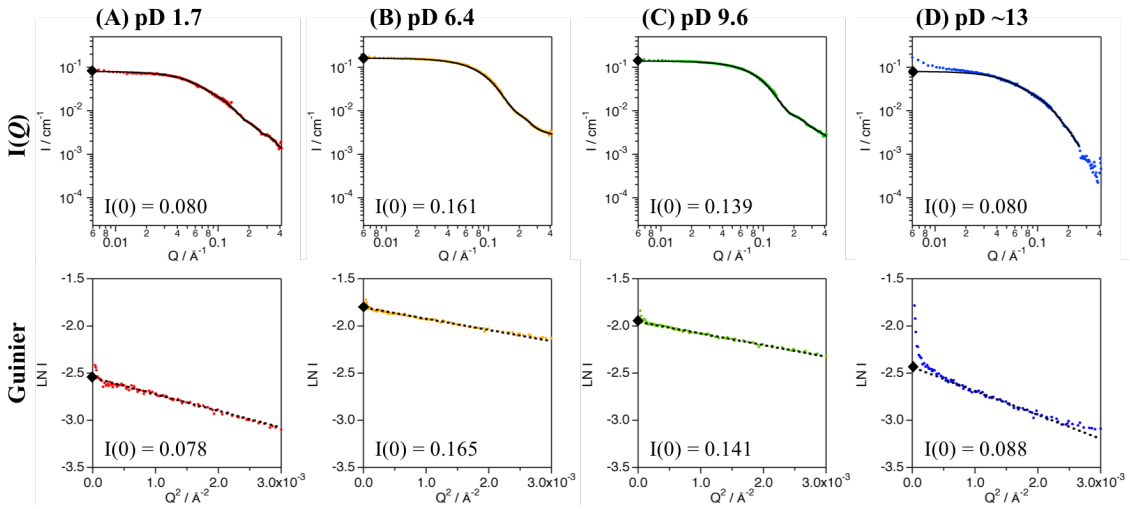


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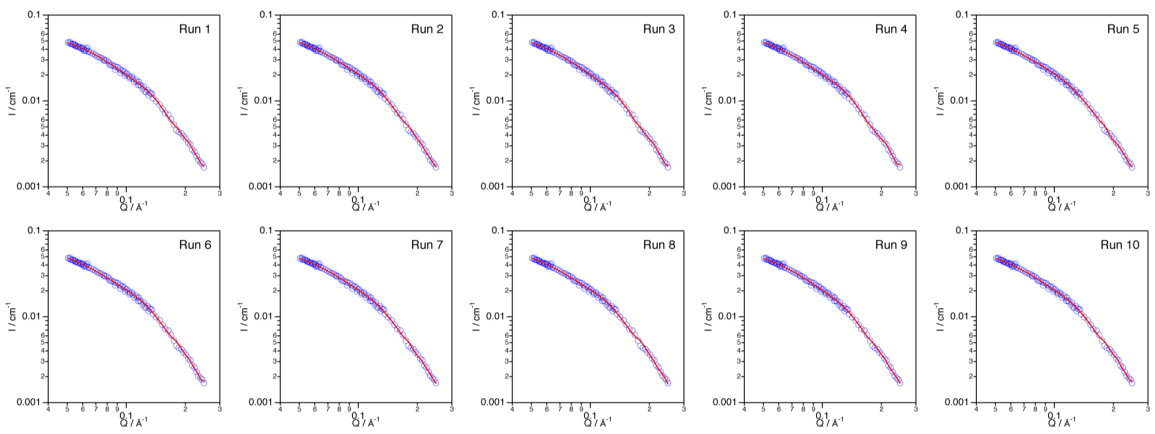


Figure S5. Fitting of BUNCH simulations (red line) versus the experimental SANS curve at pD ~13 (blue circle).

Table S1. Details of sample used in SANS experiment.

	Cytochrome *c′*
Organism	*Alcaligenes xylosoxidans*
Source	NCIMB 11015
UniProt Sequence ID (residues in construct)	P00138(1-127)
Molar Extinction Coefficient / M^{-1} cm^{-1}	96,000 (Soret band)
Molecular Weight (Da) for Monomer	14227.2 (determined by ESI-MS)
Concentration / mg mL^{-1}	5.3-5.5

Table S2. SANS data collection parameters.

Instrument	KWS-1 (JCNS/FRM-II)
Wavelength / Å	5 Å with a spread $\Delta\lambda/\lambda$ = 10%
Beam size / µm	10,000 x 10,000
Camera length	C+D = 8+8 m (Low-Q), 4+4 m (Medium-Q, transm.), 4+1.5 m (High-Q)
Q measurement range / $Å^{-1}$	0.005997535 - 0.4162468
Absolute scaling method	Comparison with scattering intensity of secondary standard Plexiglas
Normalization	Secondary Plexiglas
Exposure time	4800 s (8m), 900 s (4m), 7200 s (1.5m)
Sample temperature / K	293 K

Table S3. Software employed for SANS data reduction, analysis, and interpretation.

SANS data reduction	qtiKWS
Basic analyses: Guinier, P(r)	PRIMUS in ATSAS v. 2.8.2
Shape/bead modelling	DENFERT v.2
Preparation of volumetric map/model docking	pdb2vol/collage in Situs v.2.8. package
Numerical model fitting	IGOR Pro v. 6.04, Maple

Table S4. Structural parameters determined from SANS curves.

	pD 1.7	pD 6.4	pD 9.6	pD ~13
Guinier analysis				
I(0) / cm^{-1}	0.078±0.001	0.165±0.001	0.141±0.001	0.088±0.002
R_g / Å	23.02±0.57	18.93±0.45	19.24±0.49	27.58±0.59
QR_g limit	0.73-1.03	0.60-0.84	0.61-0.86	0.87-1.23
P(r) analysis				
I(0) / cm^{-1}	0.0804±0.0004	0.1609±0.0008	0.1389±0.0002	0.0804±0.0014
R_g / Å	25.03±0.19	18.24±0.08	18.10±0.05	25.54±0.60
d_{max} / Å	86.21	54.87	53.16	89.52
Q range / $Å^{-1}$	0.0197-0.4160	0.0197- 0.4160	0.0197-0.4160	0.0321-0.2540