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Supporting information for article:

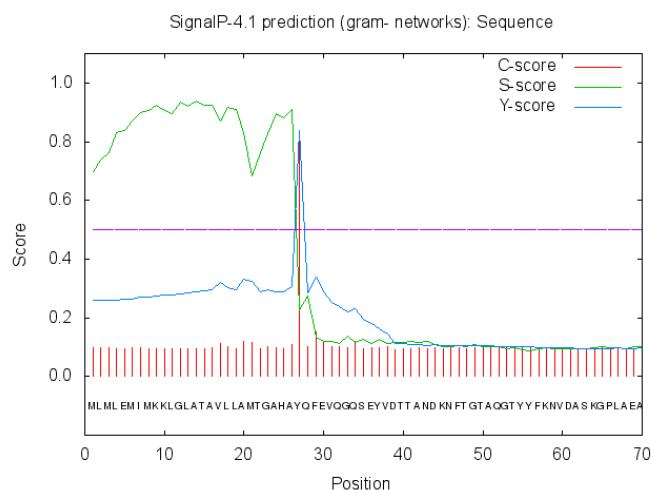
Crystal structure of the *Acinetobacter baumannii* outer membrane protein Omp33

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(a) Signal sequence prediction.

Measure	Position	Value	Cutoff	signal peptide?
max. C	27	0.818		
max. Y	27	0.839		
max. S	14	0.939		
mean S	1-26	0.861		
D	1-26	0.849	0.570	YES

SP='YES' Cleavage site between pos. 26 and 27:
AHA-YQ (Petersen et al., 2011)

**(b) Predicted transmembrane (TM)-strands positions by BOCTOPUS2**

TM1	TM2	TM3	TM4	TM5	TM6	TM7	TM8	TM9	TM10	TM11	TM12
2-11	21-30	51-60	74-83	90-99	114-123	127-136	168-178	184-194	230-240	244-253	265-274

(c) Predicted transmembrane (TM)-strands positions by PRED-TMBB**Viterbi method**

YQFEVQGQSEYVDTTANDKNFTGTAQGTYYFKNVDASKGPLAEAAFLNQASNVSVAYNYIKYDEKDTVNVESHTYGVKGEAYLPTPYLPVYASASYNHTINDFKDGVSDDNGDRYALEAGAMLLPNFLVAVGYTSAVDQISLDAFGVNVKYGIAKAVGESVAIDEKQDAVTARTKYVGNIDGTNMAIGFEAFGVFAEDNAYGMKTDLFVTPKLSVGASFADVSAFNSGYDHWWGGHTQYFITPAVAVGADFVKANAKDGNPRDTQTIGLNAKFRE

TM1	TM2	TM3	TM4	TM5	TM6	TM7	TM8	TM9	TM10	TM11	TM12
1-11	36-46	53-59	73-83	90-98	115-123	127-133	184-194	206-218	224-232	238-250	264-274

N-best method

YQFEVQGQSEYVDTTANDKNFTGTAQGTYYFKNVDASKGPLAEAAFLNQASNVSVAYNYIKYDEKDTVNVESHTYGVKGEAYLPTPYLPVYASASYNHTINDFKDGVSDDNGDRYALEAGAMLLPNFLVAVGYTSAVDQISLDAFGVNVKYGIAKAVGESVAIDEKQDAVTARTKYVGNIDGTNMAIGFEAFGVFAEDNAYGMKTDLFVTPKLSVGASFADVSAFNSGYDHWWGGHTQYFITPAVAVGADFVKANAKDGNPRDTQTIGLNAKFRE

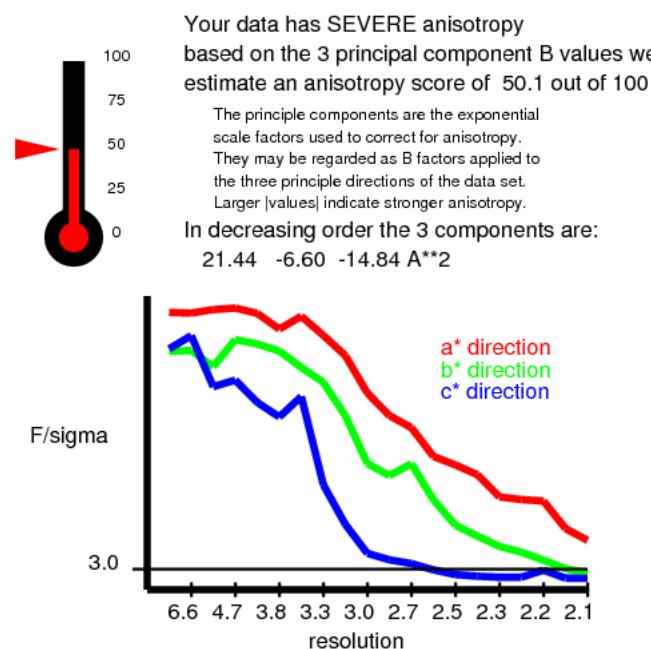
TM1	TM2	TM3	TM4	TM5	TM6	TM7	TM8	TM9	TM10	TM11	TM12
1-11	36-46	53-59	73-83	90-98	115-123	127-133	184-194	206-218	224-232	238-250	264-274

Posterior decoding method

**YQFEVQGQSEYVDTTANDKNFTGTAQGTYYFKNVDASKGPLAEAAFLNQASNVSVAYNYIKYDEKD
TVNVESHTYGVKGEAYLPTPYLPVYASASYNHTINDFKDGVSDDNGDRYALEAGAMLLPNFLVAVG
YTSVADQISLDAFGVNKYGIAKAVGESVAIDEKQDAVTARTKYVGNIIDGTNMAIGFEAFGVFAEDN
AYGMKTDLFVTPKLSVGASFADVSAFNNSGYDHVWGHTQYFITPAVAVGADFVKANAKDGNPRDTQ
TIGLNAKFRF**

TM1	TM2	TM3	TM4	TM5	TM6	TM7	TM8	TM9	TM10	TM11	TM12
2-11	38-46	53-59	73-83	88-98	115-	127-	169-	184-	200-	212-	228-
					123	133	177	194	210	218	236
TM13 TM14											
238-	264-										
250	273										

Figure S1 Predicted signal sequence and topology of Omp33. (a) SignalP 4.1 prediction of the cleavage of the signal sequence for Omp33. (b) predicted transmembrane residues from boctopus2. (c) PRED-TMBB predicted transmembrane residues from the three different methods that the software uses, Viterbi, N-best and posterior decoding. In red the transmembrane strands, green for the periplasmic regions (turns) and blue for the extracellular loops.



The recommended resolution limits along/near to a^*, b^*, c^* are
2.1 Ång 2.1 Ång 2.6 Ång

These are the resolutions at which F/σ
drops below an arbitrary cutoff of 3.0
 F/σ in highest shell is greater than cutoff. Consider reprocessing data to higher resolution.

16532 reflections were in the initial data set. 3111 were discarded because they fell outside the specified ellipsoid with dimensions 1/2.1, 1/2.1, 1/2.6 Å⁻² along a^*, b^*, c^* , respectively. These discarded reflections had an average F/σ of 2.87.

13421 reflections remain after ellipsoidal truncation. Anisotropic scale factors were then applied to remove anisotropy from the data set. Lastly, an isotropic B of -19.57 Å⁻² was applied to restore the magnitude of the high resolution reflections diminished by anisotropic scaling. The following pseudo precession images illustrate the individual steps.

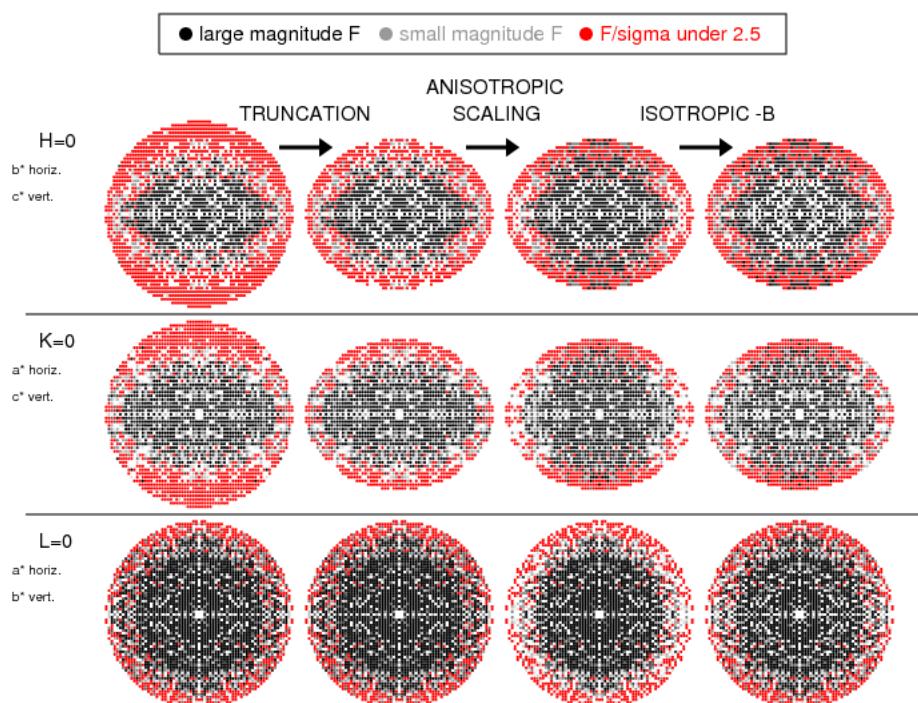


Figure S2 Diffraction anisotropy server results.

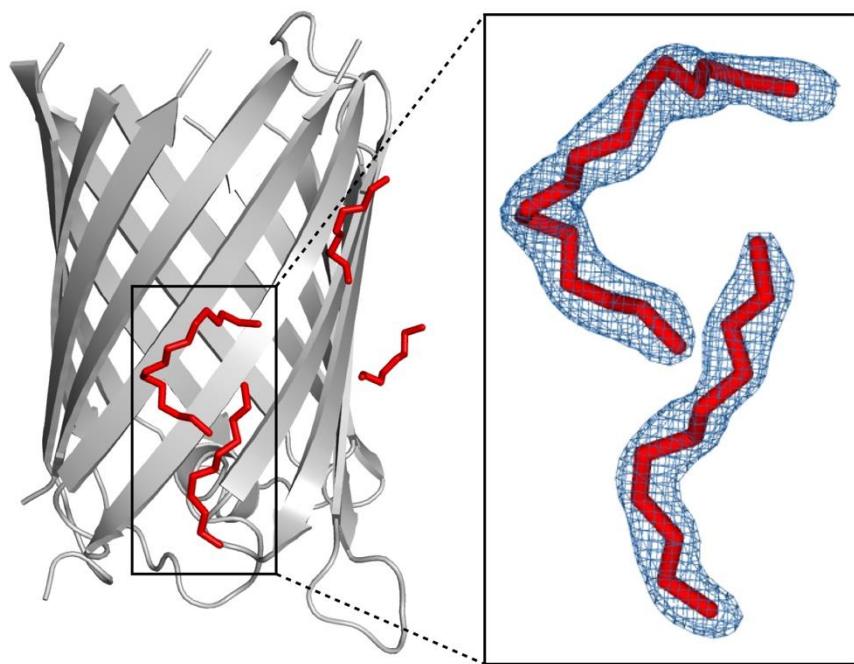


Figure S3 Modelled C₈E₄ molecules. Left panel, cartoon representation of the structure of Omp33 (in grey) showing the four molecules of C₈E₄ (in red). Notice that not all the molecules of detergent are fully modelled due to the lack of electron density for the whole molecule. Right panel, a close-up showing 2Fo-Fc electron density at 1.5 σ for the best-ordered C₈E₄ molecules.

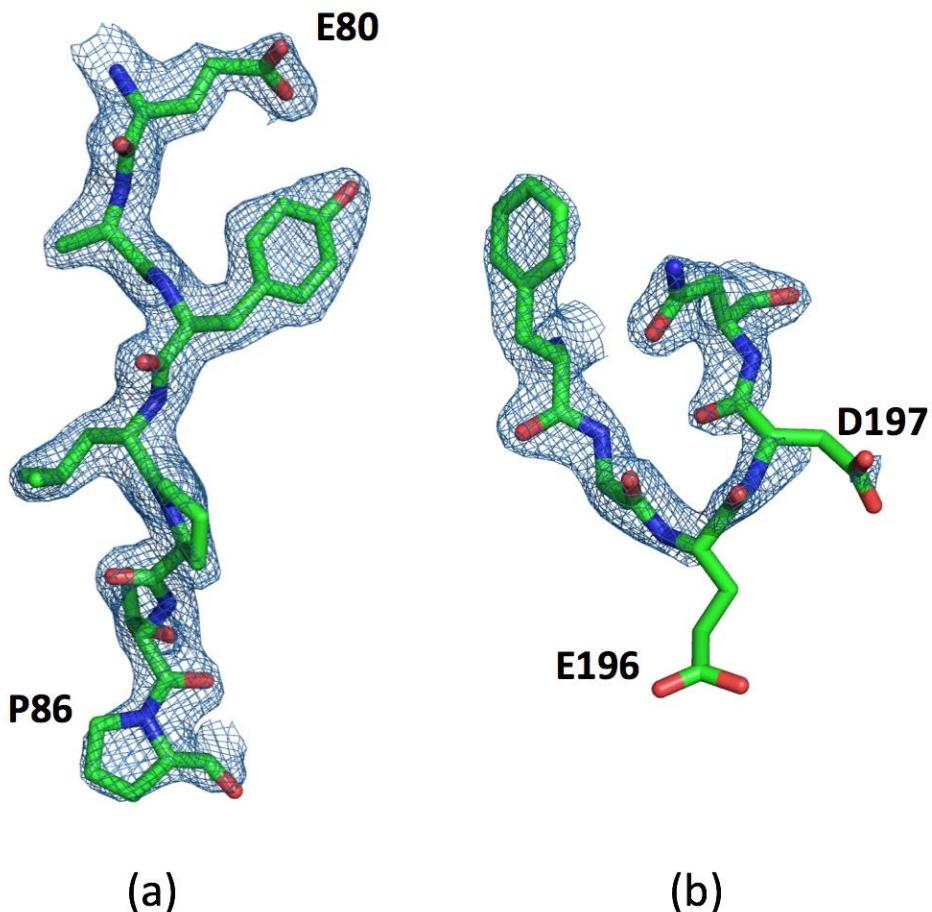


Figure S4 Close up view of Omp33. Both panels show the $2\text{Fo}-\text{Fc}$ electron density contoured at 1.5σ . (a) Diagram showing the fit of the electron density for a well-defined region of the model. (b) Figure showing the lack of electron density for the side chains of Glu196 and Asp197, probably due to flexibility.

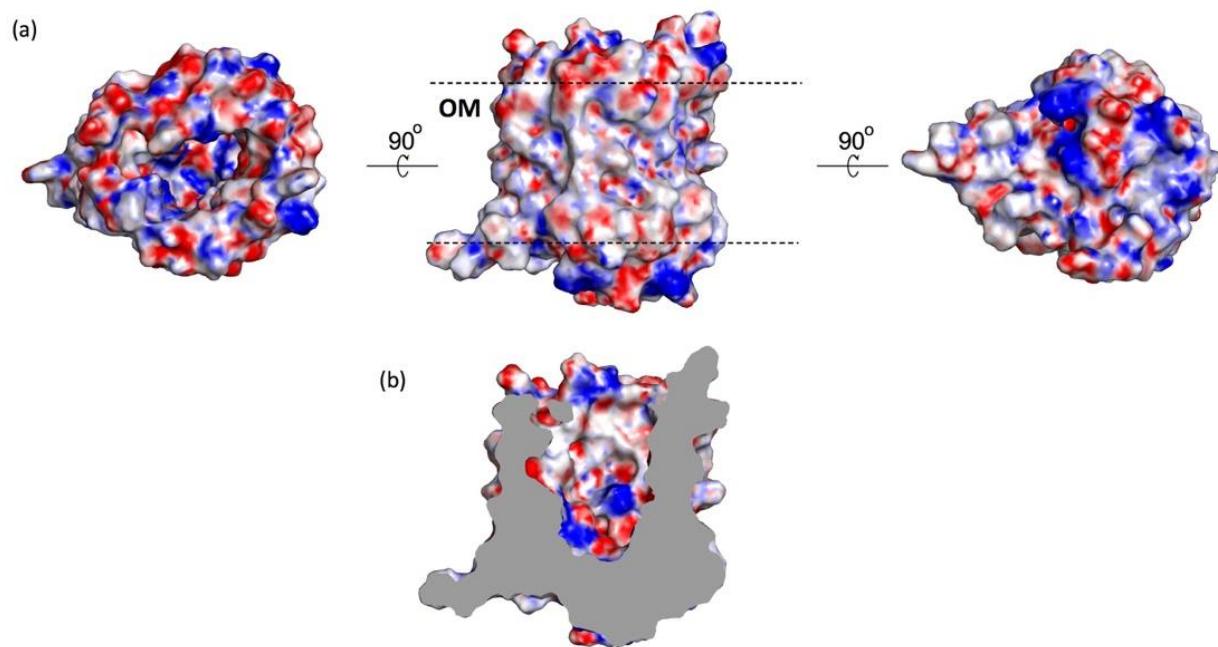


Figure S5 Omp33 electrostatics. (a) Views from the extracellular side (left), OM plane (middle) and periplasmic space (right). The electronegative areas are presented in red, neutral in white and electropositive in blue. The electrostatic potential was calculated by PBEQ Solver (Jo et al., 2008) and coloured from -14 kT/e (red) to +14 kT/e (blue). The missing extracellular loops are not modelled. (b) Surface slice through the centre of the Omp33 β -barrel, showing the hydrophilic nature of the interior of the barrel.

Table S1 Omp33 resembles outer membrane protein G (OmpG) and CymA

Summary of a DALI comparison between the Omp33 X-ray crystal structure and those in the Protein Data Bank.

No	Chain	Z-score	Rmsd (Å)	lali	nres	%id	PDB Description
1	3dwo-X	17.4	2.5	181	444	12	PROBABLE OUTER MEMBRANE PROTEIN
2	4ctd-B	17.2	3.2	187	231	10	OUTER MEMBRANE PROTEIN G
3	4ctd-A	17.2	3.1	187	234	9	OUTER MEMBRANE PROTEIN G
4	2f1c-X	16.6	3.1	187	252	9	OUTER MEMBRANE PROTEIN G
5	2wvp-A	16.4	3.1	187	262	9	OUTER MEMBRANE PROTEIN G
6	4d5d-A	16.0	2.6	190	311	7	CYMA
7	4d5b-A	16.0	2.6	190	312	7	CYMA
8	4d5d-B	16.0	2.6	190	309	7	CYMA
9	4d51-A	15.9	2.6	190	312	7	CYMA
10	4d5b-B	15.9	2.6	190	309	7	CYMA
11	4v3h-B	15.9	2.6	190	307	7	CYMA PROTEIN
12	2iwv-B	15.9	2.9	191	277	6	OUTER MEMBRANE PROTEIN G
13	2iwv-C	15.9	2.9	191	277	6	OUTER MEMBRANE PROTEIN G
14	2iwv-D	15.9	2.9	190	277	6	OUTER MEMBRANE PROTEIN G
15	2iwv-A	15.9	2.9	190	277	6	OUTER MEMBRANE PROTEIN G
16	4v3g-B	15.8	2.6	191	322	6	CYMA PROTEIN
17	2x9k-A	15.8	3.2	187	278	9	OUTER MEMBRANE PROTEIN G
18	2iww-B	15.8	3.2	187	277	9	OUTER MEMBRANE PROTEIN G
19	4v3h-A	15.7	2.6	190	314	7	CYMA PROTEIN
20	4v3g-A	15.5	2.6	190	330	7	CYMA PROTEIN
21	3pgr-A	15.4	2.4	181	376	11	LONG-CHAIN FATTY ACID TRANSPORT PROTEIN
22	3bry-A	15.3	2.4	184	389	12	TBUX
23	1t16-A	15.0	2.5	181	427	11	LONG-CHAIN FATTY ACID TRANSPORT PROTEIN
24	2iww-A	14.6	3.1	187	277	8	OUTER MEMBRANE PROTEIN G

Reference

Petersen, T. N., Brunak, S., Heijne, von, G. & Nielsen, H. (2011). *Nature Methods*. **8**, 785–786.