



3D reconstruction of disc-shaped native membranes of *E. coli* containing unliganded ELIC. A) Unfiltered and unsharpened full map obtained using CryoSparc. B) Various FSC curves. The curve that includes the correction for noise overfitting ("corrected") crosses the 0.143 threshold at ~ 2.5 Å. C) Local-resolution map calculated using *MonoRes* (from the Xmipp/Scipion package). D) and E) Side and top views (parallel to the membrane and from the extracellular side, respectively) of the locally sharpened full map obtained with *LocalDeblur* (from the Xmipp/Scipion package) using the local-resolution information provided by *MonoRes*. For clarity of display, the 3D reconstructions in panels C, D, and E were masked so as to exclude the lipid disc.

Table. Cryo-EM data-collection and processing statistics of unliganded ELIC in native *E. coli* membranes

Data collection	
Grids	Quantifoil gold grids
Vitrification method	Leica GP-EMM
Microscope	Krios
Voltage (kV)	300
Magnification factor	22,500
Detector	K2 Summit
Recording mode	Counting
Total electron dose ($e^- \text{Å}^{-2}$)	56.228
Total exposure time (s)	8
Set defocus range (mm)	0.11–4.85
EM-data processing	
Number of micrographs	4639
Number of picked particles	2,215,589
Number of particles used for refinement	1,239,532
Symmetry imposed	C5
Resolution (Å)	2.5