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Supplementary Information

Elucidating a role for the cytoplasmic domain in the *Mycobacterium tuberculosis* mechanosensitive channel of large conductance

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Maps were calculated at 8 Å resolution using the Molrep program in CCP4i. Angles are Chi = 180° , 120° , 90° , 72° , 60° , and 51.7° . All the maps were calculated with the same 180° , 120° , and 90° sections to represent equal contours.



Figure S2. Rotation functions calculated for 2, 3, 4, 5, 6, and 7 fold symmetry for Native *Mt*MscL_{\(\Delta\)}C crystals (A-F, respectively).

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Figure S3. Examples of data used to generate Figure 5 in manuscript.

(A) Representative traces of tension (or pressure) induced currents in patches derived from BL21 DE3 $\Delta mscL$ cells expressing *Ec*MscL, *Mt*MscL ΔC , or *Mt*MscL full-length. The midpoint pressure for *Ec*MscS and the relevant MscL variant is indicated, as well as the point on the curve where current saturation is achieved. (B) Representative single channel traces for *Ec*MscL, *Mt*MscL ΔC , and *Mt*MscL. Gating events were observed in all of the patches shown upon application of the triangular pressure ramp (suction to the pipette) of the indicated amplitude (mm of mercury). The suction applied to the patch pipette in each experiment is indicated underneath the trace and the maximum value of negative pressure reached is indicated.



Figure S4. Crossing angle analysis of MscL structures.

A categorization of the conformational states of MscL can be made by plotting the helixhelix crossing angles between TM1-TM1' and TM1-TM2. Structures in the closed or expanded states can be differentiated by the characteristic relationships between these crossing angles in the two states. TM1-TM1' is plotted on the X axis and TM1-TM2 is plotted on the Y axis, values are represented in degrees.