

Table S1. List of parameters used in the model, related to **Figure 2A**.

| Parameter | Meaning | Value |
|----------------|---|--------------|
| g_L | Leak conductance | 0.0025 1/s |
| V_L | Rest potential | -90 mV |
| g_{C1} | Initial cation channel conductance | 0.0125 1/s |
| g_{C2} | Final cation channel conductance | 0.003125 1/s |
| δ_C | Cation channel decay rate | 0.025 1/s |
| V_0 | Cation Nernst potential when $C = C_e$ | 1 mV |
| C_e | Extracellular cation concentration | 2 mM |
| α_{in} | Import rate | 0.075 1/s |
| α_{out} | Export rate | 0.075 1/s |
| F | Conductance coefficient | 0.0067 mM/mV |
| β_T | ThT input rate | 0.01 mM/s |
| δ_{hyp} | ThT export rate under hyperpolarization | 0.00025 1/s |
| δ_{dep} | ThT export rate under depolarization | 0.005 1/s |
| V_S | Steepness of the ThT response function | 0.1 mV |

Table S2. Proteins enriched uniquely in the L22^{*} mutant, related to **Figure 5**.

Proteins marked in bold are ion transporters. The table lists a description of overexpressed proteins and its expression level in wild-type in the presence of spectinomycin (Spec), ΔyhdP, ΔL34, and L22^{*} mutant normalized to wild-type (WT) expression level.

| Protein | Description | WT | Spec | ΔyhdP | ΔL34 | L22* |
|-------------|--|----|------|-------|------|------|
| YodF | Uncharacterized symporter, similar to amino acid permease | 1 | 0.83 | 0.69 | 0.87 | 1.94 |
| MgtE | Magnesium transporter MgtE | 1 | 0.94 | 0.57 | 0.99 | 1.87 |
| YybJ | Uncharacterized ABC transporter ATP-binding protein | 1 | 0.91 | 1.04 | 0.92 | 1.77 |
| YoaB | 2-oxoglutarate permease (proton symporter) | 1 | 0.77 | 0.73 | 0.87 | 1.68 |
| YhdP | Magnesium efflux pump | 1 | 0.93 | 0.27 | 1.09 | 1.57 |
| YbeC | Probable amino acid-proton symporter | 1 | 1.06 | 0.72 | 0.97 | 1.56 |
| NtdC | Glucose-6-phosphate 3-dehydrogenase | 1 | 0.68 | 0.51 | 0.67 | 6.25 |
| NtdA | 3-oxo-glucose-6-phosphate:glutamate aminotransferase | 1 | 0.69 | 0.55 | 0.66 | 5.02 |
| NtdB | Kanosamine-6-phosphate phosphatase | 1 | 0.70 | 0.57 | 0.62 | 4.90 |
| YqjL | General stress protein, putative hydrolase involved in oxidative stress resistance | 1 | 0.92 | 1.09 | 1.00 | 2.31 |
| YdiP | Similar to DNA-3-methyladenine glycosidase II | 1 | 0.94 | 0.93 | 0.87 | 1.90 |
| EpsN | UDP-2,6-dideoxy 2-acetamido 4-keto glucose aminotransferase | 1 | 0.66 | 0.83 | 0.71 | 1.75 |
| BioD | ATP-dependent dethiobiotin synthetase | 1 | 0.87 | 1.00 | 0.94 | 1.71 |
| YokE | SPBc2 prophage-derived uncharacterized protein | 1 | 1.07 | 1.09 | 1.06 | 1.68 |
| MtnK | Methylthioribose kinase | 1 | 0.69 | 0.96 | 0.96 | 1.65 |
| PxpC | 5-oxoprolinase subunit C | 1 | 1.05 | 0.98 | 0.87 | 1.65 |
| YvyE | IMPACT family member | 1 | 1.02 | 0.97 | 1.07 | 1.57 |
| KipR | HTH-type transcriptional regulator KipR | 1 | 0.91 | 0.85 | 0.71 | 1.56 |
| RapG | Response regulator aspartate phosphatase G | 1 | 0.99 | 0.97 | 1.05 | 1.54 |
| MtnA | Methylthioribose-1-phosphate isomerase | 1 | 0.95 | 1.03 | 0.90 | 1.51 |

Table S3. Summary values of parameters measured at the population level for the conditions listed, related to **Figure 7**.

The table lists mean ThT level with its standard deviation (ThT std), hyperpolarized cell fraction and its confidence interval (CI), and number of analyzed cells.

| Condition | ThT mean (a.u.) | ThT std | Hyperpolarized cell fraction | CI | Number of cells |
|------------------------------|-----------------|---------|------------------------------|------|-----------------|
| WT | 211.39 | 20.15 | 3.68 | 0.03 | 1,459,602 |
| WT Spec | 229.56 | 42.41 | 28.54 | 0.07 | 1,404,264 |
| WT Kan | 219.89 | 34.24 | 15.76 | 0.06 | 1,298,188 |
| WT Cm | 219.44 | 41.03 | 23.41 | 0.07 | 1,415,308 |
| WT+20 mM Mg ²⁺ | 191.36 | 31.98 | 2.94 | 0.04 | 781,747 |
| L22* | 197.35 | 15.35 | 0.29 | 0.01 | 896,718 |
| L22* Spec | 197.6 | 17.05 | 0.36 | 0.01 | 1,279,746 |
| ΔL34 | 237.09 | 47.30 | 35.22 | 0.07 | 1,741,790 |
| ΔL34+100 mM Mg ²⁺ | 184.42 | 21.53 | 2.3 | 0.03 | 1,249,549 |

Table S4. Summary values of parameters measured at the single-cell level for the conditions listed, related to **Figure 7**.

The table lists the elongation rate with its standard error (SE), mean number of surviving generations with its standard error (SE), and number of analyzed cells.

| Condition | Elongation rate (h^{-1}) | SE | Surviving generations | SE | Number of cells |
|------------------------------|--|------|--------------------------|------|--------------------|
| WT | 0.19 | 0.01 | 2.85 | 0.18 | 102 |
| WT Spec | 0.06 | 0.01 | 1.22 | 0.08 | 174 |
| WT Kan | 0.19 | 0.01 | 1.56 | 0.12 | 134 |
| WT Cm | 0.04 | 0.01 | 1.31 | 0.13 | 35 |
| WT+20 mM Mg ²⁺ | 0.19 | 0.02 | 3.58 | 0.10 | 50 |
| L22* | 0.26 | 0.01 | 4.02 | 0.31 | 51 |
| L22* Spec | 0.17 | 0.01 | 2.52 | 0.14 | 100 |
| ΔL34 | 0.09 | 0.02 | 1.29 | 0.28 | 35 |
| ΔL34+100 mM Mg ²⁺ | 0.23 | 0.02 | 2.56 | 0.34 | 35 |