**Consistent mutational paths predict eukaryotic thermostability**

# Supplementary Tables

Table S1 Bacterial genomes and Optimal Growth Temperature

|  |  |
| --- | --- |
| **Species** | **OGT** |
| *Carboxydothermus hydrogenoformans* | 67 |
| *Clostridium acetobutylicum* | 37 |
| *Clostridium perfringens* | 37 |
| *Clostridium tetani* | 37 |
| *Desulfitobacterium hafniense* | 38 |
| *Moorella thermoacetica* | 57 |
| *Thermoanaerobacter tengcongensis* | 75 |

Table S2 Archaeal genomes and Optimal Growth Temperature

|  |  |
| --- | --- |
| **Species** | **OGT** |
| *Methanocaldococcus jannaschii* | 85 |
| *Methanococcus maripaludis S2* | 35 |
| *Methanopyrus kandleri AV19* | 98 |
| *Methanospirillum hungatei* | 35 |
| *Pyrococcus abyssi* | 96 |
| *Pyrococcus furiosis* | 100 |
| *Pyrococcus horikoshii* | 98 |

Table S3 correlations with OGT in bacterial and archaeal clades containing thermophiles.

|  |  |  |
| --- | --- | --- |
| **Amino acid** | **Correlation bacteria** | **Correlation archaea** |
| A | 0.32 | 0.13 |
| C | -0.81\*\* | -0.72 |
| D | -0.60\* | -0.53 |
| E | 0.37 | 0.77\* |
| F | 0.047 | 0.17 |
| G | 0.18 | -0.11 |
| H | 0.026 | -0.43 |
| I | -0.27 | -0.18 |
| K | -0.16 | 0.11 |
| L | 0.45 | 0.86\*\* |
| M | -0.69\*\* | -0.91\*\*\* |
| N | -0.53 | -0.47 |
| P | 0.50\* | 0.48 |
| Q | -0.057 | -0.70 |
| R | 0.37 | 0.50\* |
| S | -0.62\*\* | -0.98\*\*\* |
| T | -0.58 | -0.96\*\*\* |
| V | 0.68\* | 0.60 |
| W | 0.16 | 0.70 |
| Y | 0.025 | 0.35 |
| IVYWREL | 0.89\*\*\* | 0.94\*\* |

\* P < 0.1; \*\* P < 0.05; \*\*\*P < 0.005

# Supplementary figure legends

**Figure S1 Phylogenetic tree of Sordariomycetes.** A maximum likelihood tree was calculated with RaXML based on the concatenated alignments of 2,064 single copy orthologs in Sordariomycetes.Numbers on the branches indicate bootstrap support.

**Figure S2 Intergenic length distribution of *N. crassa*, *C.globosum* and *C.thermophilum****.* Intergenic regions of C. thermophilum (blue) are significantly smaller than Neurospora crassa (red) and Chaetomium globosum (green), due to genome compaction.

**Figure S3 Thermostability of Wild-type and Mutant ctArx1.** The critical temperature for thermostability is higher at lower protein concentration. The thermostability test (in vitro aggregation assay) with ctArx1 mutant proteins was performed at a 6-fold lower concentration (~1.3 mg/ml) than in **Fig. 4B**. ctArx1-nondestabilizing and ctArx1-destabilizing with five neutral or adaptive mutations, respectively (see **Figure 4A, B**), and ctArx1 wild-type recombinant proteins were affinity-purified and incubated at the indicated temperatures for 1 hour, separated into supernatant (S) and pellet (P) fractions by centrifugation and subjected to SDS-PAGE and Coomassie stain in comparison to the input (I). PS: protein standard



Figure S1



Figure S2



Figure S3