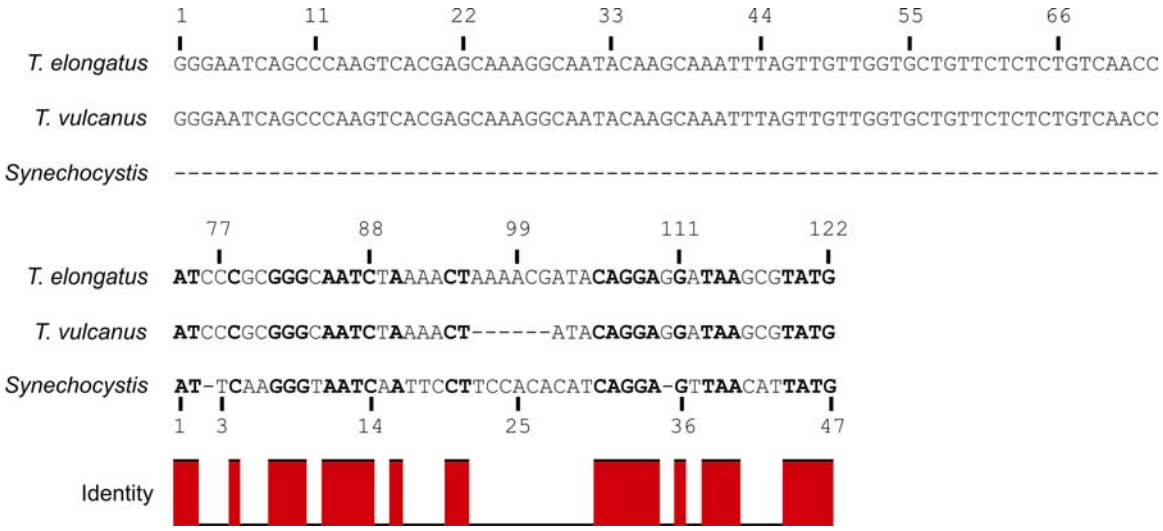


**Figure S1.** Alignment of *Synechocystis hsp17* 5'-UTR (thermometer element) with the 5'-UTRs of *T. elongates* and *T. vulcanus*. Identical nucleotides are indicated by bold letters.

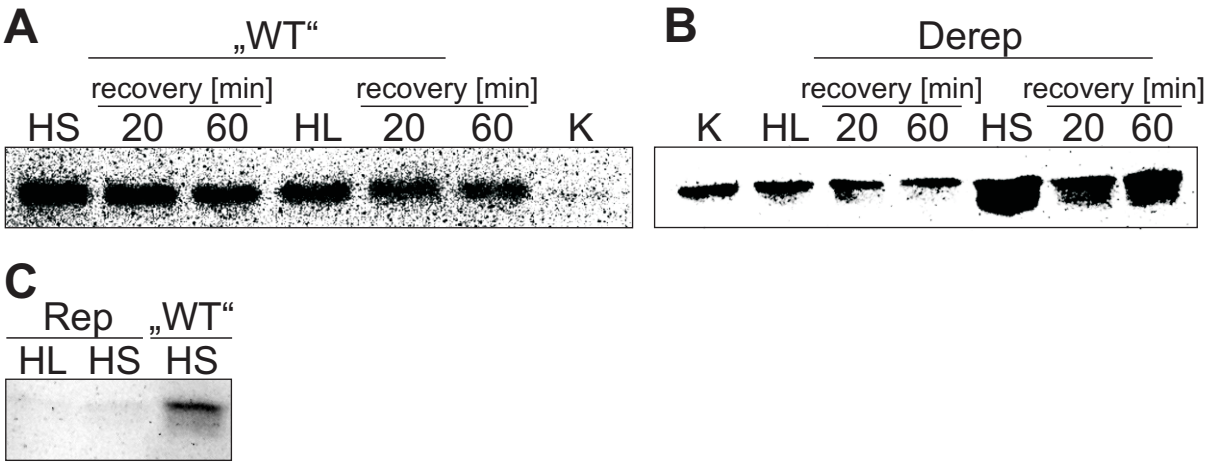


**Figure S2.** Comparative Western blot analysis of Hsp17 protein levels after heat shock (HS) and high light stress (HL) and in the recovery phase. (A) *Synechocystis* „WT“ and (B) Derep cells were incubated at 42°C (for 60 min) or stressed with high light (for 30 min). Following the different stress conditions, the cultures were transferred to 28°C and LL conditions (recovery phase). Total protein was extracted directly after stress (HS or HL) and at time points during recovery phase as indicated. K, control (total protein extracted from cells grown at 28°C and low light). (C) Total protein was extracted from *Synechocystis* Rep cells incubated at 42°C or under high light conditions and blotted beside total protein originating from heat shocked „WT“ cells. Hsp17 protein was detected via monoclonal α-Hsp17 antibody. Equal amounts of total protein were checked by Coomassie-stained SDS-PAGE gels (data not shown).

**Table SI.** Strains, plasmids and oligonucleotides used in this study

| Strain, plasmid or oligonucleotide | Relevant characteristic(s) or sequence[a] | Source or reference |
|---|---|---|
| **Strains** | | |
| *E. coli* DH5α | *supE44 ΔlacU169* (Φ80*lacZ*ΔM15) *hsdR17 recA1 gyrA96* | Gibco-BRL |
| *E. coli* DH5αZ1 | Δ(*argF-lac*)I69φ 80Δ*lac*Z58(M15) *gln*V44 (AS) *rfb*D1 *gyr*A96 ($Nal^R$) *rec*A1 *end*A1 *sρo*T1 *thi*-1*hsdRI7* ZI (*lacR tetR* $Sp^R$) | Lutz and Bujard, 1997 |
| *Synechocystis* sp. PCC6803 HK-1 | Δ *hsp17* ($Km^R$) | Kosaka and Fukuzawa |
| **Plasmids** | | |
| pBAD-bgaB (pBO415) | Translational *bgaB* fusion vector, *bgaB*: heat-stable β-galactosidase, $Amp^R$ | Waldminghaus et al., 2007 |
| pXG-10 | Translational *gfp* fusion vector, $Cm^R$, $P_{LTet-O}$ Promotor | Urban and Vogel, 2006 |
| pNaive.16 (pAZ877) | Vector for integration into the *hsp17* locus, Restriction sites used for subcloning: BamHI (upstream region); CpoI (*hsp17* coding region), $Spec^R$, $Amp^R$ | Giese and Vierling, 2002 |
| pBO1292 | *Synechocystis hsp17* 5'-UTR (thermometer element) in pUC18 (QuikChange® template) | This study |
| pBO1347 | fragment of ORF sll1514 containing *hsp17* thermometer element in pUC18, (pNaive-QuikChange® template) | This study |
| pBO1293 | *Synechocystis hsp17* 5'-UTR-*bgaB* fusion in pBAD-bgaB | This study |
| pBO1312 | *Synechocystis hsp17* 5'-UTR-AAC39-41G-*bgaB* fusion in pBAD-bgaB (M1-rep) | This study |
| pBO1310 | *Synechocystis hsp17* 5'-UTR-T17C-*bgaB* fusion in pBAD-bgaB (M2) | This study |
| pBO1311 | *Synechocystis hsp17* 5'-UTR-T22G-*bgaB* fusion in pBAD-bgaB (M3) | This study |
| pBO1316 | *Synechocystis hsp17* 5'-UTR-C19T-*bgaB* fusion in pBAD-bgaB (M4.1) | This study |
| pBO1314 | *Synechocystis hsp17* 5'-UTR-CC19/20GA-*bgaB* fusion in pBAD-bgaB (M4.2) | This study |
| pBO1315 | *Synechocystis hsp17* 5'-UTR-CC19/20AG-*bgaB* fusion in pBAD-bgaB (M4.3) | This study |
| pBO1313 | *Synechocystis hsp17* 5'-UTR-CC19/20GG-*bgaB* fusion in pBAD-bgaB (M4.4-derep) | This study |
| pBO602 | *Salmonella agsA-bgaB* fusion in pBAD-bgaB (Control C.1) | Waldminghaus et al., 2007 |
| pBO1056 | *E. coli gyrA-bgaB* fusion in pBAD-bgaB (Control C.2) | Waldminghaus et al., 2007 |
| pBO1325 | *Synechocystis hsp17* 5'-UTR-*gfp* fusion in pXG-10 | This study |
| pBO1801 | *Synechocystis hsp17* 5'-UTR-*rep-gfp* fusion in pXG-10 | This study |
| pBO1802 | *Synechocystis hsp17* 5'-UTR-*derep-gfp* fusion in pXG-10 | This study |
| pBO1301 | *Synechocystis hsp17* 5'-UTR-WT-*in vitro* transcripts for structure probing experiments, runoff vector (via MlsI site), pUC18-based | This study |

| | | |
|---|---|---|
| pBO1304 | *Synechocystis hsp17* 5'-UTR-rep-*in vitro* transcripts for structure probing experiments, runoff vector (via MlsI site), pUC18-based | This study |
| pBO1302 | *Synechocystis hsp17* 5'-UTR-derep-*in vitro* transcripts for structure probing experiments, runoff vector (via MlsI site), pUC18-based | This study |
| pBO1305 | *Synechocystis hsp17* 5'-UTR-WT-*in vitro* transcripts (+63 bp coding region) for toeprinting experiments, runoff vector (via HpyCH4V site), pUC18-based | This study |
| pBO1349 | *Synechocystis hsp17* 5'-UTR-rep-*in vitro* transcripts (+63 bp coding region) for toeprinting experiments, runoff vector (via HpyCH4V site), pUC18-based | This study |
| pBO1348 | *Synechocystis hsp17* 5'-UTR-derep-*in vitro* transcripts (+63 bp coding region) for toeprinting experiments, runoff vector (via HpyCH4V site), pUC18-based | This study |
| pB01834 | pNaive- *hsp17* 5'-UTR-WT (pNaive.16 derivate) | This study |
| pBO1806 | pNaive- *hsp17* 5'-UTR-rep (pNaive.16 derivate) | This study |
| pBO1807 | pNaive- *hsp17* 5'-UTR-derep (pNaive.16 derivate) | This study |

**Oligonucleotides**

| | |
|---|---|
| NheI-*hsp17*therm | TTTGCTAGCATTCAAGGGTAATCAA |
| | (*pBO1293) |
| *hsp17*therm-EcoRI | TTTGAATTCAGACATAATGTTAACTCC |
| | (*pBO1293) |
| *hsp17*therm-M1-fw | CACACATCAGGAGTTGATTATGTCTGAATTC |
| | (*pBO1312, pBO1801, pBO1304 and pBO1806) |
| *hsp17*therm-M1-rv | CTCCTGATGTGTGGCAGGAATTGATTACCC |
| | (*pBO1312, pBO1801, pBO1304 and pBO1806) |
| *hsp17*therm-M2-fw | CAAGGGTAATCAACTCCTTCCACACATCAGG |
| | (*pBO1310) |
| *hsp17*therm-M2-rv | CCTGATGTGTGGAAGGAGTTGATTACCCTTG |
| | (*pBO1310) |
| *hsp17*therm-M3-fw | GGGTAATCAATTCCTGCCACACATCAGGAG |
| | (*pBO1311) |
| *hsp17*therm-M3-rv | CTCCTGATGTGTGGCAGGAATTGATTACCC |
| | (*pBO1311) |
| *hsp17*therm-M4-fw | CAAGGGTAATCAATTRRTTCCACACATCAGG |
| | (*pBO1316, pBO1314, pBO1315 and pBO1313) |
| *hsp17*therm-M4-rv | CCTGATGTGTGGAARRAATTGATTACCCTTG |
| | (*pBO1316, pBO1314, pBO1315 and pBO1313) |
| *hsp17*therm-der.-fw | CAAGGGTAATCAATTGGTTCCACACATCAGG |
| | (*pBO1802, pBO1302 and pBO1807) |

| | |
|---|---|
| *hsp17*therm-der.-rv | CCTGATGTGTGGAACCAATTGATTACCCTTG |
| | (*pBO1802, pBO1302 and pBO1807) |
| PstI-rep-fw | TTTCTGCAGATTCAAGGGTAATCAATTGG |
| | (*pBO1802) |
| derep-NheI-rv | AAAAGCTAGCAGACATAATCAACTCCTG |
| | (*pBO1801) |
| T7- *hsp17*therm | GAAATTAATACGACTCACTATAGGGATTCAAGGGTAATCAATTCC (*pBO1301 and pBO1305) |
| *hsp17*therm-runoff-SP | TGGCCAGACATAATGTTAACTCCTG |
| | (*pBO1301) |
| *hsp17*therm-runoff-toeprinting | TGCAAACAGTTGGTTCATCTGCTGCTGG |
| | (*pBO1305) |
| *hsp17*therm-ORF-fw | TTGAATTCCATTATTGCCGGGGCCGTC |
| | (*pBO1347) |
| *hsp17*therm-ORF-rv | TTCCGTGGCGGTCCGTAGGGAC |
| | (*pBO1347) |
| rep-toeprinting-fw | CACATCAGGAGTTGATTATGTCTCTCATTC |
| | (*pBO1349) |
| rep-toeprinting-rv | GAATGAGAGACATAATCAACTCCTGATGTG |
| | (*pBO1349) |
| derep-toeprinting-fw | CAAGGGTAATCAATTGGTTCCACACATCAGG |
| | (*pBO1348) |
| derep-toeprinting-rv | CCTGATGTGTGGAACCAATTGATTACCCTTG |
| | (*pBO1348) |
| fw-hsp17-probe | TGTCTCTCATTCTTTACAAT (Northern blot probe) |
| rv-hsp17-probe | TAATACGACTCACTATCATTTATTAGGAAAGCTGAAC (Northern blot probe) |
| fw-bgaB-probe | AGAGCAATGGCCAGAGGAAA (Northern blot probe) |
| rv-bgaB-probe | TAATACGACTCACTATAGATCGGCAAAGAATCTGGAT (Northern blot probe) |
| up-fw | GCGGCTAGAAATGTAATTTCGGCAATC (segregation analysis via PCR) |
| cr-rv | GTTAGGATACCGGCATCGTAATTAGC (segregation analysis via PCR) |
| cr-fw | GCTAATTACGATGCCGGTATCCTAAC (segregation analysis via PCR) |
| down-rv | GACAACTTTTTCAGCAGTCCATTCCCATGG (segregation analysis via PCR) |

[a] Introduced restriction sites are underlined. * Corresponding plasmids

**Table SII.** Absolute levels of *bgaB* fusion constructs

| *bgaB* Fusion Construct | T = 28°C | T = 42°C | Fold Induction |
|---|---|---|---|
| WT (Full-Length *hsp17* 5' UTR) | 30 +/-5.1 | 180 +/-8.8 | 6.0 |
| M1 (AAC39-41G) | 0 +/-0 | 0 +/-0 | 0.0 |
| M2 (U17-C) | 15 +/-6.3 | 40 +/-7.0 | 2.6 |
| M3 (U22-G) | 15 +/-7.2 | 75 +/-5.9 | 5.0 |
| M4.1 (C19U) | 110 +/-7.9 | 275 +/-7.2 | 2.5 |
| M4.2 (CC19/20GA) | 125 +/-8.4 | 350 +/-8.9 | 2.8 |
| M4.3 (CC19/20AG) | 175 +/-9.8 | 690 +/-9.7 | 3.9 |
| M4.4 (CC19/20GG) | 200 +/-9.7 | 790 +/-9.9 | 3.9 |
| C.1 (*Salmonella agsA* 5' UTR) | 48 +/-5.3 | 350 +/-5.7 | 7.8 |
| C.2 (*E. coli gyrA* 5' UTR) | 18 +/-2.1 | 41 +/-4.8 | 2.3 |