

Supplementary Materials

Identification of 5'-untranslated regions that function as effective translational enhancers in monocotyledonous plant cells using a novel method of genome-wide analysis

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Table S1. Gene-specific primers for construction of plasmid pBlue-35S-OsADH_5'UTR-FL-HSPT

5'UTR name	Primer sequence (5' to 3') ^a
<i>OsADH</i>	TATCGATGGAGAGAAGAATTCCAAGCAACG
	TGACGTC ^a TCCATTAATCCCCCTCTTTTTC

^a *Cla*I and *Aat*II sites are shown in red and blue, respectively.

Table S2. Gene-specific primers for construction of plasmid pBlue-35S-test_5'UTR-FL-HSPT

5'UTR ID ^a	Primer sequence (5' to 3') ^b
Os10t0530900-01_00076	AAGGAAGTTCATTATCGATGGAGAGAAATAAGCAAACCAAACAAAAAAC TTCTTTATGT TTTTGACGTCTTCCATGCCAAGTCTCTCGTTCTAGATC
Os04t0556300-01_00118	AAGGAAGTTCATTATCGATGGAGAGAAACGTCTCGTCTCTGTCTCTCG TTCTTTATGTTTTTGACGTCTTCCATGCTGGAGCGAGCGGCGGCCG
Os03t0180400-01_00112	AAGGAAGTTCATTATCGATGGAGAGAAATTTTGGTTTATTTTGCCTCC TTCTTTATGTTTTTGACGTCTTCCATCGTCGCCGATTCTCGCCGCC
Os11t0592100-01_00063	AAGGAAGTTCATTATCGATGGAGAGAAATTTCACTAGCATCTAGCAAGTG TTCTTTATGTTTTTGACGTCTTCCATCTCACCTAAG GTGGTTACTACG
Os05t0119200-01_00064	AAGGAAGTTCATTATCGATGGAGAGAAAGTTTTCTCTCTGATTTTC TTCTTTATGTTTTTGACGTCTTCCATCGTCGTCTGA TCAAGAAATC
Os01t0303800-01_00106	AAGGAAGTTCATTATCGATGGAGAGAAATCTCTCCACTTCTCCAACCAATTGC TTCTTTATGTTTTTGACGTCTTCCATCACTCTCGACAGCAGCTAGCTAG
Os06t0147400-01_00074	AAGGAAGTTCATTATCGATGGAGAGAAAGTTTTGAGTGCTGCGAACGCAG TTCTTTATGTTTTTGACGTCTTCCATGGTCCCTCGCCGTGAAGGG
Os02t0684500-00_00094	AAGGAAGTTCATTATCGATGGAGAGAAACCAAGAGCCACAGCGATTTTCAG TTCTTTATGTTTTTGACGTCTTCCATCGCCGAGACCGAGACGAGACCGAG
Os08t0117400-01_00160	AAGGAAGTTCATTATCGATGGAGAGAAACTTTGTTCCCCACCACAGCAA TTCTTTATGTTTTTGACGTCTTCCATGATCCGATCCGATCCGATTCCG
Os01t0559000-01_00115	AAGGAAGTTCATTATCGATGGAGAGAAAGGGAGTGAAGAGCGAATCAAACAC TTCTTTATGTTTTTGACGTCTTCCATCCTCTCCCCCTCTTCTCCCC
Os07t0230700-01_00066	AAGGAAGTTCATTATCGATGGAGAGAAATTAACCTGACGCCACCACCTCTC TTCTTTATGTTTTTGACGTCTTCCATTTTTAGTTAGATTGTTCGGTGAATGGAAG
Os08t0136800-01_00129	AAGGAAGTTCATTATCGATGGAGAGAAAGAACACACAACCCAGCTATAGCCAC TTCTTTATGTTTTTGACGTCTTCCATGGATGGCGCCAATTATGTA CTCTCAC
Os04t0583600-01_00068	AAGGAAGTTCATTATCGATGGAGAGAAACAAGGCTTCTCCACATCTCCACC TTCTTTATGTTTTTGACGTCTTCCATCGCCGCAGCTAACCGCTCCG
Os06t0342200-01_00113	AAGGAAGTTCATTATCGATGGAGAGAACAGAAAACCGCATCCC GCAACTC TTCTTTATGTTTTTGACGTCTTCCATGGCTTGGCTACCGGCGGCCG
Os08t0136800-01_00116	AAGGAAGTTCATTATCGATGGAGAGAAAGCTATAGCCACCTACATCTGCTGATC TTCTTTATGTTTTTGACGTCTTCCATGGATGGCGCCAATTATGTA CTCTCAC

^a Candidate 5'UTR IT consists of Gene ID from IRGSP-1.0 and TSS ID, defined based on TSS position.

^b Homologous recombination regions of plasmid pBlue-35S-FL-HSPT are shown in red.

Table S3. Gene-specific primers for construction of plasmid pBlue-35S-test_5'UTR-RL-HSPT

5'UTR name and ID ^a	Primer sequence (5' to 3') ^b
<i>OsADH</i>	AAAAGAGGGGGATTAATGGCTTCCAAGGTGTACGA TCTTCATCTTCATATGAGCTCACTAGTTTACTGCTCGTTCTTCAGCA
Os02t0684500-00_00094	TCTCGGTCTCCGGCGATGGCTTCCAAGGTGTACGA TCTTCATCTTCATATGAGCTCACTAGTTTACTGCTCGTTCTTCAGCA
Os08t0136800-01_00129	TAATTGGCGCCATCCATGGCTTCCAAGGTGTACGA TCTTCATCTTCATATGAGCTCACTAGTTTACTGCTCGTTCTTCAGCA
Os04t0583600-01_00068	CGGTTAGCTGCGGCGATGGCTTCCAAGGTGTACGA TCTTCATCTTCATATGAGCTCACTAGTTTACTGCTCGTTCTTCAGCA
Os06t0342200-01_00113	GCCGGTAGCCAAGCCATGGCTTCCAAGGTGTACGA TCTTCATCTTCATATGAGCTCACTAGTTTACTGCTCGTTCTTCAGCA

^a Candidate 5'UTR ID consists of Gene ID from IRGSP-1.0 and TSS ID, defined based on TSS position.

^b Homologous recombination regions of plasmid pBlue-35S-test_5'UTR-RL-HSPT are shown in red.

Table S4. Test 5'UTR sequences

5'UTR ID ^a	5'UTR sequences	5'UTR length
Os10t0530900-01_00076	ATAAGCAAACCAAACAAAAAACCTCACCATCATCGACCCACTTGTTCCTAGCGATCT AGAACGAGAGACTTGGCA	76
Os04t0556300-01_00118	ACGTCTCGTCTCTGTCTCTCGGCCACCGCTTCCATCGTTCGTCTCCACGCTAC CGTTTCGCAACCACCCGCTTCTCCTCCGGAGACCGCTCGGCCGCCGCTCGCTCCAGC	118
Os03t0180400-01_00112	ATTTTGGTTTATTTTGCCTCCCCAACCAAGCACAAACACAAGCCTCGCCGTATTCTCT CGGAGCAAAGCGAACGCGAAACCCACCCGCGGCGGAGAAATCCGGCGACG	112
Os11t0592100-01_00063	ATTTACTAGCATCTAGCAAGTGCACAACACATCTATAAATCGTAGTAACCACCTTAG GTGAG	63
Os05t0119200-01_00064	AGTTTTCTCCTCTGATTTTCCACCACAAGATCTCTAGTTTCTTTGATTTCTTGATCAGAC GACG	64
Os01t0303800-01_00106	ATCTCTCCACTTCTCCAACCAATTGCCGATTCATCGATCTCTCATCGTTCGTTTCGCTCT AGCTAGGTTGAAGACTCGGGAGTCTAGCTAGCTGCTGTCGAGAGTG	106
Os06t0147400-01_00074	GTTTGTGAGTGCTGCGAACGCAGAGACTAGACTTTTGACCACCGCGTGAGTCCAACCT TCACGGCGAGGGAACC	74
Os02t0684500-00_00094	ACCAAGAGCCACAGCGATTTCAGATCTCCCCAAGTAGTAGACTTCTCTCTTCTCCATC TCGGATTCTGCTCGGTCTCGTCTCGGTCTCCGGCG	94
Os08t0117400-01_00160	ACTTTGTTCCCCACCACAGCAAGCGCAACGCGCAAAGAGGCAGACGCGCAGCTGAGG AGGAGAAGCGCACGCAGCCACCGCGAGATCTCCTCACCTCCATCTCCATCTCCATCTC CATCTCCGGCGGCGAGATCCCCACCGAATCGGATCGGATCGGATC	160
Os01t0559000-01_00115	AGGGAGTGAAGAGCGAATCAAACACCACCACCATCGCCAGCGTCATTAGATTTCAG AGGGAGGGAGGGAGGGAGAGAGAAAGGAAGGGGAAGTGGGGGAGAAGGAGGGGAG AGG	115
Os07t0230700-01_00066	AATTAACCTGACGCCACCACCTCTCTTTCCCTCTCTCTCTTCCATTACCGACAATCTA ACTAAAA	66
Os08t0136800-01_00129	GAACACACAACCCAGCTATAGCCACCTACATCTGCTGATCGACATCGATCGATCTCTA CTACTCCAGTAGCTAGCAAGAATTGAATCTAGGTGAAGTGAGAGTGTGAGAGTACAT AATTGGCGCCATCC	129
Os04t0583600-01_00068	ACAAGGCTTCTTCCACATCTCCACCCAAATTCCAAATCTCTCGGTGATCGGAGCGGTT AGCTGCGGCG	68
Os06t0342200-01_00113	CAGAAAAACCGCATCCCGCAACTCCACCGCTCCCAATCTCGATCGAAACGCGAGGGTT TCGCATCTCCGCCGACGCCTAGAGGAAGAAGGGATCCGCCGCCGGTAGCCAAGCC	113
Os08t0136800-01_00116	AGCTATAGCCACCTACATCTGCTGATCGACATCGATCGATCTCTACTACTCCAGTAGCT AGCAAGAATTGAATCTAGGTGAAGTGAGAGTGTGAGAGTACATAATTGGCGCCATCC	116

^a Candidate 5'UTR name consists of Gene ID from IRGSP-1.0 and TSS ID, defined based on TSS position.

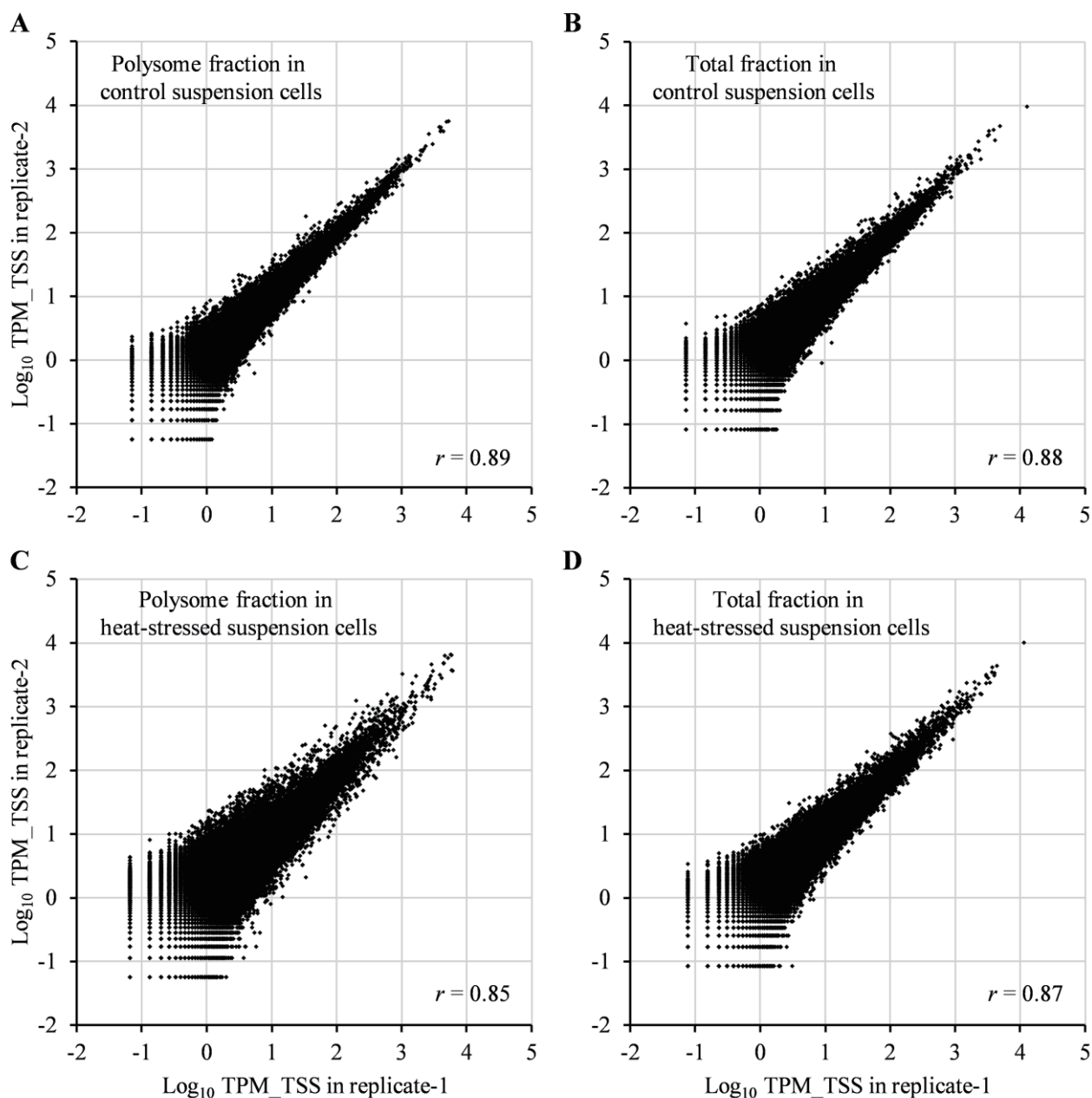


Figure S1. Reproducibility of two biological replicates of CAGE data

Scatter plots of \log_{10} -transformed TPM_TSS in polysome fraction in control suspension cells (A), total fraction in control suspension cells (B), polysome fraction in heat-stressed suspension cells (C), and total fraction in heat stressed suspension cells (D). Plots represent 418,092 (A), 395,785 (B), 384,135 (C), and 358,860 (D) transcripts from each TSS. Pearson correlation coefficients of each pair of data sets are shown.

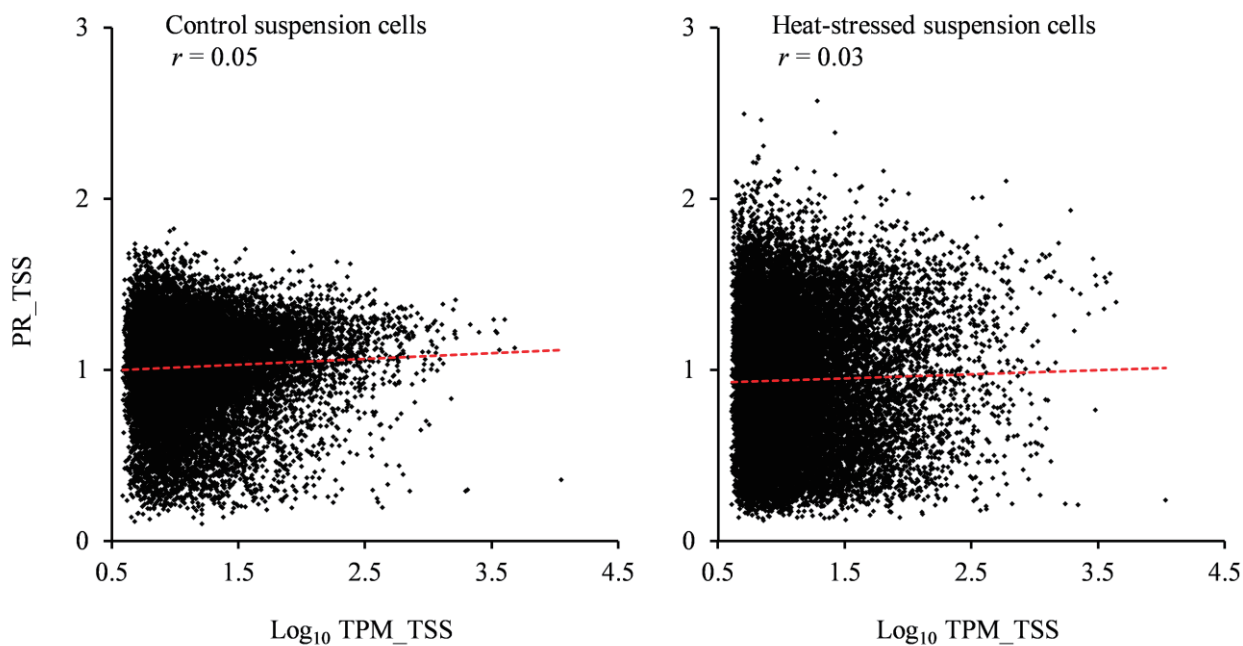


Figure S2. Relationship between accumulated mRNA level and polysome association

Scatter plots between log₁₀-transformed TPM_TSS (a reflection of accumulated mRNA level) and PR_TSS (a reflection of polysome association). Both plots represent 19,917 transcripts derived from multiple TSSs in 7,048 genes. Pearson correlation coefficients and regression line (red dotted line) for of each pair of data sets are shown.