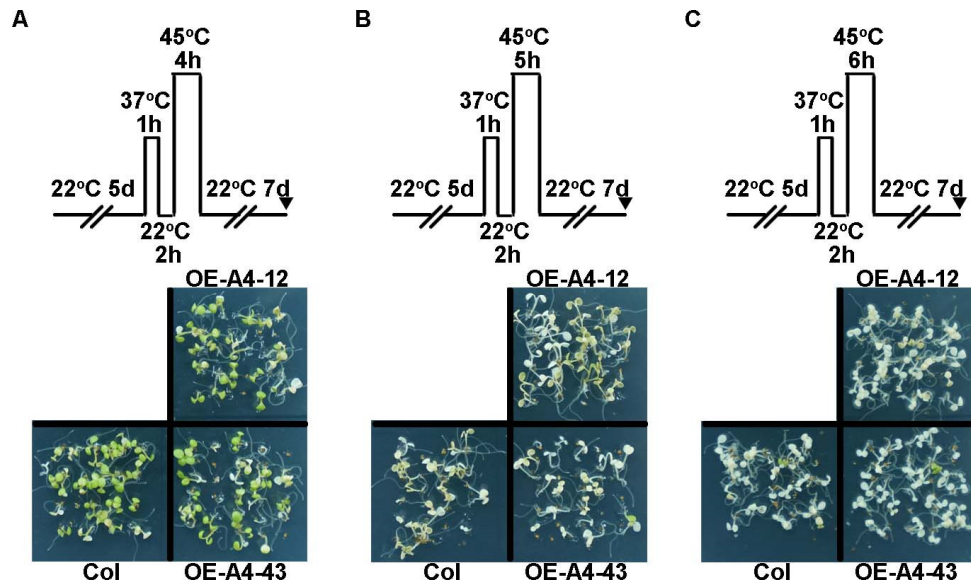
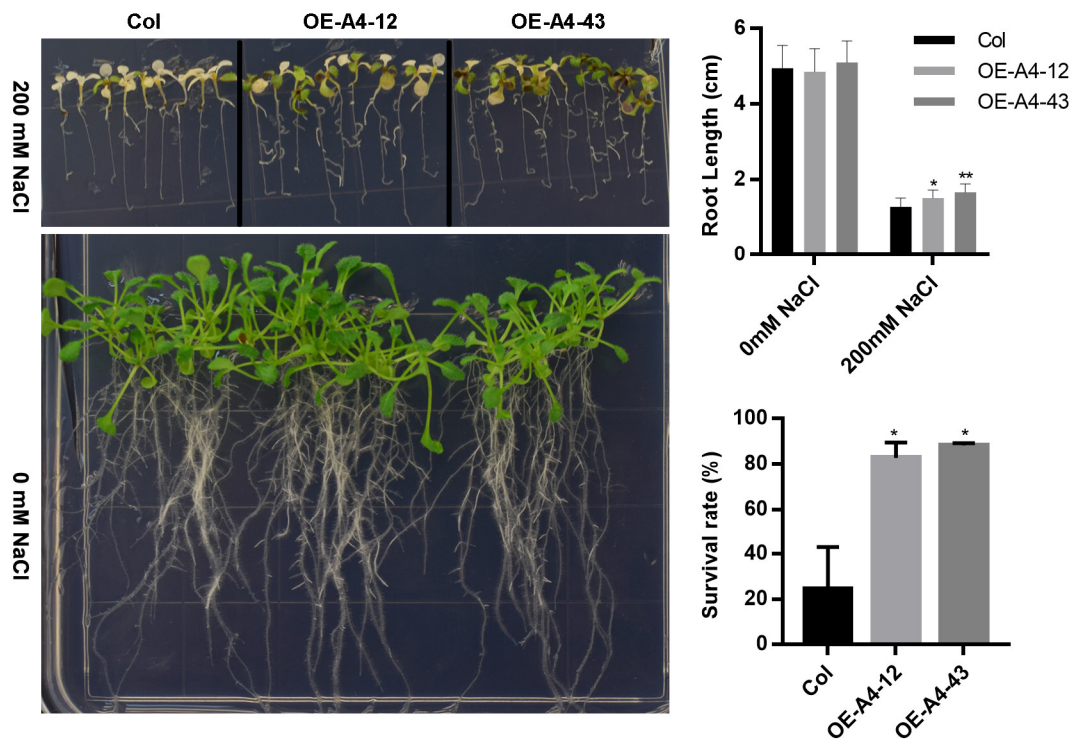




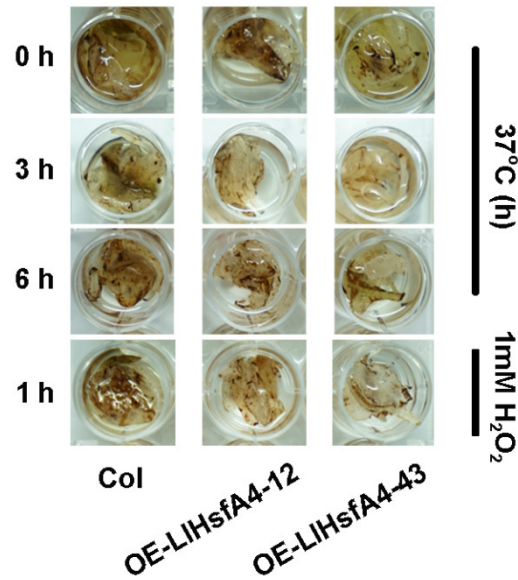
Supplementary Figure S1. Phylogenetic tree of LIHsfA4 and all Hsfs in different monocotyledonous plants and three non-plant organisms. This tree was constructed by ClustalW 2.0 and MEGA 5.0 software. The dark dot means LIHsfA4 protein.



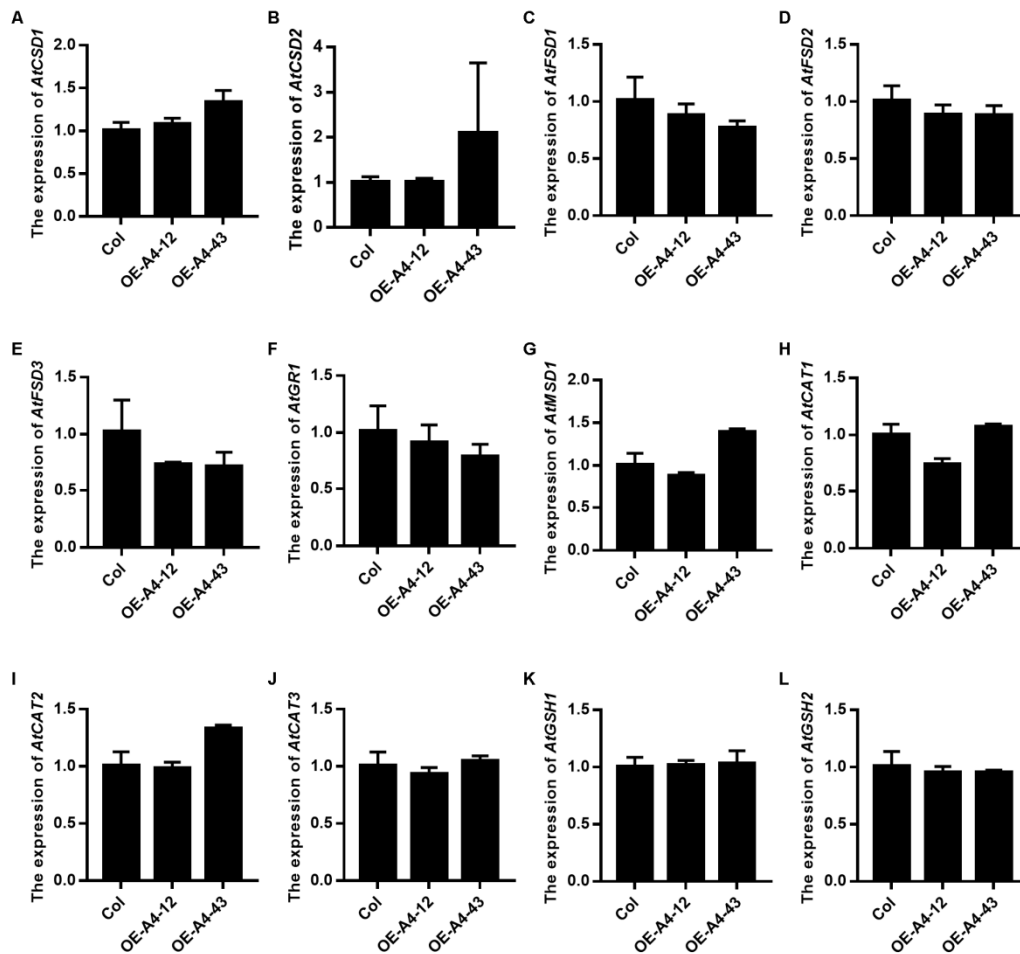
Supplementary Figure S2. Transgenic Arabidopsis plants of LIHsfA4 did not enhance acquired thermotolerance. 5-day old seedlings of two transgenic lines and wild type were treated with 37 °C for 1h, then recovery for 2 h at 22°C, and then subjected to 45°C with different times as 4 h (A), 5 h (B) and 6 h (C) to examine acquired thermotolerance.



Supplementary Figure S3. Transgenic Arabidopsis plants of LIHsfA4 enhanced salt tolerance. 5-day old seedlings of two transgenic lines and wild type were transferred into MS medium containing 200 mM NaCl for 10 days before being photographed. T-test analysis of variance was employed to identify treatment means that differed statistically. Samples with different letters are significantly different: * $p < 0.05$, ** $p < 0.01$.



Supplementary Figure S4. Content of H₂O₂ decreased in transgenic Arabidopsis plants of LIHsfA4. 10-day old seedlings of two transgenic lines and wild type were treated with 1 mM H₂O₂ for 1 h or heat (37 °C) with different time and then used for DAB staining.



Supplementary Figure S5. Expression of genes related with ROS metabolism was examined in LIHsfA4 transgenic plants. The mRNA level of *AtSODs*, *AtCATs*, *AtGSH*, and *AtGRs* were detected in wild type (Col) and transgenic lines using qRT-PCR. Expression value of these genes in Col was setting as 1 for comparison and *AtACTIN2* was used as an

internal standard. 10-day-old seedlings were collected for RNA extracting. Three independent experiments were performed each with three technical replicates and showing one experiment result. Unmarked means non-significance..

Table S1. Primers used in experiments.

Primer name	Primer sequence	Usage
<i>LIHsfA4</i> -Clone-F	5'TTYAAGCACARCAACTTCTCC 3'	Used for conserved sequence cloning
<i>LIHsfA4</i> -Clone-R	5'AGAAAYTGTTSCCARAABACATC 3'	
<i>LIHsfA4</i> -3'-F1	5'CAGAATCGTATGGAGACCTCTGTATCAGT 3'	3'RACE
<i>LIHsfA4</i> -3'-F2	5'CTTAATGAGCTTCGTGCCTCGTCTGAG 3'	
<i>LIHsfA4</i> -5'-R1	5'TCCAGTACCTGCATCTGCCGATC 3'	5'RACE
<i>LIHsfA4</i> -5'-R2	5'CTGCAGCTCGGATATCAAAGACCCC 3'	
<i>LIHsfA4</i> -F	5' GAGTCAAACCTCCTCTCTGCTTCTTCTCC 3'	LIHsfA4 full-length cDNA clone
<i>LIHsfA4</i> -R	5' GGAACAACATCAAGTCAGTACGCTAGAAAGG 3'	
<i>LIHsfA4</i> -SP1	5' ATATACGTGTACACAATCCAACCTCACC 3'	Used for cloning of <i>LIHsfA4</i> promoter.
<i>LIHsfA4</i> -SP2	5' AGCAATCTGGCCAACTGAAGATGATTTAATAC 3'	
<i>LIHsfA4</i> -SP3	5' CCACGTCAGAAATTCCAATAGAGATGATCCATG 3'	
<i>LIHsfA4</i> -F- <i>Xba</i> I-1300	5' GCTCTAGAATGGACCCCTCGCAGGGGGC 3'	Used for <i>pSuper::LIHsfA4</i> -GFP vector construction.
<i>LIHsfA4</i> -R- <i>Kpn</i> I-1300	5' GGGGTACCAGTTCTCTGTGCAGAAGCAAGATGCCC 3'	
<i>LIHsfA4</i> -F- <i>Pst</i> I-1391	5' AACTGCAGTTGTAGATGACTTTGAGAACTTGTCTATAG 3'	Used for <i>pHsfA4::GUS</i> vector construction.
<i>LIHsfA4</i> -F- <i>Xma</i> I-1391	5' TCCCCCGGGGGCGATCGGATCGATCGGAAC 3'	
<i>LIHsfA4</i> -PGBK-F- <i>Nde</i> I	5' GGAATTCCATATGATGGACCCCTCGCAGGGGGC 3'	Used for pBD-LIHsfA4 vector construction.
<i>LIHsfA4</i> -PGBK-R- <i>Bam</i> HI	5' CGGGATCCTTAAGTTCTCTGTGCAGAAGCAAGATGCCC 3'	

<i>LlHsfA4</i> -D1-R	5' CGGGATCCTTACACCCCAGTGGTCGCAGC 3'	
<i>LlHsfA4</i> -D2-R	5' CGGGATCCTTAGCTTTTCAGCGTAATTCCTCGACTC 3'	
<i>LlHsfA4</i> -D3-R	5' CGGGATCCTTACGAAGGCACACGAGGCAAG 3'	
<i>LlHsfA4</i> -D4-R	5' CGGGATCCTTACATACGATTCTGACTCTCTGTATCAGC 3'	
<i>LlHsfA4</i> -RT-F	5' CCGAGGGAGTATTTGGTGGAAC 3'	qPCR of <i>LlHsfA4</i> .
<i>LlHsfA4</i> -RT-R	5' CTCAAAATAGATGGCCACAGAAGAAG 3'	
<i>18S rRNA</i> -F	5' CTGAATCAGGATTGGATATCTGAGG 3'	qPCR of the <i>18S rRNA</i> .
<i>18S rRNA</i> -R	5' AACTAGGTTACTGTCACTGGATAAC 3'	
<i>AtActin2</i> -F	5' AGGAACTGGATCTGGTATGGGAACAT 3'	
<i>AtActin2</i> -R	5' GCAAATCCAGCCTTCACCAT 3'	
<i>AtHsp17.6</i> -F	5' CCAAAGAAAAAGCCAAGAAGC 3'	
<i>AtHsp17.6</i> -R	5' TGGAAACCTTCCAAACTCCA 3'	
<i>AtZat6</i> -F	5' GTGACCTTGACCTGCCTTCTTC 3'	
<i>AtZat6</i> -R	5' CTCCGGCAGATTGAGTAAGC 3'	Primers used for qPCR
<i>AtWRKY30</i> -F	5' AGAGCGATGATTCCGATCAAG 3'	
<i>AtWRKY30</i> -R	5' CATCGTCCAGCGTTCTATCAA 3'	
<i>AtMBF1c</i> -F	5' AGCAGATACCCAGGAGCAGT 3'	
<i>AtMBF1c</i> -R	5' TTCGGATCGCGTAGGTCTTG 3'	
<i>AtHsp25.3</i> -F	5' GATCAAGATGCGTTTCGACAT 3'	

<i>AtHsp25.3</i> -R	5' TTCTACAGAGATTTTGACGTCTTCTT 3'
<i>AtGolS1</i> -F	5' AGCCACCGGCTCTTTACTTC 3'
<i>AtGolS1</i> -R	5' GTTCAGCGAAAGGAGTCGGA 3'
<i>AtHsp22</i> -F	5' ACTACTCCAGGCAGCTTGCTA 3'
<i>AtHsp22</i> -R	5' CTTGAATGGATCAGGGAACC 3'
<i>AtZat12</i> -F	5' GACGCTTTGTCTGTCTGGATT 3'
<i>AtZat12</i> -R	5' GTGTCCTCCCAAAGCTTGTC 3'
<i>AtAPX1</i> -F	5' GTCCATTTCGGAACAATGAGGTTTGAC 3'
<i>AtAPX1</i> -R	5' GTGGGCACCAGATAAAGCGACAAT 3'
<i>AtAPX2</i> -F	5' TGATGTGAAGACGAAGACAGGAGGAC 3'
<i>AtAPX2</i> -R	5' CCCATCCGACCAAACACATCTCTTA 3'
<i>AtAPX3</i> -F	5' CCCAAAATCACATACGCAGACCTGTA 3'
<i>AtAPX3</i> -R	5' AGTTGTCAAACCTTCAGCGGCTCTTG 3'
<i>AtAPX4</i> -F	5' CTACTAAATCCGGGGGAGCCAATG 3'
<i>AtAPX4</i> -R	5' CTCTGTTGCATCACTCCTTCCAAAAT 3'
<i>AtAPX5</i> -F	5' AGCTAAACCGTCCACACAACAAAGGT 3'
<i>AtAPX5</i> -R	5' GTCCCAAAGTGTGACCTCCAGAGAGA 3'
<i>AtAPX6</i> -F	5' TGCAAAACGAAATAAGGAAAAGTGGTG 3'
<i>AtAPX6</i> -R	5' CACTCAGGGTTTCTGGAGGTAGCTTG 3'

<i>AtsAPX-F</i>	5' TGCTAATGCTGGTCTTGTGAATGCTT 3'
<i>AtsAPX-R</i>	5' CCACTACGTTCTGGCCTAGATCTTCC 3'
<i>AttAPX-F</i>	5' CAGAATGGGACTTGATGACAAGGAAA 3'
<i>AttAPX-R</i>	5' ATGCAGCCACATCTTCAGCATACTTC 3'
<i>AtCSD1 -F</i>	5' AGTAACCAAAGAGAGACGAAGCA 3'
<i>AtCSD1 -R</i>	5' CCTTCCTGGGTGAAAAAGATAG 3'
<i>AtCSD2 -F</i>	5' CGAAGGAGTTGTTACTTTGACCC 3'
<i>AtCSD2 -R</i>	5' GAACCACAAAGGCTCTTCCAAC 3'
<i>AtFSD1 -F</i>	5' CTCAAGCCACCTCCATTCG 3'
<i>AtFSD1 -R</i>	5' GCGTTGTTGAAAGCAGGGA 3'
<i>AtFSD2 -F</i>	5' TGGATTATCACTGGGGCAAAC 3'
<i>AtFSD2 -R</i>	5' GGATAGACTCCCAGAAGAACTCG 3'
<i>AtFSD3 -F</i>	5' GTGAACCCAACATCCCAATCG 3'
<i>AtFSD3 -R</i>	5' TTGCGTCACTAACATTACTGTCACC 3'
<i>AtGRI-F</i>	5' GGGTTTTGATGACGAAATGAGG 3'
<i>AtGRI-R</i>	5' ATAGGACGACATCTGCCACGA 3'
<i>AtMSD1-F</i>	5' GTTTGGGAGCACGCCTACTAC 3'
<i>AtMSD1-R</i>	5' GTTCATCTCCTTATGTCATCGTGTA 3'
<i>AtCAT1-F</i>	5' CGCCGATTTGCGAGATACA 3'

<i>AtCAT1</i> -R	5' ACCCTCTCAGGAATCCGCTC 3'
<i>AtCAT2</i> -F	5' TATCCAAC TCCGCCTGCTGTCT 3'
<i>AtCAT2</i> -R	5' ATGCGTGGGTCGGATAGGG 3'
<i>AtCAT3</i> -F	5' GGTGACACTCAGAGACATCGCC 3'
<i>AtCAT3</i> -R	5' AAACCTGTCTTGCCTGTCTGG 3'
<i>AtGSH1</i> - F	5' TTTGAGCAGTATGTTGACTACGCAC 3'
<i>AtGSH1</i> - R	5' GCAGTTCACCAGGGAGACAGG 3'
<i>AtGSH2</i> -F	5' GGAAATGCTTTGCTGGGC 3'
<i>AtGSH2</i> -R	5' TCTCCATAGATGTTGTTTCCTCC 3'