

Transcriptional profiles of drought-responsive genes in modulating transcription, signal transduction and biochemical pathways in tomato

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Supplementary materials

Table S1: Primer information of genes for the RT-PCR analysis

Accession No.	Primer set	
	Forward primer (5'-3')	Reverse primer (5'-3')
SGN-U213363	ATGGCTATTTACAAAGTTGCTCTCC	AAGTAACACAAGCCGTGGTAAAGGT
SGN-U231627	CATGGAGAGTATATATGACAGTGCC	TTTAAAACTCGCAATCCACCTCTG
SGN-U214777	AGCTACACCACCACTTAAAGTTCTC	ACCAACCAAAAACAAAAAAGGAACC
SGN-U213276	TCCGACACAGCGCATTCCGCCAAAG	TTTCTTTAGCCGACTCCATCATTTT
SGN-U219218	CCAAAAGCTATGTATAACACAAGAA	CAGAGAAAGGGAAAGAGCTTAATAT
SGN-U227655	GTCTCATTTGTATGGCTGCTTATCA	TACTCATGGATAACCCCAGAGACTA
SGN-U218605	ATGGATTGTGGATTCAATTATGAAT	ATAATACCTTGTTCCTGCTCTTGGC
SGN-U223525	AACGAAAGTTGATCGATAACAAGTAA	AAGGATACTATATGAACAAACACCC
X14449	TTGCTTGCTTTCACCCTTGG	TTGGCACCAGTTGGGTCCTT

Table S2: Seedling survival rates of ILs, M82 and *S. pennellii* under drought stress treatment at different seasons in 2006

Gnotype name	Survival rate (%)		
	May	Sep	Nov
IL2-5	70.00	80.00	73.33
IL9-1	73.33	80.00	80.00
IL11-1	50.00	66.67	63.33
IL8-1	63.33	56.67	56.67
IL6-3	53.33	63.33	56.67
IL1-4	50.00	53.33	53.33
IL3-2	43.33	50.00	46.67
M82	13.33	6.67	16.67
<i>S. pennellii</i>	100.00	93.33	90.00

Table S3: Leaf damage rates of two selected ILs, M82 and *S. pennellii* under different drought stresses.

Data are shown as means  $\pm$  standard error of triplicate samples.

Mild stress: tomato seedlings immersed in 2% (w/v) PEG nutrient solution 12h;

Moderate stress: tomato seedlings immersed in 4% (w/v) PEG nutrient solution 12h;

Severe stress: tomato seedlings immersed in 6% (w/v) PEG nutrient solution 12h.

Genotype name	Mild stress	Moderate stress	Severe stress
<i>S. pennellii</i>	15.36 $\pm$ 0.19*	16.89 $\pm$ 0.37*	17.66 $\pm$ 0.88*
M82	19.29 $\pm$ 0.54	23.76 $\pm$ 0.68	24.50 $\pm$ 0.17
IL9-1	16.37 $\pm$ 0.21*	16.73 $\pm$ 0.12*	22.37 $\pm$ 0.11*
IL2-5	16.05 $\pm$ 0.52*	16.55 $\pm$ 0.65*	17.77 $\pm$ 0.72*

\* Significantly different from M82 at  $p < 0.01$ .

Table S5: Functional classification of drought responsive genes

The percentage of genes involved in each biological process was shown in parenthesis.

Biological Process	Number of differently expressed genes		
	M82	IL2-5	IL9-1
Cellular process	547 (38.52%)	379 (38.01%)	315 (37.63%)
Metabolic process	456 (32.11%)	287 (28.79%)	229 (27.36%)
Response to stress	403 (28.38%)	302 (30.29%)	267 (31.90%)
Biosynthetic process	312 (21.97%)	223 (22.37%)	194 (23.18%)
Response to abiotic stimulus	249 (17.54%)	172 (17.25%)	155 (18.52%)
Transport	201 (14.15%)	150 (15.05%)	118 (14.10%)
Response to endogenous stimulus	193 (13.59%)	142 (14.24%)	117 (13.98%)
Signal transduction	187 (13.17%)	127 (12.74%)	108 (12.90%)
Transcription	182 (12.82%)	149 (14.94%)	126 (15.05%)
Cellular amino acid and derivative metabolic process	169 (11.90%)	119 (11.94%)	98 (11.71%)
Lipid metabolic process	169 (11.90%)	117 (11.74%)	105 (12.54%)
Protein modification process	166 (11.69%)	113 (11.33%)	84 (10.04%)
Catabolic process	163 (11.48%)	89 (8.93%)	80 (9.56%)
Carbohydrate metabolic process	150 (10.56%)	88 (8.83%)	71 (8.48%)
Cellular component organization	149 (10.49%)	110 (11.03%)	91 (10.87%)
Secondary metabolic process	147 (10.35%)	119 (11.94%)	102 (12.19%)
Multicellular organismal development	131 (9.23%)	94 (9.43%)	73 (8.72%)
Response to biotic stimulus	121 (8.52%)	100 (10.03%)	78 (9.32%)
Protein metabolic process	114 (8.03%)	73 (7.32%)	66 (7.89%)
Generation of precursor metabolites and energy	95 (6.69%)	65 (6.52%)	50 (5.97%)
Photosynthesis	82 (5.78%)	57 (5.72%)	40 (4.78%)
Reproduction	73 (5.14%)	50 (5.01%)	41 (4.90%)

Response to external stimulus	73 (5.14%)	58 (5.82%)	55 (6.57%)
Cell differentiation	61 (4.30%)	51 (5.11%)	35 (4.18%)
Nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	59 (4.16%)	41 (4.11%)	38 (4.54%)
Post-embryonic development	58 (4.09%)	47 (4.71%)	37 (4.42%)
Anatomical structure morphogenesis	52 (3.67%)	39 (3.91%)	16 (1.91%)
Flower development	52 (3.67%)	46 (4.61%)	35 (4.18%)
Cell death	47 (3.31%)	40 (4.01%)	31 (3.70%)
Cell growth	42 (2.96%)	29 (2.91%)	27 (3.23%)
Cell cycle	41 (2.89%)	33 (3.31%)	28 (3.35%)
Embryonic development	36 (2.54%)	17 (1.70%)	16 (1.91%)
Ripening	31 (2.18%)	24 (2.40%)	7 (0.84%)
Cellular homeostasis	25 (1.76%)	16 (1.60%)	13 (1.55%)
Translation	22 (1.55%)	19 (1.90%)	14 (1.67%)
Response to extracellular stimulus	20 (1.41%)	14 (1.40%)	12 (1.43%)
DNA metabolic process	17 (1.20%)	13 (1.30%)	6 (0.72%)
Tropism	16 (1.13%)	8 (0.80%)	6 (0.72%)
Cell communication	15 (1.06%)	12 (1.20%)	12 (1.43%)
Growth	14 (0.99%)	8 (0.80%)	5 (0.60%)
Pollination	4 (0.28%)	1 (0.10%)	1 (0.12%)
Behavior	3 (0.21%)	1 (0.10%)	0 (0.00%)
Regulation of gene expression, epigenetic	3 (0.21%)	1 (0.10%)	2 (0.24%)
Cell-cell signaling	2 (0.14%)	1 (0.10%)	1 (0.12%)
Respiratory electron transport chain	1 (0.07%)	1 (0.10%)	1 (0.12%)

Table S7: Biochemical pathways and their corresponding genes affected by drought stress in all three genotypes  
Significant difference (FDR <0.05 and fold change  $\geq 2$ ) in relative level is shown in bold.

Accession no.	Annotation	e-value	M82	IL2-5	IL9-1
<b>Carotenoid biosynthesis</b>					
SGN-U212843	Phytoene synthase 1, chloroplast precursor	0	<b>-2.63</b>	<b>-2.56</b>	<b>-2.50</b>
SGN-U232158	Phytoene synthase 1, chloroplast precursor	7e-010	<b>-3.00</b>	<b>-2.91</b>	<b>-2.99</b>
SGN-U212844	Phytoene synthase 2, chloroplast precursor	6e-083	<b>-3.34</b>	<b>-2.16</b>	<b>-2.00</b>
SGN-U212842	Mutant phytoene synthase	1e-041	<b>-2.46</b>	<b>-2.38</b>	-1.94
SGN-U230217	Putative phytoene synthase	1e-066	<b>-3.15</b>	<b>-2.29</b>	<b>-2.30</b>
M84744	TOMCBPe Tomato phytoene synthetase		<b>-2.89</b>	<b>-2.71</b>	<b>-2.94</b>
SGN-U221920	Lycopene beta-cyclase	1e-155	<b>-2.11</b>	<b>-2.04</b>	<b>-2.19</b>
Y14809	Beta-carotene hydroxylase		<b>-2.93</b>	<b>-7.17</b>	<b>-6.33</b>
Y14810	beta-carotene hydroxylase		<b>3.02</b>	<b>2.56</b>	<b>2.01</b>
SGN-U215829	Beta-carotene hydroxylase	1e-160	<b>-2.65</b>	<b>-7.18</b>	<b>-10.90</b>
SGN-U233360	Beta-carotene hydroxylase	1e-083	<b>-3.17</b>	<b>-5.79</b>	<b>-5.92</b>

SGN-U221254	Zeaxanthin epoxidase	1e-123	-1.32	<b>-2.27</b>	<b>-2.27</b>
<b>Nitrogen reduction and fixation</b>					
SGN-U218881	Nitrate reductase	0.0	<b>-97.10</b>	<b>-24.50</b>	<b>-20.40</b>
SGN-U225781	Nitrate reductase	1e-156	<b>-68.50</b>	<b>-19.70</b>	<b>-23.30</b>
SGN-U221852	Nitrite reductase	2e-080	<b>-2.16</b>	<b>-2.27</b>	<b>-2.07</b>
SGN-U218320	Nitrite reductase	0.0	<b>-9.01</b>	<b>-4.70</b>	<b>-5.15</b>
SGN-U215084	Glutamine synthetase GS56	1e-142	<b>-14.20</b>	<b>-7.96</b>	<b>-3.27</b>
SGN-U223400	Aminomethyl transferase	1e-148	<b>-2.74</b>	<b>-2.06</b>	<b>-2.04</b>
SGN-U217684	Phenylalanine ammonia-lyase	0.0	<b>-2.94</b>	<b>-3.49</b>	<b>-2.19</b>
SGN-U215486	Asparagine synthetase	6e-032	<b>3.51</b>	<b>2.17</b>	1.38
<b>Lignin biosynthesis</b>					
SGN-U217714	4-coumarate:coenzyme A ligase	1e-144	<b>-2.63</b>	<b>-3.18</b>	<b>-2.65</b>
SGN-U218477	4-coumarate-CoA ligase-like protein	2e-073	<b>-2.44</b>	<b>-2.47</b>	<b>-2.60</b>
SGN-U214174	4-coumarate--CoA ligase 1 (4CL 1)	0.0	<b>-2.81</b>	<b>-3.86</b>	<b>-3.09</b>
SGN-U218376	4-coumarate:coenzyme A ligase	1e-138	<b>-2.19</b>	<b>-2.34</b>	<b>-2.28</b>
SGN-U231278	4-coumarate-CoA ligase-like protein	5e-045	-1.86	<b>-2.02</b>	<b>-2.53</b>
SGN-U215109	O-methyltransferase/S-adenosylmethionin e-dependent methyltransferase	3e-079	<b>10.11</b>	<b>14.06</b>	<b>16.45</b>
<b>Suberin biosynthesis</b>					
SGN-U232238	Phenylalanine ammonia-lyase (EC 4.3.1.5)	6e-097	<b>-3.83</b>	<b>-4.14</b>	<b>-2.78</b>
SGN-U232239	Phenylalanine ammonia-lyase (PAL)	2e-056	<b>-5.5</b>	<b>-4.90</b>	<b>-3.27</b>
SGN-U217684	Phenylalanine ammonia-lyase	0.0	<b>-2.94</b>	<b>-3.49</b>	<b>-2.19</b>
SGN-U213271	Cinnamic acid 4-hydroxylase	3e-052	<b>-3.37</b>	<b>-3.25</b>	<b>-2.93</b>
SGN-U213270	Cinnamic acid 4-hydroxylase	0.0	<b>-2.96</b>	<b>-3.03</b>	<b>-2.31</b>
SGN-U215381	Putative p-coumaroyl 3'-hydroxylase CYP98A-C1	0.0	<b>-3.17</b>	<b>-4.21</b>	<b>-3.32</b>
SGN-U215109	O-methyltransferase/S-adenosylmethionin e-dependent methyltransferase	3e-079	<b>10.11</b>	<b>14.06</b>	<b>16.45</b>
<b>polyamine biosynthesis</b>					
SGN-U213684	Putative spermine synthase	0.0	<b>-2.27</b>	<b>-2.55</b>	<b>-2.11</b>
SGN-U213682	Putative spermine synthase	7e-018	<b>-2.62</b>	<b>-2.64</b>	<b>-2.51</b>
SGN-U213683	Putative spermine synthase	4e-037	-1.98	<b>-2.26</b>	<b>-2.24</b>
SGN-U232590	Putative spermine synthase	2e-067	<b>-2.11</b>	<b>-2.43</b>	<b>-2.62</b>
SGN-U213682	Putative spermine synthase	7e-018	<b>-2.62</b>	<b>-2.64</b>	<b>-2.51</b>
AY335900	Putative spermine synthase (SPM1)		-1.53	<b>-2.24</b>	-1.89
<b>Sulfate assimilation and dissimilation</b>					
SGN-U213833	Sulfate adenylyltransferase	0.0	<b>-7.56</b>	<b>-4.39</b>	<b>-5.08</b>
SGN-U225535	Adenylyl-sulfate reductase	0.0	<b>-17.10</b>	<b>-4.82</b>	<b>-6.37</b>
SGN-U240835	Adenylyl-sulfate reductase	5e-017	<b>-8.67</b>	<b>-3.31</b>	<b>-3.73</b>
SGN-U214793	Cytosolic cysteine synthase	1e-153	<b>5.20</b>	<b>3.45</b>	<b>3.88</b>
<b>Phenylpropanoid biosynthesis</b>					
SGN-U232238	Phenylalanine ammonia-lyase (EC 4.3.1.5)	6e-097	<b>-3.83</b>	<b>-4.14</b>	<b>-2.78</b>

SGN-U232239	Phenylalanine ammonia-lyase (PAL)	2e-056	<b>-5.50</b>	<b>-4.90</b>	<b>-3.27</b>
SGN-U226405	Phenylalanine ammonia-lyase	6e-042	-1.88	<b>-2.04</b>	<b>-2.08</b>
SGN-U217684	Phenylalanine ammonia-lyase	0.0	<b>-2.94</b>	<b>-3.49</b>	<b>-2.19</b>
SGN-U213271	Cinnamic acid 4-hydroxylase	3e-052	<b>-3.37</b>	<b>-3.25</b>	<b>-2.93</b>
SGN-U213270	Cinnamic acid 4-hydroxylase	0.0	<b>-2.96</b>	<b>-3.03</b>	<b>-2.31</b>
<b>Glutamate degradation III</b>					
SGN-U212562	Glutamate decarboxylase isozyme 2	1e-144	<b>2.88</b>	<b>2.44</b>	1.90
SGN-U212561	Glutamate decarboxylase isozyme 1	0.0	<b>2.84</b>	<b>2.77</b>	<b>2.28</b>
SGN-U212563	Glutamate decarboxylase isozyme 3	1e-103	<b>2.08</b>	<b>2.14</b>	1.87
SGN-U213860	Glutamate decarboxylase	0.0	<b>10.01</b>	<b>3.47</b>	<b>2.14</b>
SGN-U213861	Putative glutamate decarboxylase	1e-045	<b>16.55</b>	<b>4.71</b>	<b>2.97</b>
<b>Arginine degradation VI and VII</b>					
AY656837	Arginase 1 (ARG1)		<b>2.68</b>	<b>2.27</b>	<b>2.23</b>
SGN-U214917	Arginase 1	0.0	<b>3.56</b>	<b>2.03</b>	<b>2.18</b>
SGN-U216285	Pyrroline-5-carboxylate reductase	1e-121	<b>3.08</b>	<b>2.65</b>	<b>2.48</b>
<b>Salicylic acid biosynthesis</b>					
SGN-U232238	Phenylalanine ammonia-lyase (EC 4.3.1.5)	6e-097	<b>-3.83</b>	<b>-4.14</b>	<b>-2.78</b>
SGN-U232239	Phenylalanine ammonia-lyase (PAL)	2e-056	<b>-5.50</b>	<b>-4.90</b>	<b>-3.27</b>
SGN-U226405	Phenylalanine ammonia-lyase	6e-042	-1.88	<b>-2.04</b>	<b>-2.08</b>
SGN-U217684	Phenylalanine ammonia-lyase	0.0	<b>-2.94</b>	<b>-3.49</b>	<b>-2.19</b>
<b>Glycosylglyceride desaturation pathway</b>					
SGN-U217678	FAD5 (FATTY ACID DESATURASE 5); oxidoreductase	1e-150	<b>-23.40</b>	<b>-9.25</b>	<b>-10.40</b>
SGN-U214130	Omega-3 fatty acid desaturase	0.0	<b>-6.97</b>	<b>-3.82</b>	<b>-3.65</b>
<b>Canavanine degradation and citrulline biosynthesis</b>					
AY656837	Arginase 1 (ARG1)		<b>2.68</b>	<b>2.26</b>	<b>2.23</b>
SGN-U214917	Arginase 1	0.0	<b>3.56</b>	<b>2.03</b>	<b>2.18</b>
<b>Gibberellin biosynthesis</b>					
SGN-U226448	Gibberellin 20-oxidase-1; 20ox-1	0.0	<b>-15.70</b>	<b>-5.48</b>	<b>-3.55</b>
<b>Formaldehyde assimilation I (serine pathway)</b>					
SGN-U237093	Hydroxypyruvate reductase	2e-011	<b>-2.01</b>	<b>-2.15</b>	-1.92
SGN-U233374	HPR; oxidoreductase	2e-026	<b>-2.79</b>	<b>-2.32</b>	<b>-2.05</b>
SGN-U233373	NADH-dependent hydroxypyruvate reductase	7e-055	<b>-2.41</b>	<b>-2.41</b>	<b>-2.29</b>
<b>Glycogen biosynthesis II (from UDP-D-Glucose)</b>					
SGN-U221404	Starch branching enzyme II, SBE-II	2e-058	<b>2.54</b>	<b>2.74</b>	<b>3.19</b>
<b>NAD biosynthesis I (from aspartate)</b>					
SGN-U226241	Putative L-aspartate oxidase	5e-077	<b>-4.32</b>	<b>-3.17</b>	<b>-2.71</b>
<b>Glycine degradation I</b>					
SGN-U223400	Aminomethyltransferase	1e-148	<b>-2.74</b>	<b>-2.06</b>	<b>-2.04</b>
<b>Glycosylglyceride biosynthesis</b>					
SGN-U220224	Digalactosyldiacylglycerol synthase 2	1e-044	<b>-3.41</b>	<b>-2.46</b>	<b>-2.14</b>
SGN-U226648	Putative UDP-sulfoquinovose synthase	1e-058	<b>3.76</b>	<b>2.65</b>	<b>3.04</b>

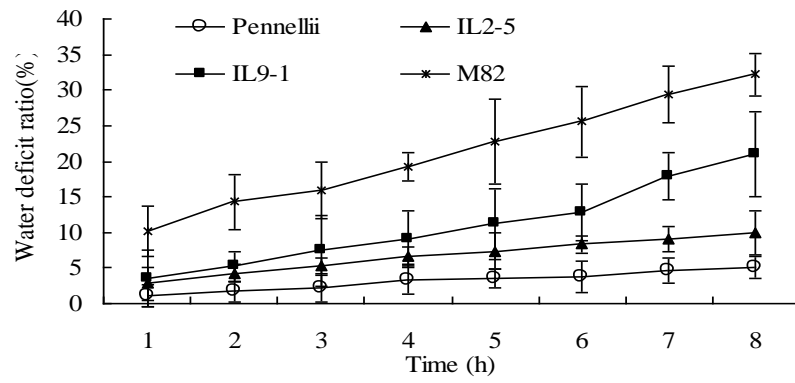


Figure S1: Water deficit ratio of detached leaves of two selected ILs, M82 and *S. pennellii* under 25°C.

Results are means  $\pm$  standard error of triplicate.

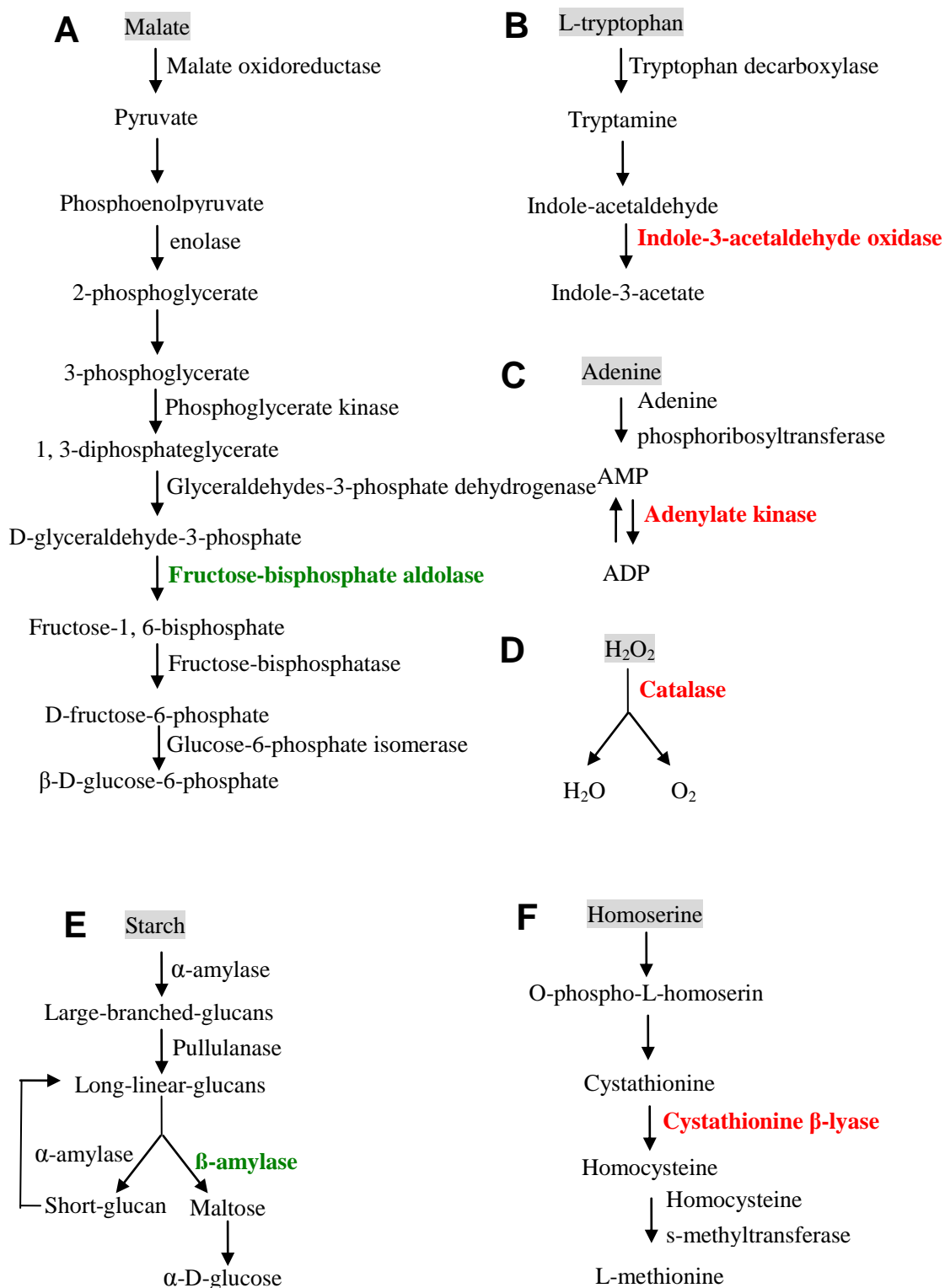


Figure S2: Biochemical pathways affected by drought stress specifically in the tolerant genotypes.

A: Gluconeogenesis; B: Tryptophan degradation; C: Salvage pathways of purine and pyrimidine nucleotides; D: Removal of superoxide radicals; E: Starch degradation, F: Methionine biosynthesis. Enzymes encoded by specifically expressed drought-responsive genes were shown in red (up-regulated) or green (down-regulated).