Additional file 2

| Sequence | Sequence type | Number of | GC% | URL |
|----------|---------------------|-----------|------|--|
| origin | | sequences | | |
| Genome | Chromosomes | 10 | 41.6 | http://www.phytozome.net/sorghum |
| | Super scaffold | 3394 | 40.9 | http://www.phytozome.net/sorghum |
| UniGene | Unclustered UniGene | 199087 | 52.9 | http://www.ncbi.nlm.nih.gov/UniGene/ |
| | Clustered UniGene | 14057 | 52.0 | http://www.ncbi.nlm.nih.gov/UniGene/ |
| ESTs | TIGR transcripts | 209835 | 53.3 | http://www.ncbi.nlm.nih.gov/dbEST/ http://www.plantgdb.org/prj/ESTCluster/ ftp://occams.dfci.harvard.edu/pub/bio/tgi/data/ |
| | NCBI dbEST | 20199 | 50.7 | http://www.ncbi.nlm.nih.gov/dbEST/ |
| RNA-seq | Maize GEO data | 3000000 | NA | http://www.ncbi.nlm.nih.gov/geo/ |
| | (GSE40070) | reads | | |

Table S1 Summary of sorghum transcript and genomic data

| Main parts of plants | Tissues | Number of lib ^a | Library ID | Name of Library | Sequence number |
|-------------------------|--------------------|-------------------------------|---|---|--|
| Body site | Callus | 1 | Lib.13735 | Callus culture/cell suspension | 10449 |
| | embryo | 2 | Lib.5437 | Embryo 1 (EM1) | 9843 |
| | leaf | 12 | Lib.15546 Lib.10086 Lib.14497 Lib.14500 8 not shown | Wounded leaves Pathogen-infected compatible 1 (PIC1) Drought-stressed after flowering Drought-stressed before flowering | 11221 9092 6295 3597 each <1000 |
| | ovary | 3 | Lib.5439 Lib.7266 1 not shown | Ovary 1 (OV1) Ovary 2 (OV2) | 5052 4983 <1000 |
| | panicle | 2 | Lib.9519 1 not shown | Immature panic le 1 (IP1) | 9588 <1000 |
| | pollen | 1 | Lib.14372 | Pollen | 10212 |
| | chloroplast | 1 | not shown | | each <1000 |
| | root | 3 | Lib.15544 Acid- and alkali Lib.16897 roots Lib.20762 Anaerobic roots Sorghum bicolo Root hair | | 7744 6113 5468 |
| | shoot | 2 | Lib.16898 1 not shown | GA- or brassinolide- treated seedlings | 11134 <1000 |
| | Whole | 4 | not shown | | each <1000 |
| | Unspecified tissue | 1 | not shown | | <1000 |
| | mixed | 13 | Lib.4037 Lib.13713 Lib.15545 Lib.4038 Lib.13770 Lib.13736 Lib.13769 Lib.12996 Lib.14297 Lib.14297 Lib.14296 | Dark Grown 1 (DG1) Heat-shocked seedlings Oxidatively-stressed leaves and roots Water-stressed 1 (WS1) Ethylene-treated seedlings Salicylic acid-treated seedlings Abscisic acid-treated seedlings | 11099 10558 10086 10039 6942 6737 5801 4907 3984 3849 3723 |

Table S2 Overview of UniGene libraries (build # 30)

| Main parts of plants | Tissues | Number of lib ^a | Library ID | Name of Library | Sequence number |
|-------------------------|---|---------------------------------------|---|---|--|
| | | | 2 not shown | Iron-deficient seedlings Nitrogen-deficient seedlings Phosphorous-deficient seedlings | each < 1000 |
| | not yet classified | 4 | Lib.5441 Lib.2801 2 not shown | Pathogen induced 1 (PI1) Light Grown 1 (LG1) | 9569 9451 each <1000 |
| Developmental stage | Germinating seed seedling vegetative flowering ripening unknown developmental stage not yet classified | 2 25 9 1 1 1 3 2 | Not shown Not shown Not shown Not shown Not shown Not shown Not shown | | each <1000 each < 1000 each < 1000 < 1000 < 1000 < 1000 each < 1000 each < 1000 |

| Chrom | ן | UniGene clusters | that | UniGene | clusters that | mapped | Grand Total | | |
|--------|--------|-------------------------|-------|---------|----------------|--------|-------------|--|--|
| osomes | ove | erlapped Known | genes | to in | ntergenic regi | ion | | | |
| - | DR^1 | Non-DR(DR) ² | Total | DR | Non-DR | Total | | | |
| Chr1 | 20 | 1733 (37) | 1753 | 22 | 91 | 113 | 1866 | | |
| Chr2 | 14 | 1181 (33) | 1195 | 20 | 69 | 89 | 1284 | | |
| Chr3 | 16 | 1322 (39) | 1338 | 15 | 73 | 88 | 1427 | | |
| Chr4 | 13 | 1150 (35) | 1163 | 12 | 64 | 76 | 1239 | | |
| Chr5 | 4 | 432 (13) | 436 | 7 | 49 | 56 | 493 | | |
| Chr6 | 19 | 876 (21) | 895 | 10 | 64 | 74 | 966 | | |
| Chr7 | 9 | 670 (23) | 679 | 9 | 61 | 70 | 749 | | |
| Chr8 | 8 | 544 (13) | 552 | 7 | 63 | 70 | 622 | | |
| Chr9 | 14 | 883 (27) | 897 | 12 | 66 | 78 | 976 | | |
| Chr10 | 6 | 750 (15) | 756 | 7 | 54 | 61 | 817 | | |
| Super | 0 | 99 (2) | 99 | 7 | 74 | 81 | 180 | | |
| Total | 123 | 9640 (258) | 9763 | 128 | 728 | 856 | 10619 | | |

Table S3 Chromosomal distribution of UniGene clusters mapped to genome

Key to legend:

¹UniGene clusters that represent purely drought responsive

² UniGene clusters that basically represent non-drought responsive, however, contain shorter ESTs which were expressed under drought conditions dispersed within the clusters.

| D | Existing a | nnotation | Current annotation | | | | | | |
|-------------|--------------------|-----------|--------------------|-------|-------------|------|--|--|--|
| Description | | | Upd | ated | Novel | | | | |
| | gene models mRNA g | | gene models | mRNA | gene models | mRNA | | | |
| Original | 34211 | 37205 | 30937 | 33862 | - | - | | | |
| Updated | - | - | 3274 | 3343 | 210 | 210 | | | |
| Total | 34211 | 37205 | 34211 | 37205 | 210 | 210 | | | |

Table S4 Comparison and update of annotation

This supplementary table shows the comparison and update of annotation between existing and current prediction of sorghum genome based on PASA alignment evidence.

| Known | Complete genes | | | | | |
|-----------------|----------------|-------------|-------------|--------------|--------------|-------|
| drought-related | 3' and 5' UTR | only 3' UTR | only 5' UTR | at-least one | truncated at | Total |
| function | attained | attained | attained | end attained | both ends | |
| DR | 12 | 15 | 2 | 29 | 137 | 166 |
| NDR | 0 | 0 | 0 | 0 | 44 | 44 |
| Total | 12 | 15 | 2 | 29 | 181 | 210 |

 Table S5 Functional distribution of the novel gene structure models

| Scaffolds | Chr | Chromosomal distribution of the modified existing gene models | | | | | | | | | |
|-----------|-------|---|-----------------------|----------------|------------------|------------------|-------|------------------|----------------|------|--|
| | A^1 | B^2 | C ³ | D^4 | E^{5} | \mathbf{F}^{6} | G^7 | H^{8} | \mathbf{I}^9 | - | |
| chr1 | 3 | 16 | 24 | 642 | 0 | 0 | 1 | 0 | 0 | 685 | |
| chr2 | 6 | 11 | 7 | 373 | 0 | 0 | 0 | 1 | 0 | 397 | |
| chr3 | 2 | 19 | 14 | 445 | 0 | 0 | 0 | 1 | 0 | 480 | |
| chr4 | 2 | 13 | 11 | 402 | 0 | 0 | 1 | 0 | 0 | 428 | |
| chr5 | 2 | 3 | 7 | 170 | 0 | 0 | 0 | 0 | 0 | 182 | |
| chr6 | 2 | 8 | 5 | 220 | 0 | 0 | 0 | 0 | 0 | 235 | |
| chr7 | 0 | 5 | 10 | 193 | 0 | 0 | 0 | 0 | 0 | 208 | |
| chr8 | 0 | 4 | 4 | 182 | 0 | 0 | 0 | 0 | 0 | 190 | |
| chr9 | 3 | 2 | 3 | 254 | 0 | 1 | 0 | 0 | 0 | 263 | |
| chr10 | 1 | 4 | 11 | 248 | 0 | 0 | 0 | 0 | 1 | 265 | |
| super | 0 | 0 | 0 | 10 | 0 | 0 | 0 | 0 | 0 | 10 | |
| Total | 21 | 85 | 96 | 3139 | 0 | 1 | 2 | 2 | 1 | 3343 | |

Table S6 Chromosomal distribution of existing genes modified corresponding to the position and structure of modification.

Legend to key:

A¹ Transcript A; B² Transcript B; C³ Transcript C; D⁴ Transcript D; E⁵ Transcript E;

F⁶ Transcript F; G⁷ Transcript G; H⁸ Transcript H and I⁹ Transcript I (Figure 1)

| | | | Partial gene structure | | | | | | | | |
|-----------|------|---|------------------------|--------------|--------|------------------|-------|--|--|--|--|
| Scaffolds | Comp | lete gene structure | One e | end retained | trunca | ted at both ends | | | | | |
| | DRª | $\mathbf{ND}\text{-}\mathbf{DR}^{\mathrm{b}}$ | DRª | $ND-DR^{b}$ | DRª | $Non-DR^{b}$ | Total | | | | |
| chr1 | 3 | 0 | 0 | 0 | 12 | 13 | 28 | | | | |
| chr2 | 3 | 0 | 0 | 0 | 9 | 8 | 20 | | | | |
| chr3 | 1 | 0 | 2 | 0 | 15 | 3 | 21 | | | | |
| chr4 | 1 | 0 | 4 | 0 | 10 | 8 | 23 | | | | |
| chr5 | 0 | 0 | 2 | 0 | 11 | 6 | 19 | | | | |
| chr6 | 0 | 0 | 1 | 0 | 8 | 3 | 12 | | | | |
| chr7 | 1 | 0 | 4 | 0 | 10 | 5 | 20 | | | | |
| chr8 | 2 | 0 | 0 | 0 | 16 | 7 | 25 | | | | |
| chr9 | 0 | 0 | 2 | 0 | 16 | 2 | 20 | | | | |
| chr10 | 1 | 0 | 2 | 0 | 11 | 6 | 20 | | | | |
| super | 0 | 0 | 0 | 0 | 1 | 1 | 2 | | | | |
| Total | 12 | 0 | 17 | 0 | 119 | 62 | 210 | | | | |

 Table S7 Chromosomal distribution of the novel gene structure models

Key to legend: ^a Drought responsive; ^b Non-drought responsive

| | Intron | | | | | | | | | | |
|-------|-------------------|-------|-------|-------|-------------------|-----------------------|-------|-------|--|--|--|
| S^1 | | Sp | liced | | Retained | | | | | | |
| | Coordinate | O^2 | A^3 | M^4 | Coordinate | O ² | A^3 | M^4 | | | |
| chr1 | 1819870-1820018 | + | 64 | 1 | 1819870-1820018 | + | 65 | 1 | | | |
| chr1 | 2245082-2245182 | - | 83 | 5 | 2245082-2245182 | | 84 | 2 | | | |
| chr1 | 2630315-2630410 | - | 101 | 1 | 2630315-2630410 | - | 102 | 2 | | | |
| chr1 | 8195826-8195903 | - | 306 | 1 | 8195826-8195903 | - | 307 | 1 | | | |
| chr1 | 47862385-4786251 | + | 638 | 6 | 47862385-4786251 | + | 639 | 6 | | | |
| chr1 | 56966890-56966985 | - | 779 | 2 | 56966890-56966985 | - | 780 | 1 | | | |
| chr1 | 58887490-58887568 | - | 819 | 3 | 58887490-58887568 | - | 820 | 1 | | | |
| chr1 | 58887601-58887781 | - | 819 | 1 | 58887601-58887781 | - | 820 | 1 | | | |
| chr1 | 61110177-61110310 | - | 896 | 9 | 61110177-61110310 | - | 897 | 2 | | | |
| chr1 | 67980410-67980506 | + | 1125 | 6 | 67980410-67980506 | + | 1126 | 2 | | | |
| chr2 | 69162027-69162097 | - | 2358 | 2 | 69162027-69162097 | - | 2357 | 2 | | | |
| chr3 | 62293574-62293685 | - | 3153 | 3 | 62293574-62293685 | - | 3154 | 1 | | | |
| chr4 | 65061928-65062017 | + | 4139 | 2 | 65061928-65062017 | + | 4140 | 1 | | | |
| chr6 | 45323794-45323870 | - | 4626 | 18 | 45323794-45323870 | - | 4627 | 1 | | | |
| chr6 | 60237462-60237612 | + | 4942 | 2 | 60237462-60237612 | + | 4943 | 1 | | | |
| chr7 | 8345323-8345537 | + | 5138 | 5 | 8345323-8345537 | + | 5139 | 4 | | | |
| chr8 | 6642051-6642134 | + | 5536 | 7 | 6642051-6642134 | + | 5537 | 1 | | | |
| chr8 | 6641856-6641938 | + | 5536 | 4 | 6641856-6641938 | + | 5537 | 1 | | | |
| chr8 | 44687799-44687915 | - | 5622 | 1 | 44687799-44687915 | - | 5621 | 1 | | | |
| chr9 | 2436568-2436638 | - | 5784 | 4 | 2436568-2436638 | - | 5785 | 1 | | | |
| chr9 | 52085543-52085653 | - | 6061 | 2 | 52085543-52085653 | - | 6060 | 4 | | | |
| chr9 | 46038980-46039077 | + | 5954 | 32 | 46038980-46039077 | + | 5955 | 15 | | | |
| chr9 | 46038375-46038506 | + | 5953, | 178 | 46038375-46038506 | + | 5955 | 4 | | | |
| | | | 5954 | | | | | | | | |
| chr9 | 54606686-54606830 | + | 6117 | 2 | 54606686-54606830 | + | 6118 | 2 | | | |
| chr9 | 57490227-57490310 | - | 6208 | 5 | 57490227-57490310 | - | 6209 | 1 | | | |
| chr10 | 58496008-58496090 | _ | 1717 | 2 | 58496008-58496090 | - | 1718 | 1 | | | |

Table S8 Genomic distribution of spliced and retained intron based on PASA analysis

Key to legend: ¹ Chromosomes number 1-10; ² Orientation of the strand; ³ Assembly number that the transcripts belong to and ⁴ Maximum evidence support.

| | Exon | | | | | | | | | | | |
|-------|-------------------|-------|----------------|-------|--------------------|-------|----------------|-------|--|--|--|--|
| S^1 | Skip | ped | | | Retained | | | | | | | |
| | Coordinate | O^2 | A ³ | M^4 | Coordinate | O^2 | A ³ | M^4 | | | | |
| chr2 | 15949877-15955756 | + | 1983 | 1 | 15951004-15951091, | + | 1984 | 1 | | | | |
| | | | | | 15951181-15951253 | | | | | | | |
| chr2 | 75790551-75791220 | + | 2540 | 1 | 75790984-75791097 | + | 2539 | 7 | | | | |
| chr3 | 71528810-71529369 | - | 3403 | 2 | 71529086-71529156 | - | 3404 | 1 | | | | |
| chr4 | 4717042-4717529 | + | 3609 | 1 | 4717411-4717488 | + | 3608 | 11 | | | | |
| chr6 | 1517417-1519399 | - | 4521 | 1 | 1518211-1518383 | - | 4520 | 2 | | | | |
| chr6 | 60298761-60299726 | - | 4947 | 1 | 60299069-60299251 | - | 4946 | 9 | | | | |
| chr9 | 54972512-54974544 | - | 6127 | 2 | 54972660-54973133 | - | 6126 | 10 | | | | |
| chr10 | 2956478-2957725 | - | 1340 | 1 | 2956662-2956707 | - | 1339 | 1 | | | | |
| chr10 | 59988260-59988901 | + | 1779 | 3 | 59988341-59988410 | + | 1780 | 2 | | | | |

Table S9 Genomic distribution of skipped and retained exons based on PASA analysis

Key to legend: refer to Table S6.

| Types of AS | AS event | Non-AS events |
|----------------------|----------|---------------|
| Alternative acceptor | 42 | - |
| Alternative donor | 14 | - |
| Alternative 5' site | - | - |
| Alternative 3' site | - | - |
| Alternative exon | 16 | - |
| Ends in intron | 10 | - |
| Retained exon | - | 9 |
| Retained Intron | 27 | - |
| Skipped exon | 9 | - |
| Spliced intron | - | 27 |
| Starts in intron | 18 | - |
| Total | 136 | 36 |

Table S10 PASA based identification of alternative splicing (AS) for the novel genes

| Features ^a | Feature | Single | Double | Triple | Quadruple | Mul | Multiple features | | Total | | |
|--------------------------------|------------------|------------------|--------|--------|-----------|-----|-------------------|---|-------|---|-----|
| | less | | | | | 5 | 6 | 7 | 8 | 9 | |
| Total Exons | 0 | 210 | 105 | 62 | 30 | 17 | 8 | 5 | 2 | 2 | 441 |
| Total introns | 0 | 105 | 62 | 30 | 17 | 8 | 5 | 2 | 2 | 0 | 231 |
| Gene per exonic ^b | 0 | 105 ^d | 43 | 32 | 13 | 9 | 3 | 3 | 0 | 2 | 210 |
| Gene per intronic ^c | 105 ^e | 43 | 32 | 13 | 9 | 3 | 3 | 0 | 2 | 0 | 210 |

Table S11 Patterns of exonic and intronic features in the novel gene structure models

Key to legend: ^a Genes, Exons and Introns; ^bGene number per exonic feature; ^cGene number per intronic feature; ^dSingle exonic genes and ^eIntronless genes.

| GO-D ^A | GO ID | GO-term | E/T ^B | GO/Bg ^C | P ^D |
|-------------------|------------|---------------------------------|------------------|--------------------|----------------|
| BP | GO:0006950 | Response to stress | 1102/1102 | 3705/26245 | 0.00 |
| BP | GO:0006950 | Response to water deprivation | 118/1102 | 374/26245 | 4.98e-55 |
| BP | GO:0009269 | Response to dessication | 21/1102 | 71/26245 | 5.4e-09 |
| BP | GO:0009408 | Response to heat | 91/1102 | 324/26245 | 1.35e-38 |
| BP | GO:0009737 | Response to ABA stimulus | 109/1102 | 664/26245 | 2.87e-28 |
| BP | GO:0009738 | ABA mediated signalling pathway | 37/1102 | 192/26245 | 4.87e-11 |
| CC | GO:0044444 | Cytoplasmic part | 477/1102 | 7660/26245 | 2.07e-20 |
| CC | GO:0009536 | Plastid | 161/1102 | 2109/26245 | 5.95e-11 |
| CC | GO:0009507 | Chloroplast | 144/1102 | 183/26245 | 1.45e-10 |
| CC | GO:0043227 | Membrane-bounded organelle | 628/1102 | 10408/26245 | 1.24e-27 |
| CC | GO:0005886 | Plasma membrane | 136/1102 | 1557/26245 | 6.93e-13 |
| MF | GO:0051082 | Unfolding protein binding | 36/1102 | 184/26245 | 1.83e-10 |
| MF | GO:0048037 | Cofactor binding | 81/1102 | 792/26245 | 5.83e-10 |
| MF | GO:0005506 | Ion binding | 91/1102 | 890/26245 | 3.17e-11 |
| MF | GO:0016209 | Antioxidant activity | 72/1102 | 276/26245 | 3.88e-28 |
| MF | GO:0009055 | Electron transfer activity | 86/1102 | 984/26245 | 1.59e-07 |

Table S12 GO functional enrichment of DR sorghum genes based on orthology groups

Key to legend: ^AGO-D denotes GO domain; ^BE/T denote enriched genes Vs test set: the numerator and denominator represent the number of enriched genes and test set respectively; ^C GO/Bg denote genes with GO-terms Vs back ground set: the numerator and denominator represent the number of genes for which corresponding GO-terms were assigned and the background set respectively; ^D P denote P-value (FDR). The first top GO-term in each GO-domain, eg. BP: response to stress (GO:0006950), CC: Cytoplasmic part (GO:0044444) and MF: Unfolding protein binding (GO:0051082), represent the most likely term associated to the corresponding number of enriched genes. As such, 1102, 477, and 36 genes respectively were assigned to these GO-terms.

| No. | Databases | Link or reference | | | |
|-----|--------------------------------------|---|--|--|--|
| 1 | Genome database: | | | | |
| | Phytozome | http://www.phytozome.net/sorghum or | | | |
| | | ftp://ftp.jgi- | | | |
| | | psf.org/pub/compgen/phytozome/v9.0/Sbicolor_ v1.4/; Goodstein <i>et al.</i> , 2012 | | | |
| | | | | | |
| | | http://www.gramene.org/; Ware et al., 2002 | | | |
| | Gramene | http://www.ensembl.org/index.html; Flicek et al., | | | |
| | Ensembl | 2012 and 2013 | | | |
| 2 | Sequences databases: | | | | |
| | RefSeq | http://www.ncbi.nlm.nih.gov/refseq/ | | | |
| | NCBI | http://www.ncbi.nlm.nih.gov/ | | | |
| | UniGene | http://www.ncbi.nlm.nih.gov/UniGene/; Rudd et al., | | | |
| | | 2003 | | | |
| | dbESTs | http://www.ncbi.nlm.nih.gov/dbEST/; Boguski | | | |
| | | et al., 1993 | | | |
| | Uniprot | http://www.uniprot.org/ | | | |
| 3 | Protein domain databases | | | | |
| | Uniref | www.uniprot.org/help/uniref | | | |
| | Uniprot | http://www.uniprot.org/ | | | |
| | Swissprot | http://www.expasy.ch/sprot/ | | | |
| | | http://www.ebi.ac.uk/swissprot/ | | | |
| | Interoroscan | https://www.ebi.ac.uk/interpro/ | | | |
| | Signal peptides | http://www.signalpeptide.com/ | | | |
| 4 | Expression databases: | | | | |
| | Gene Expression Omnipus (GEO): | http://www.ncbi.nlm.nih.gov/geo/ | | | |
| | experimental genome microarray data, | | | | |
| | RNA-seq data | | | | |

Table S13 Databases containing potential source for candidate drought responsive genes

| Ser. # | Tools | Links or references | | | |
|--------|--------------------------------------|--|--|--|--|
| 1 | alignment tools: | | | | |
| | Blast, | http://blast.ncbi.nlm.nih.gov/Blast.cgi | | | |
| | Blat, | http://genome.ucsc.edu/FAQ/FAQblat.html | | | |
| | est2genome | http://bioinfo.nhri.org.tw/cgi-bin/emboss/est2genome | | | |
| | exonerate | http://www.csc.fi/english/research/sciences/bioscience/programs/ | | | |
| | ClustalW | exonerate/index_html | | | |
| | ClustalX | http://www.clustal.org/clustal2/ | | | |
| 2 | Gene structure prediction tools: | | | | |
| | Augustus, | http://augustus.gobics.de/binaries/ | | | |
| | est2genome | ftp://ftp.hgc.jp/pub/mirror/ebi/software/exonerate/ | | | |
| | exonerate, and | http://bioinfo.nhri.org.tw/cgi-bin/emboss/est2genome | | | |
| | PASA | http://www.evidencemodul | | | |
| | Evidence Moduler (EVM), | http://evidencemodeler.sourceforge.net/ | | | |
| 3 | Gene enrichment analysis tools: | | | | |
| | AgriGO | http://www.agrigo.org/ | | | |
| | Blast2go | http://www.blast2go.de | | | |
| | MeV, | http://www.tm4.org/mev.html | | | |
| | Genevestigator, | https://www.genevestigator.ethz.Ch/ | | | |
| 4 | Genome viewer tools: | | | | |
| | Galaxy, | http://usegalaxy.org; https://main.g2.bx.psu.edu/ | | | |
| | Gbrowse | http://gmod.org/wiki/GBrowse/tool_data; | | | |
| | USCS-Gbrowse | http://genome.ucsc.edu/ | | | |
| | Biomart-ensemble, | http://www.ensembl.org/info/data/biomart.html; | | | |
| | Biomart-Gramene | http://archive.gramene.org/biomart/martview/334378 | | | |
| | | ea281f8c0071af7d45f4ab01f9 | | | |
| | Biomart Phytozome | http://www.phytozome.net/biomart/martview/6ae06d | | | |
| | | 47637222933cd94d5beb507c71 | | | |
| 5 | Expression profiling analysis tools: | | | | |
| | Multivariate Experiment Viewer | http://www.tm4.org/mev.html | | | |
| | (MeV) | | | | |

Table S14 Relevant tools for identification of the candidate gene

6 Sequence quality filtering tools:

| | RepeatMasker | http://www.repeatmasker.org/ | | |
|---|-----------------------------|---|--|--|
| | Statistical analysis tools: | | | |
| | R programming, | http://www.r-project.org/ | | |
| | MeV | http://www.tm4.org/mev.html | | |
| 7 | Programming tools: | | | |
| | linux OS | http://www.gnu.org/ | | |
| | Perl, | http://www.perlmonks.org/ | | |
| | Python, | http://www.tutorialspoint.com/python/python_tools_u | | |
| | R, programming modules, | tilities.htm | | |
| | Bioperl | http://www.r-project.org/ | | |
| | Biopython | www.bioperl.org/ | | |
| | | www.biopython.org/;http://biopython.org/wiki/Main_ | | |
| | | Page | | |

Table S15 Functional description of sorghum drought related metabolic pathways

| Pathway | Enzyme | Enzyme ID | Pathway map ID | Pathway ID | gene (id) involved |
|---|---|---------------------------|------------------------|------------------------|----------------------------------|
| Aminoacyl-tRNA biosynthesis | ligase | EC:6.1.1.16 | map00970 | KO:K01883 | *genes |
| Cysteine and methionine metabolism | Dioxygenase (iron(II)-requiring) | EC:1.13.11.54 | map00270 | KO:K08967 | Sb01g046360 |
| Drug metabolism - other enzymes | ali-esterase | EC:3.1.1.1 | map00983 | *KO | novel |
| Glucosinolate biosynthesis | CYP79A1, tyrosine N-mono-oxygenase | EC:1.14.13.41 | Map00966 (sbi00966) | KO:K13027 | Sb01g001200 |
| Glycerophospholipid metabolism | kinase (ATP dependent) | EC:2.7.1.107 | map00564 | KO:K00901 | Sb01g032250 |
| Glycerolipid metabolism | kinase (ATP dependent) | EC:2.7.1.107 | map00561 | KO:K00901 | Sb01g032250 |
| Phosphatidylinositol signalling system | kinase (ATP dependent) | EC:2.7.1.107 | map04070 | KO:K00901 | Sb01g032250 |
| Nicotinate and nicotinamide metabolism | kinase | EC:2.7.1.23 | map00760 | KO:K00858 | Sb09g019130 |
| Oxidative phosphorylation | Cytochrome c oxidase subunit 1 (Oxidase); Inorganic diphosphatase | EC:1.9.3.1; EC:3.6.1.1 | Map00190 map00190 | KO:K02256 KO:K02256 | Sb09g022400, COX1; **genes |
| Pyrimidine metabolism | RNA polymerase | EC:2.7.7.6 | map00240 | **KO | ***genes |
| Purine metabolism | RNA polymerase; adenylpyrophosphatase | EC:2.7.7.6; EC:3.6.1.3 | Map00230 map00230 | **KO KO:K01509 | novel |
| Pantothenate biosynthesis | BCAT | EC:2.6.1.42 | sbi 00770 | KO:K00826 | ****genes |
| VLIB | BCAT | EC:2.6.1.42 | sbi 00290 | KO:K00826 | ****genes |
| VLID | BCAT | EC:2.6.1.42 | sbi00280 | KO:K00826 | ****genes |
| Key to legend: *KO K01044 carboxylesterase 1: K03927 carboxylesterase 2: K03928 carboxylesterase; K15743 carboxylesterase 3/5 **KO K0:K03006; KO:K03009; KO:K03002; KO:K03006; KO:K03018; KO:K03021; KO:K03043; KO:K03046 *genes Sb01g047380; Sb02g032450 **genes Sb09g001530; Sb10g009880; Sb09g004450; Sb09g021610; Sb01g022340; Sb04g036230; Sb04g005710; Sb04g034340; Sb03g013530; Sb03g040910 ***genes Sb05g019520; Sb03g017630; Sb03g020184; Sb04g001790; Sb04g009491; Sb05g019520; Sb06g021120; Sb07g003680; Sb09g027223; Sb09g027230; Sb10g006995 ****genes Sb04g010240; Sb06g025140; and Sb09g008180 | | | | | |



Figure S1. Identified genes by mapping to existing annotation and their characterization status



Figure S2 Pattern of exon and intron number and the average length for novel identified genes.

Key to legend for exon and intron number: 1=exon1, intron1; 2=exon2, intron2; 3=exon3, intron3; 4=exon4, intron4; 5=exon5, intron5; 6=exon6, intron6; 7=exon7, intron7; 8=exon8, intron8; 9=exon9, intron9 and average = exon total average; intron total average.



Novel complete gene structure models

Figure S3 Nearest intergenic distances.

Key to legend: RNID (right nearest intergenic distance) and LNID (left nearest intergenic distance). The intergenic distances between the NGSM and the nearest existing neighbour gene were described based on the fourteen NGSMs known to have complete gene structure. The average RNID represents 57687 bases long while the average LNID represents 67754 bases long.



Figure S4 GO annotation based on blasting and mapping to non-redundant databases. GO annotation based on blasting and mapping sequences to non-redundant databases based on balst2go: a) patterns of sequences distribution based on blast hits associated to the GO-terms for the biological process; b) Species specific score for the total blast-hit; c) species specific blast top hits and d) GO-level distribution of annotations





Figure S5 Aminoacyl-tRNA biosynthesis (upper) and Cysteine and methionine metabolism (lower).





Figure S6 Drug metabolism - other enzymes (upper) and Glucosinolate biosynthesis (lower).



Figure S7 Glycerophospholipid metabolism (upper) and Glycerolipid metabolism (lower).





Figure S8 Phosphatidylinositol signalling system (upper) and nicotinate and nicotinate metabolism (lower).



Figure S9 Pyrimidine metabolism (upper) and purine metabolism (lower).



Figure S10 Valine, Luecine and Isoleucine Biosynthesis (upper) and Valine, Luecine and Isoleucine degradation (lower).



Figure S11 Pantothenate and CoA biosynthesis.



Figure S12 Description of interpro-domain analysis: List of protein signatures identified.





Figure S13. Volcano plots for expression profiles based on sorghum drought responsive genes. This figure shows volcano plots with differential expression of genes with most significant at the top of the plot. Volcano plot (a) represents unpaired t-test based on the evaluation of tissue type contributing to the gene expression and (b) on the evaluation of treatment effect on the experimental samples. The red dots indicate a statistical significance for the up and down-regulated genes at the fold-changes (two) above which all genes have p < 0.01 and below which p > 0.01.



Figure S14 Volcano plots showing expression profiles based on based on maize expression data.

This figure describes volcano plot showing differential expression of genes with the most significant genes at the top of the plot. The red dots indicate genes-of-interest that display both large-magnitude of fold-changes (the change in mean values of group A and group B, in our case, x-axis) and a fairly high value of statistical significance (-log10 of p-value, y-axis). The upper line

across the plot shows a point where p = 0.01 (i.e. where the fold-change is equal to two (log2 = 1) above which lie all genes having p < 0.01 and below which having p > 0.01. Volcano plots a) represent unpaired t-test based on treatment grouping, b) depicts the gene expression pattern based on between subject variance with tissue based grouping of samples. The plot shows higher number of genes expressed under drought condition in tissue based grouping with more down-regulated tissue specific genes than in treatment based grouping which shows not only relatively fewer number of genes expressed in total under the same stress condition but also relatively less down-regulated genes.



Figure S15 Venn diagram showing distribution of significantly expressed genes.

Key to legend: Fisher's ET = Fishers Exact Test; Unparired tT = Unpaired t test and RP = Rank product. The figure illustrates the number of statistically significantly expressed genes identified by each statistical model which is equivalent to the sum of values in each respective circle. An overlapping shows the number of significantly expressed genes supported by multiple models. The number at the centre represents a total number of expressed genes supported by all statistical models.



Figure S16 Sorghum % GO-terms assigned genes identified from maize orthologs. Significantly expressed 45 genes supported by all statistical models based on analysis of geneexpression are shown (left) and significantly expressed total 1079 genes supported by individual statistical models based on analysis of gene-expression are also shown (right). The percentage of sorghum genes identified from maize orthologs that were assigned to different GO-terms associated to drought responses are also shown on gene expression in maize reproductive and Leaf meristem tissue.



Figure S17 Mapping of GO terms related to responses to drought stress based on the biological process of the genes identified using orthology groups. This figure represents significantly overrepresented GO-terms most related to drought tolerance selected based on tree traversing mode using AGRIGO software package. The GO term descriptors that related to drought tolerance included but not limited to "response to drought stress", "response to desiccation tolerance", "response to water deprivation", "cellular response to drought stress", "cellular response to desiccation" and "cellular response to water deprivation". Drought stress regulated genes were represented as an expression of mRNAs with more than two-fold changes under differential conditions. The number inside the coloured box represent GO accessions, p-values, number of enriched genes in the test set per total number of genes involved in the test set and those in the sorghum database associated with GO-term per total number of genes in the database involved in the background set of the GO-term.