Supporting information

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Table S1. Methodology for measuring the drought-tolerant related traits

|  |  |
| --- | --- |
| **Trait** | **Measurement** |
| **Plant height (cm)** | Measured from the base of plants at the ground to the stem tip at seedling stage. |
| **Biomass (g)** | The aerial part of the plants is dried in an oven (~80 ℃) to constant weight |
| **LAWC (%)** | (fresh mass - dry mass)/ dry mass ×100% |
| **LRWC (%)** | (fresh mass- dry mass)/ (turgid mass- dry mass)× 100% |

Table S2. Effect of drought stress (SAWS=10%) on drought tolerant related traits in Z141 and NY-17.

|  |  |  |
| --- | --- | --- |
|  | **Z141** | **NY-17** |
| **Trait** | **Control** | **Drought** | **Control** | **Drought** |
| **Plant height (cm)** | 19.00±0.21 | 13.25±0.21 | 26.23±0.03 | 17.74±0.44 |
| **Biomass (g)** | 0.64±0.02 | 0.35±0.03 | 0.22±0.01 | 0.15±0.01 |

Table S3. Effect of drought stress on LAWC and LRWC in Z141 and NY-17

|  |  |  |
| --- | --- | --- |
|  | **Z141****ASWC (%)** | **NY-17****ASWC(%)** |
| **Trait** | **70** | **50** | **30** | **10** | **0** | **70** | **50** | **30** | **10** | **0** |
| **ALWC (%)** | 87.50±0.14 | 85.22±0.18 | 84.22±0.22 | 81.13±0.37 | 72.32±1.10 | 86.17±0.19 | 86.23±0.59 | 84.25±0.32 | 83.05±0.31 | 68.34±0.80 |
| **RLWC (%)** | 75.99±0.46 | 73.45±1.35 | 72.40±1.81 | 60.54±0.58 | 31.81±0.51 | 76.23±1.60 | 70.91±2.36 | 74.49±0.93 | 56.09±0.52 | 28.74±0.77 |

Table S4. Sequence summary of PacBio SMRT Cells

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Library** | **Cell** | **Total basees (bp)** | **Reads Number** | **Mean Length (bp)** | **Read N50 (bp)** |
| Z141 1<3k | C01 | 12,903,767,113 | 472,461 | 27,312 | 49,640 |
| Z141 >3k | D01 | 15,358,282,321 | 652,592 | 23,534 | 40,804 |
| NY-17 1<3k | D01 | 16,863,639,348 | 559,270 | 30,153 | 53,923 |
| NY-17 >3k | H01 | 10,630,578,914 | 343,939 | 30,908 | 52,130 |
| Total |  | 55,756,267,696 | 2,028,262 |  |  |

Table S5. Sequence summary of PacBio subreads.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Library** | **Cell** | **Total basees (bp)** | **NO. of subreads** | **Mean Subreaad Length (bp)** |
| Z141 1<3k | C01 | 12,431,934,278 | 11,036,887 | 1,126 |
| Z141 >3k | D01 | 15,126,701,717 | 5,498,149 | 2,751 |
| NY-17 1<3k | D01 | 16,287,449,271 | 13,605,149 | 1,197 |
| NY-17 >3k | H01 | 10,513,142,382 | 3,020,688 | 3,480 |
| Total |  | 54,359,227,648 |  |  |

Table S6. Full length evaluation

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Sample** | **Library** | **Cell** | **# of CCS** | **5' primer** | **3' primer** | **Poly-A** | **Full-Length** | **# of FLNC** | **Mean FLNC Length(bp)** |
| Z141<3k | 1-3k | C01 | 388,621 | 372,744 | 372,381 | 365,490 | 353,313 | 348,808 | 1,163 |
| Z141>3k | 3k+ | D01 | 471,760 | 442,441 | 443,569 | 401,451 | 378,932 | 285,995 | 3,299 |
| NY-17<3k | 1-3k | D01 | 470,518 | 452,489 | 452,026 | 443,192 | 429,356 | 423,067 | 1,277 |
| NY-17>3k | 3k+ | H01 | 268,516 | 259,575 | 259,399 | 254,005 | 245,360 | 235,264 | 3,811 |
| Total | - | - | 1,599,415 | 1,527,249 | 1,527,375 | 1,464,138 | 1,406,961 | 1,293,134 | - |

Table S7. Gene structure annotation.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Feature | Loci | Loci <1K | Loci 1-2K | Loci 2-3K | Loci >=3K |
| Annotation.loci | 43,484 | 22,026(50.65%) | 15,465(35.56%) | 4,119(9.47%) | 1,874(4.31%) |
| PacBio.loci | 28,686 | 4,304(15.00%) | 12,932(45.08%) | 5,137(17.91%) | 6,313(22.01%) |

Table S8. Illumina RNA-seq data of each stress.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Sample | Total Reads | Total mapping rate | Left reads mapped | Left reads multiple mapped | Right reads mapped | Right reads multiple mapped |
| z141-CK-1 | 46,611,198 | 41,250,992(88.5%) | 20,748,876(89.0%) | 620,466(3.0%) | 20,502,116(88.0%) | 608,496(3.0%) |
| z141-CK-2 | 42,135,638 | 37,104,476(88.1%) | 18,604,423(88.3%) | 480,228(2.6%) | 18,500,053(87.8%) | 475,817(2.6%) |
| z141-DS-1 | 38,108,244 | 33,333,847(87.5%) | 16,772,482(88.0%) | 474,619(2.8%) | 16,561,365(86.9%) | 467,027(2.8%) |
| z141-DS-2 | 43,607,676 | 37,517,971(86.0%) | 18,782,617(86.1%) | 546,373(2.9%) | 18,735,354(85.9%) | 546,559(2.9%) |
| z141-RW-1 | 40,664,846 | 35,139,155(86.4%) | 17,797,644(87.5%) | 816,005(4.6%) | 17,341,511(85.3%) | 785,701(4.5%) |
| z141-RW-2 | 41,738,502 | 35,375,400(84.8%) | 18,174,414(87.1%) | 815,141(4.5%) | 17,200,986(82.4%) | 757,114(4.4%) |
| z141-RD-1 | 48,517,024 | 41,924,285(86.4%) | 21,117,750(87.1%) | 567,022(2.7%) | 20,806,535(85.8%) | 565,001(2.7%) |
| z141-RD-2 | 41,681,234 | 35,361,967(84.8%) | 17,679,464(84.8%) | 483,765(2.7%) | 17,682,503(84.8%) | 486,816(2.8%) |
| NY17-CK-1 | 49,471,150 | 43,313,708(87.6%) | 21,801,450(88.1%) | 598,560(2.7%) | 21,512,258(87.0%) | 587,005(2.7%) |
| NY17-CK-2 | 49,885,912 | 44,166,031(88.5%) | 22,147,899(88.8%) | 616,686(2.8%) | 22,018,132(88.3%) | 609,831(2.8%) |
| NY17-DS-1 | 42,575,312 | 37,097,611(87.1%) | 18,574,715(87.3%) | 573,381(3.1%) | 18,522,896(87.0%) | 572,676(3.1%) |
| NY17-DS-2 | 50,269,748 | 44,424,566(88.4%) | 22,361,186(89.0%) | 642,218(2.9%) | 22,063,380(87.8%) | 631,256(2.9%) |
| NY17-RW-1 | 42,248,852 | 37,632,290(89.1%) | 18,878,334(89.4%) | 472,723(2.5%) | 18,753,956(88.8%) | 468,138(2.5%) |
| NY17-RW-2 | 39,559,908 | 34,367,100(86.9%) | 17,256,409(87.2%) | 448,232(2.6%) | 17,110,691(86.5%) | 444,370(2.6%) |
| NY17-RD-1 | 44,407,456 | 38,393,679(86.5%) | 19,289,002(86.9%) | 520,149(2.7%) | 19,104,677(86.0%) | 518,670(2.7%) |
| NY17-RD-2 | 50,412,482 | 44,583,922(88.4%) | 22,430,612(89.0%) | 545,864(2.4%) | 22,153,310(87.9%) | 538,218(2.4%) |

Table S9. Classification of FLNC sequences with genome alignment.

|  |  |  |  |
| --- | --- | --- | --- |
| feature | Pre correction | Post correctio | Merge |
| unmap | 8,248(0.64%) | 6,347(0.49%) | 5,837(0.45%) |
| multiple-best | 8,617(0.67%) | 8,693(0.67%) | 8,443(0.65%) |
| low pid | 294,844(22.80%) | 197,552(15.28%) | 185,568(14.35%) |
| high quality map | 981,421(75.89%) | 1,080,538(83.56%) | 1,093,282(84.55%) |

Note :

Multiple\_best: both entire\_PID and region\_PID mapped to multiple site of genome.

low pid : entire\_PID < 94%, region\_PID<96%.

high quality map ：entire\_PID >= 94%, region\_PID >= 96%.

Merge: Integrating the results of post correction and before correction

Table S11 Comparison of our predicated TFs with that released by PlantTFDB

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **TF Family** | **Predicted in our analysis** | **PlantTFDB** |  | **TF Family** | **Predicted in our analysis** | **PlantTFDB** |
| bHLH | 322 | 195 |  | CAMTA | 41 | 9 |
| MYB-related | 306 | 108 |  | NF-YC | 40 | 15 |
| C2H2 | 273 | 136 |  | AP2/ERF-AP2 | 35 | 35 |
| C3H | 273 | 61 |  | NF-YA | 35 | 11 |
| MYB | 269 | 206 |  | BBR-BPC | 29 | 8 |
| NAC | 264 | 191 |  | NF-X1 | 29 | 4 |
| bZIP | 245 | 107 |  | CPP | 28 | 9 |
| AP2/ERF-ERF | 235 | 193 |  | GARP-ARR-B | 27 | 15 |
| B3 | 180 | 107 |  | BES1 | 26 | 12 |
| WRKY | 167 | 108 |  | C2C2-YABBY | 24 | 15 |
| GRAS | 166 | 119 |  | E2F-DP | 21 | 14 |
| GARP-G2-like | 165 | 65 |  | GRF | 21 | 16 |
| B3-ARF | 143 | 34 |  | HB-WOX | 21 | 18 |
| HB-HD-ZIP | 111 | 62 |  | C2C2-LSD | 20 | 9 |
| HB-other | 103 | 23 |  | GeBP | 19 | 12 |
| SBP | 96 | 29 |  | CO-like | 17 | 12 |
| MADS-M-type | 90 | 67 |  | Whirly | 16 | 4 |
| Trihelix | 88 | 54 |  | zf-HD | 15 | 15 |
| MADS-MIKC | 69 | 47 |  | VOZ | 14 | 4 |
| HSF | 64 | 40 |  | DBB | 13 | 9 |
| C2C2-Dof | 56 | 43 |  | SRS | 12 | 13 |
| C2C2-GATA | 56 | 35 |  | AP2/ERF-RAV | 9 | 3 |
| FAR1 | 50 | 6 |  | EIL | 8 | 4 |
| NF-YB | 46 | 32 |  | HB-PHD | 4 | 3 |
| TCP | 45 | 23 |  | S1Fa-like | 4 | 4 |

Table S16. Primers designed for RT-PCR validation

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Gene ID** | **Primer** | **Sequence (5’-3’)** | **TM (℃)** | **Length (hp)** |
| Lus10014603 | F | GCTGCACATTTGAAGGGTGG | 57.5 | 578 |
| R | ATTCCAGCCTTGGCATCGAA | 57.2 |
| Lus10021585 | F | GAACATCTCGCGTCTGCAAC | 57.0 | 593 |
| R | GCTCATCAACCTAGCCTGCA | 57.8 |
| Lus10012962 | F | GGTGGCCAATTTCTGCAAGG | 57.6 | 514 |
| R | TGTCATGACATCCCACAGCC | 57.8 |
| Lus10001832 | F | CAGTGGTGGAGCAGTCTCAG | 58.3 | 532 |
| R | TCCCCTATCCGGTTTGCCTA | 58.1 |
| Lus10004697 | F | TCCTAGAGGCAGCAACAAGC | 57.7 | 562 |
| R | ATCCACTACTTGCCCACTGC | 57.8 |
| Lus10001016 | F | TTCCTGTTCTGGGTCATGCC | 57.8 | 572 |
| R | GCCTCTCAATATCCACCGGG | 58.3 |
| Lus10038490 | F | CAGGTCTCATCAGGAAGCCC | 58.3 | 521 |
| R | CAGGCATTGTTGTTGACCGG | 57.4 |
| Lus10027966 | F | CAGCTATGGGGAAGTAGCCG | 58.3 | 539 |
| R | GGGTCATCAGCCAATCCCAA | 57.9 |

Table S10. The list of Z141 and NY-17 DEGs under DS or RD

Table S12. Detail lists of 15 clusters of differentially expressed transcription factors.

Table S14. All sample FPKM sheet

Table S15. All sample reads count sheet