

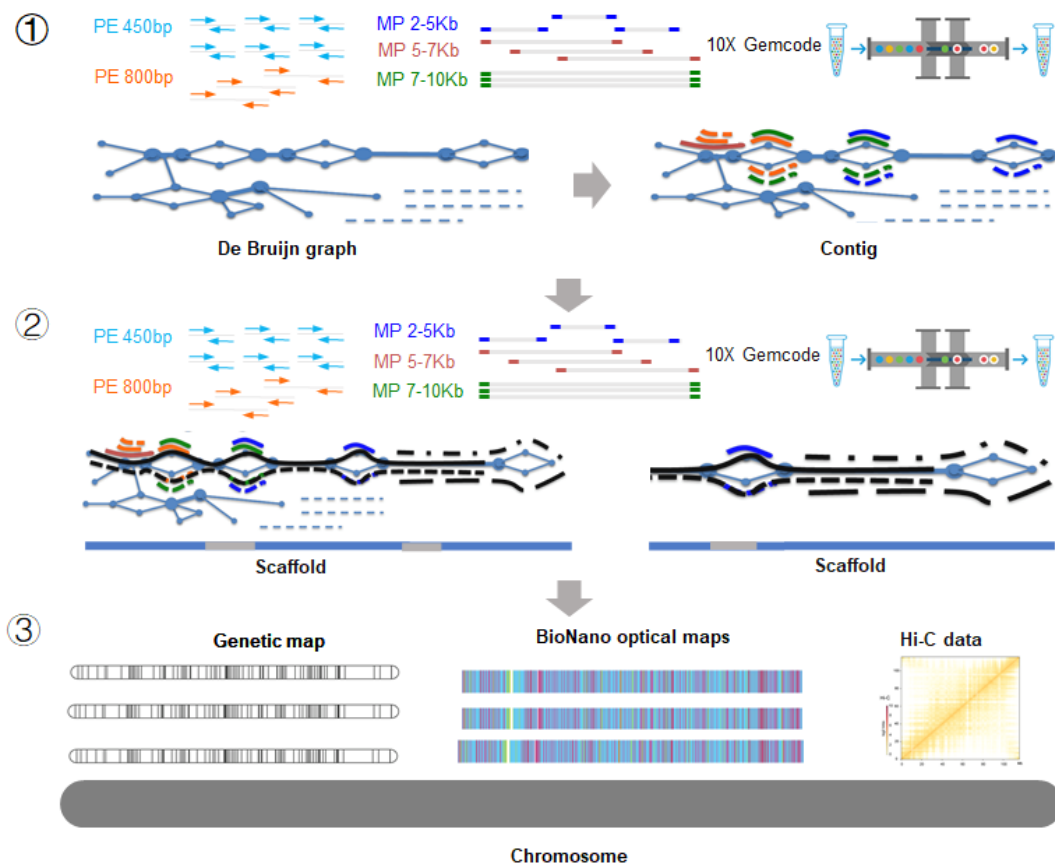
In the format provided by the authors and unedited.

Gossypium barbadense and *Gossypium hirsutum* genomes provide insights into the origin and evolution of allotetraploid cotton

Yan Hu^{1,2,11}, Jiedan Chen^{1,11}, Lei Fang^{1,2,11}, Zhiyuan Zhang^{1,11}, Wei Ma^{1,11}, Yongchao Niu^{3,11}, Longzhen Ju², Jieqiong Deng², Ting Zhao^{1,2}, Jinmin Lian³, Kobi Baruch⁴, David Fang⁵, Xia Liu⁶, Yong-ling Ruan^{1,7}, Mehboob-ur Rahman⁸, Jinlei Han⁹, Kai Wang⁹, Qiong Wang², Huitong Wu², Gaofu Mei², Yihao Zang², Zegang Han², Chenyu Xu², Weijuan Shen², Duofeng Yang², Zhanfeng Si¹, Fan Dai¹, Liangfeng Zou³, Fei Huang³, Yulin Bai⁶, Yugao Zhang⁶, Avital Brodt⁴, Hilla Ben-Hamo⁴, Xiefei Zhu², Baoliang Zhou², Xueying Guan^{1,2}, Shuijin Zhu¹, Xiaoya Chen¹⁰ and Tianzhen Zhang^{1,2*}

¹Institute of Crop Science, Plant Precision Breeding Academy, Zhejiang Provincial Key Laboratory of Crop Genetic Resources, College of Agriculture and Biotechnology, Zhejiang University, Hangzhou, China. ²State Key Laboratory of Crop Genetics and Germplasm Enhancement, Nanjing Agricultural University, Nanjing, China. ³Genosys, Inc., Shenzhen, China. ⁴NRGene, Ltd, Ness Ziona, Israel. ⁵Cotton Fiber Bioscience Research Unit, US Department of Agriculture–Agricultural Research Service–Southern Regional Research Center, New Orleans, LA, USA. ⁶Esquel Group, Wanchai, Hong Kong, China. ⁷School of Environmental and Life Sciences and Australia–China Research Centre for Crop Improvement, University of Newcastle, Newcastle, New South Wales, Australia. ⁸Plant Genomics and Molecular Breeding Laboratory, National Institute for Biotechnology and Genetic Engineering (NIBGE), Faisalabad, Pakistan. ⁹Center for Genomics and Biotechnology, Fujian Provincial Key Laboratory of Haixia Applied Plant Systems Biology, Haixia Institute of Science and Technology, Fujian Agricultural and Forestry University, Fuzhou, China. ¹⁰National Center for Gene Research, State Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Shanghai Institute of Plant Physiology and Ecology, Chinese Academy of Sciences, Shanghai, China. ¹¹These authors contributed equally: Yan Hu, Jiedan Chen, Lei Fang, Zhiyuan Zhang, Wei Ma, Yongchao Niu. *e-mail: cotton@zju.edu.cn

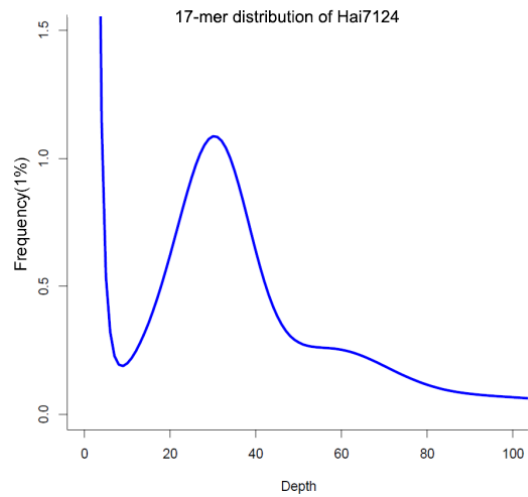
Supplementary Figures



Supplementary Figure 1

The flowchart of contig, scaffold and chromosome assembly in this study.

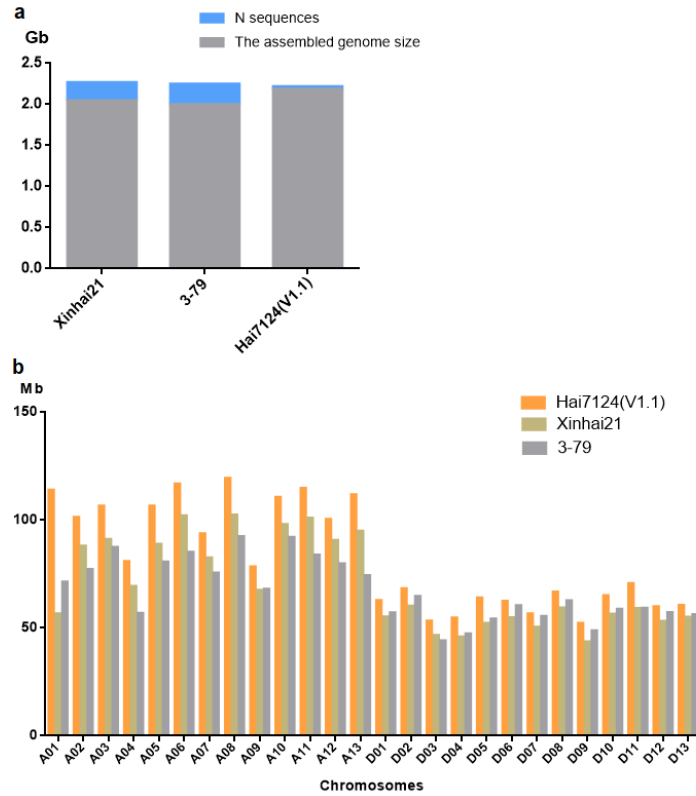
Step 1. Contig assembly and refinement. Using PE and MP reads to generate De Bruijn graph; Noise filtering by mapping reads with varying insert size (including 10× barcoded reads) on contig graph. **Step 2. Scaffold assembly, elongation and refinement.** Iterative resolve of duplication based on reliable paths identification; Using PE, MP links and De Bruijn graph information to detect a unique path connecting the gap edges; Using 10× barcoded reads for elongation and refinement. **Step 3. Chromosomes assembly.** Using genetic map, BioNano optical map and Hi-C-based chromatin interaction map to correct and order scaffolds, and finally build chromosomes efficiently.



Supplementary Figure 2

17-mers depth distribution of the sequence data from libraries with 800-bp insert size in Hai7124.

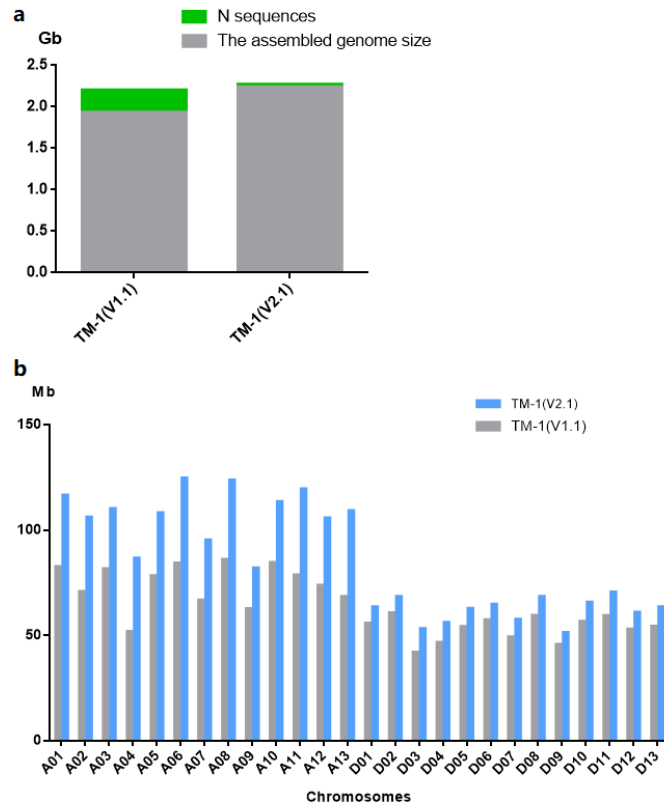
The peak depth was at 30 for Hai7124. The genome size of *G. barbadense* cv. Hai7124 is estimated to be 2.43 Gb on the basis of the formula “genome size = total_kmer_num/kmer_depth”.



Supplementary Figure 3

Comparison of the size of assembled genomes Hai7124 (V1.1) and the previous released *G. barabdense* genome assemblies (Xinhai21(ref. 5) and 3-79(ref. 7)).

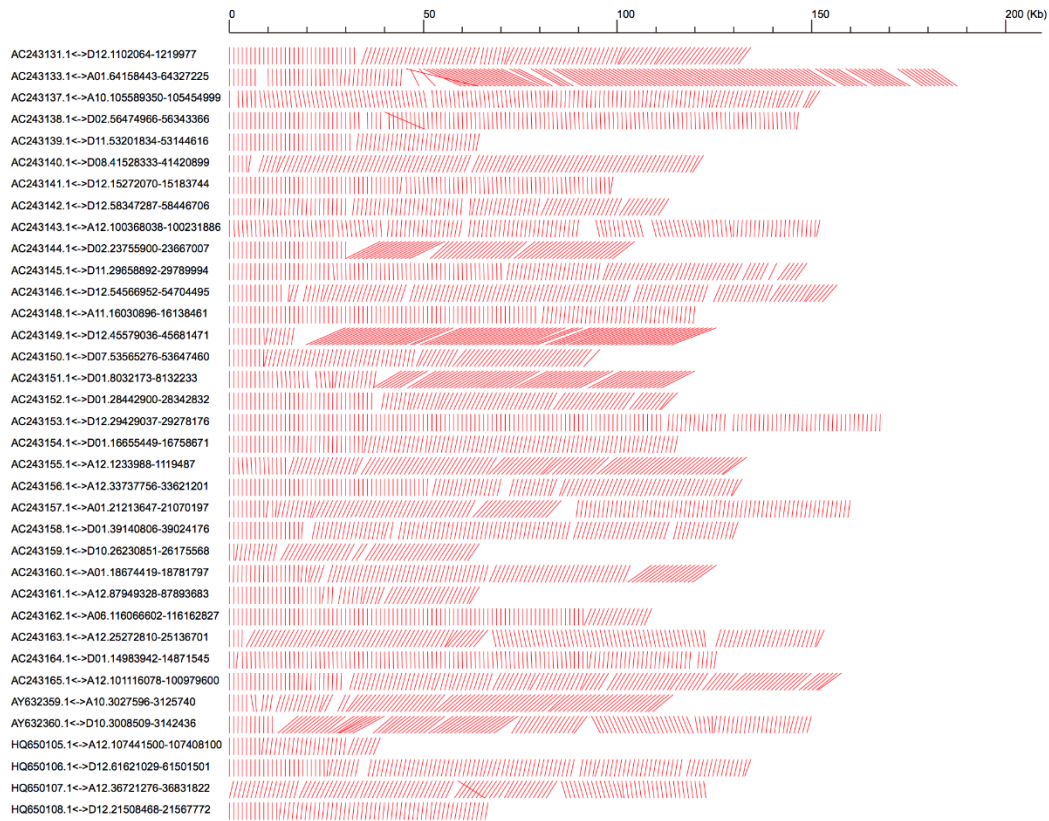
(a) Comparison of the total size of assembled genome and the connective N sequences. **(b)** Comparison of the assembled size of each chromosome without N string. The chromosome size was remarkably increased compared to the previous versions.



Supplementary Figure 4

Comparison of the assembled genome sizes of TM-1 (V2.1) and TM-1 (V1.1)⁴.

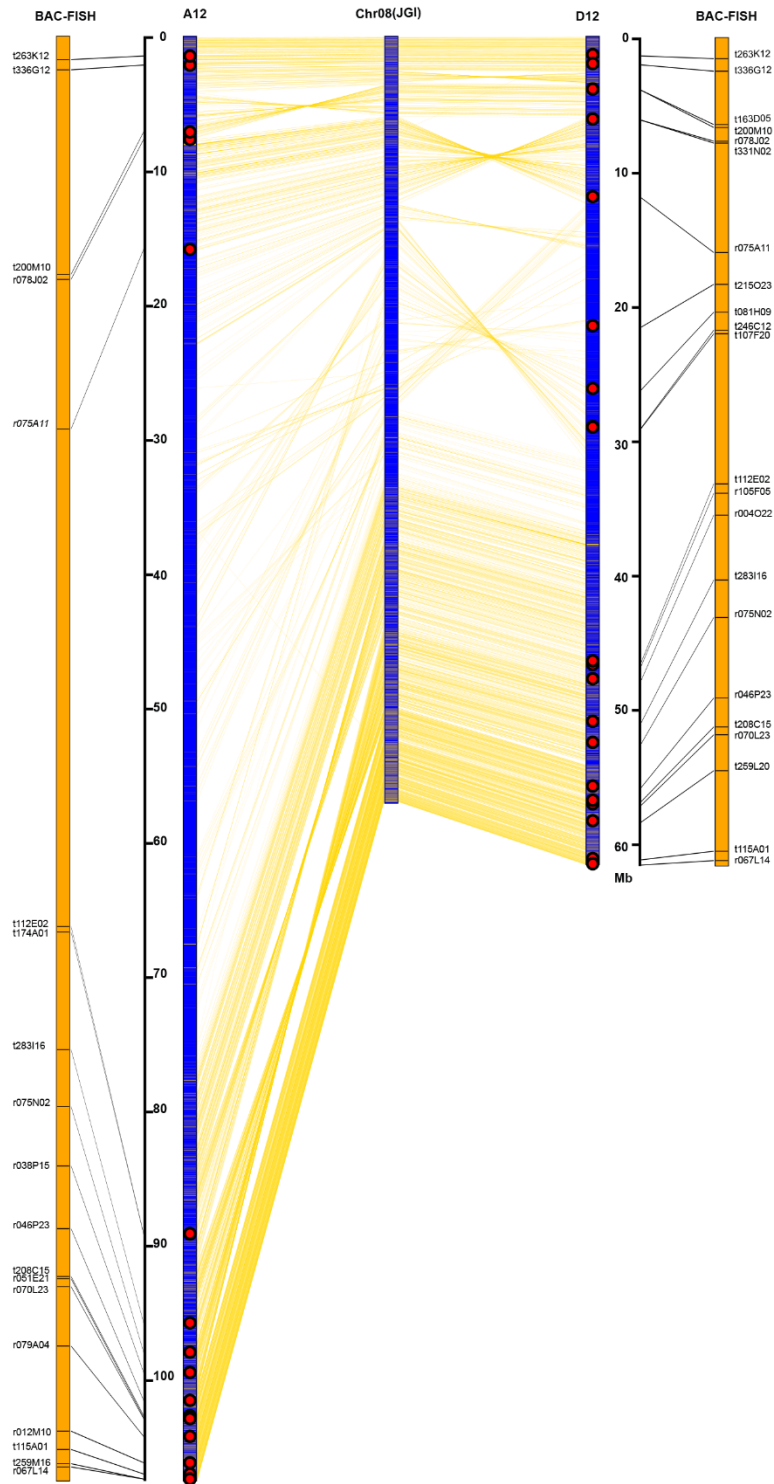
(a) Comparison of the total size of assembled genome and the connective N sequences. **(b)** Comparison of the assembled size of each chromosome without N insertion. The chromosome size was remarkably increased compared to the previous version.



Supplementary Figure 5

Assessment of the TM-1 genome assembly (V2.1) using whole sequence of 36 BACs from *G. hirsutum*.

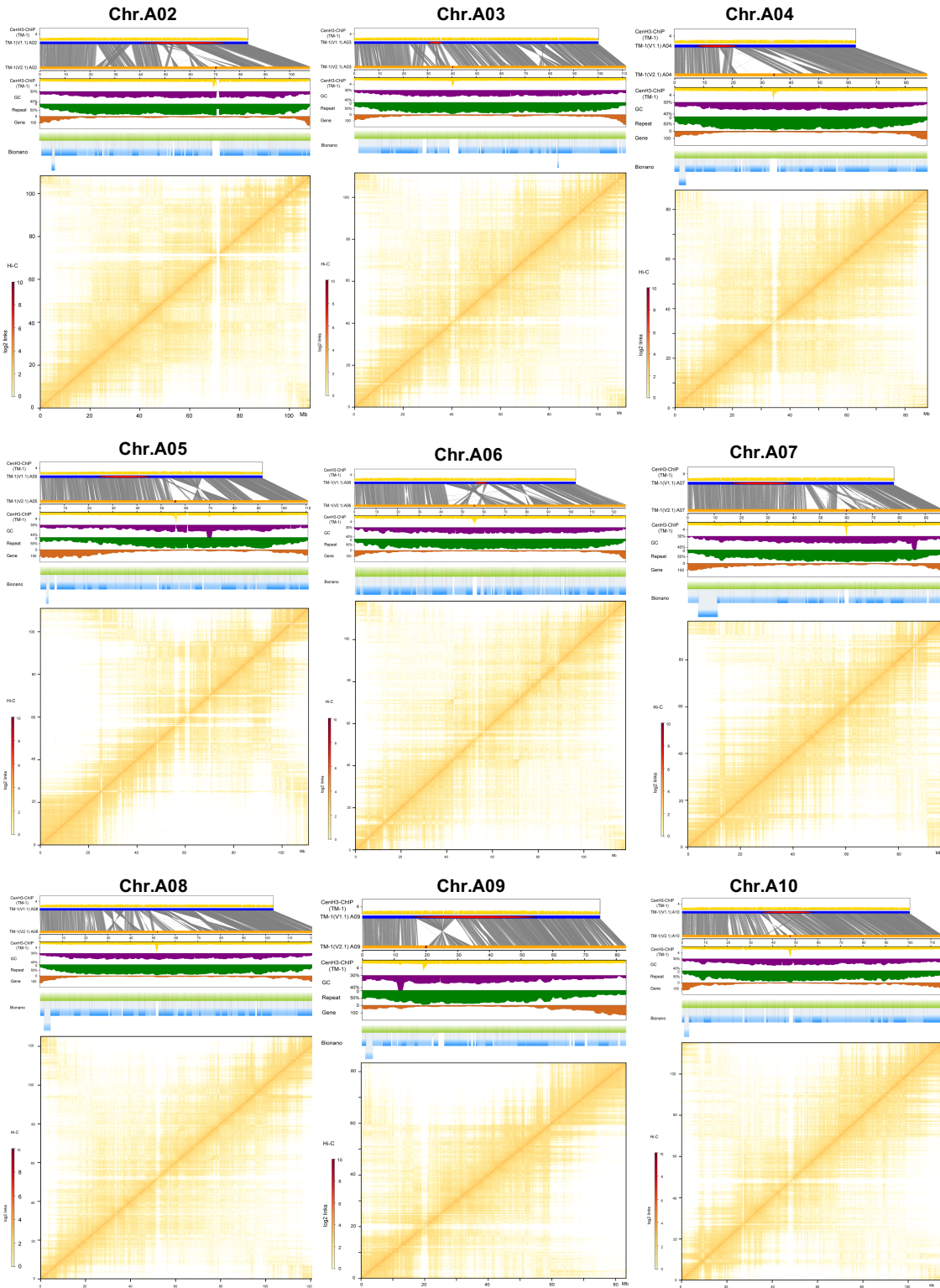
Each red line represents a 1-kb sequence. The upper end of the red line represents the position on the BAC sequences, and the lower end of the red line represents the position on the TM-1 (V2.1) assembly.

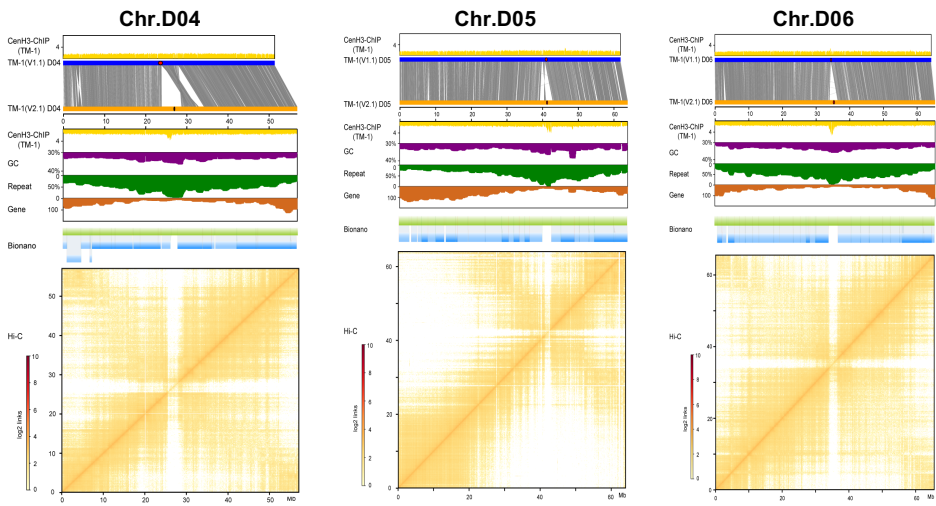
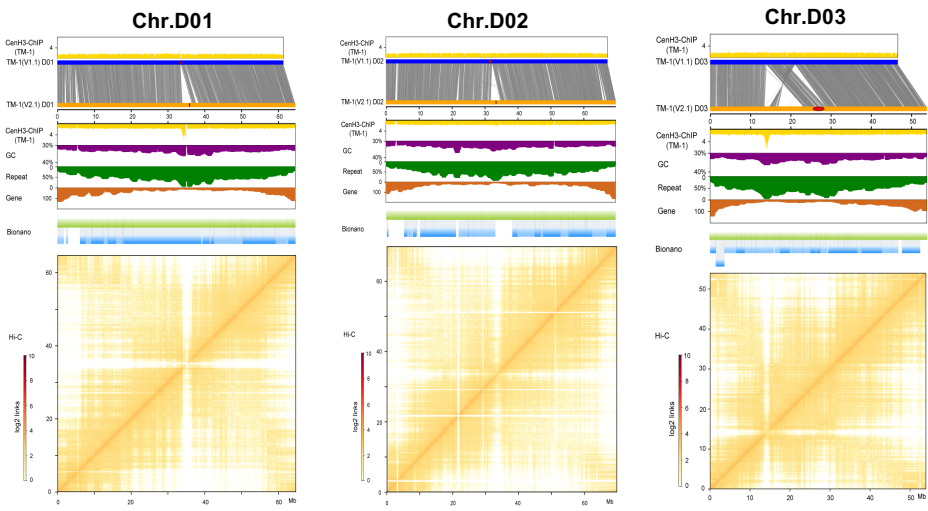
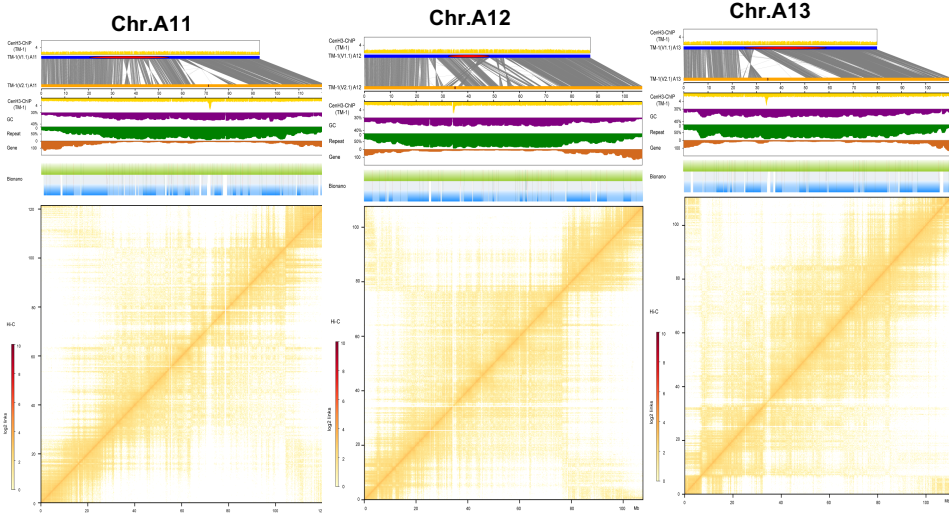


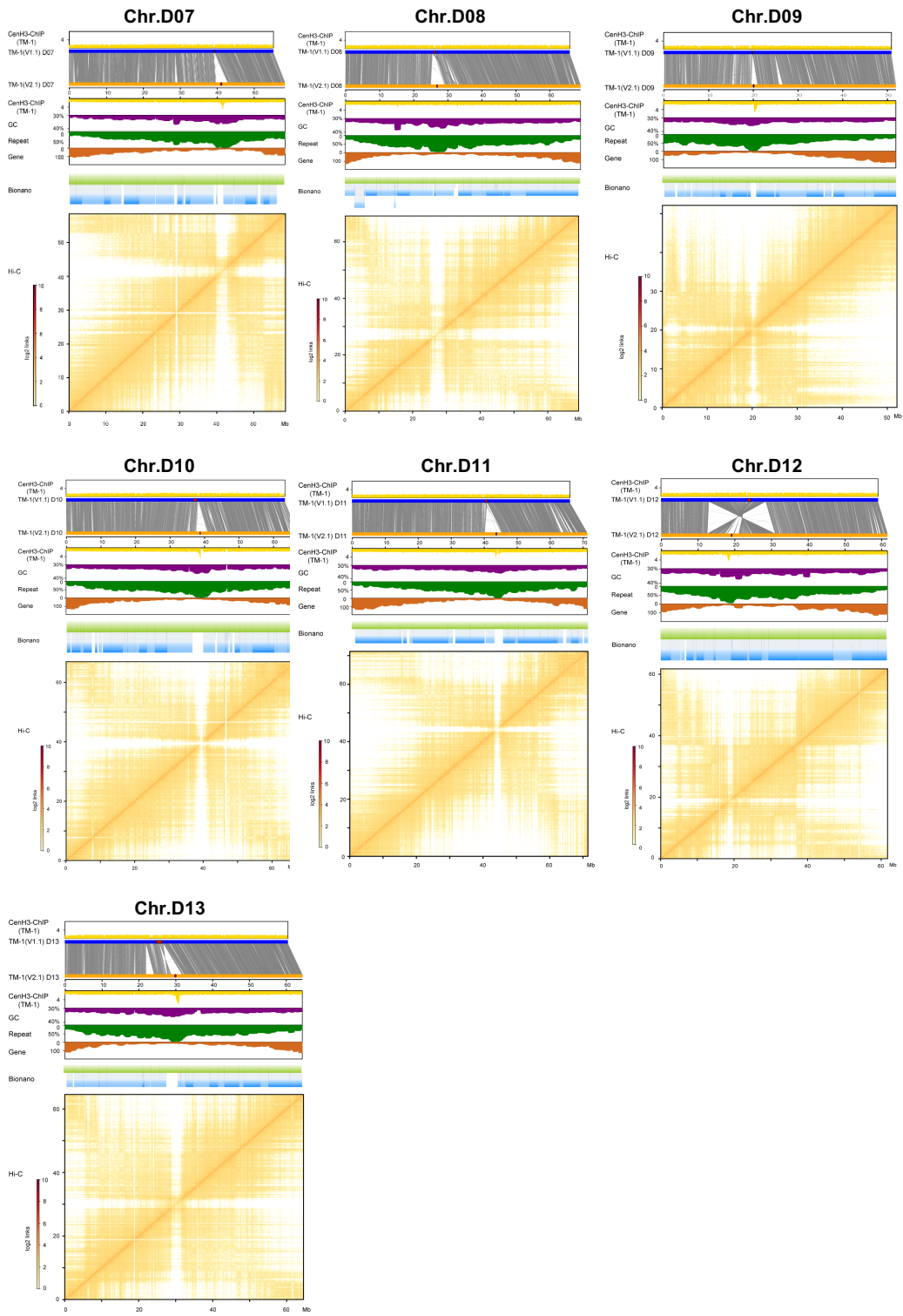
Supplementary Figure 6

Assessment of the TM-1 assembly using BAC-FISH cytogenetic maps of A12 and D12 homoeologous chromosomes.

Yellow lines, synthetic gene pairs; red circle, physical position of BACs on chromosome A12 and D12; black bars, relative position of BACs on cytogenetic map of A12 and D12 (ref.⁴).



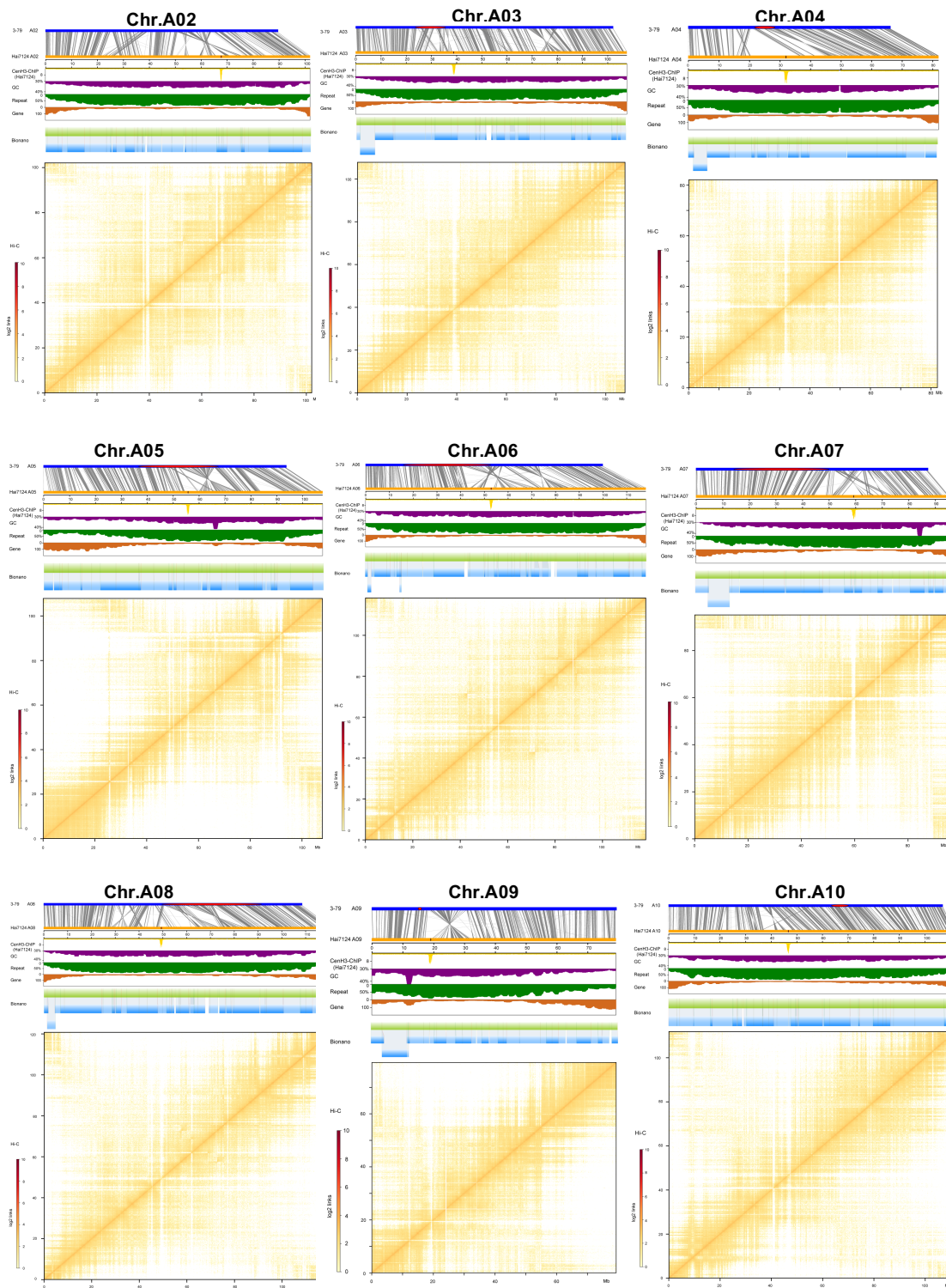


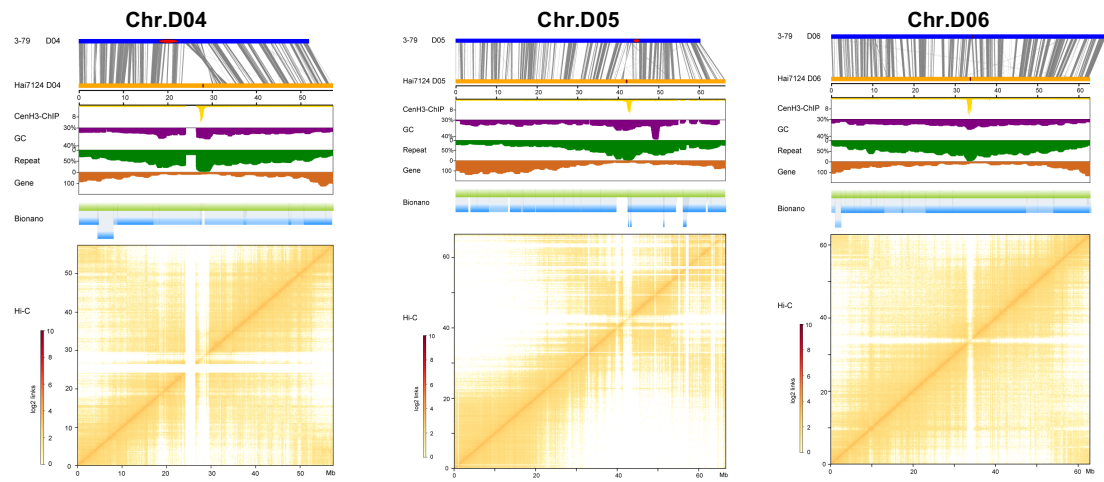
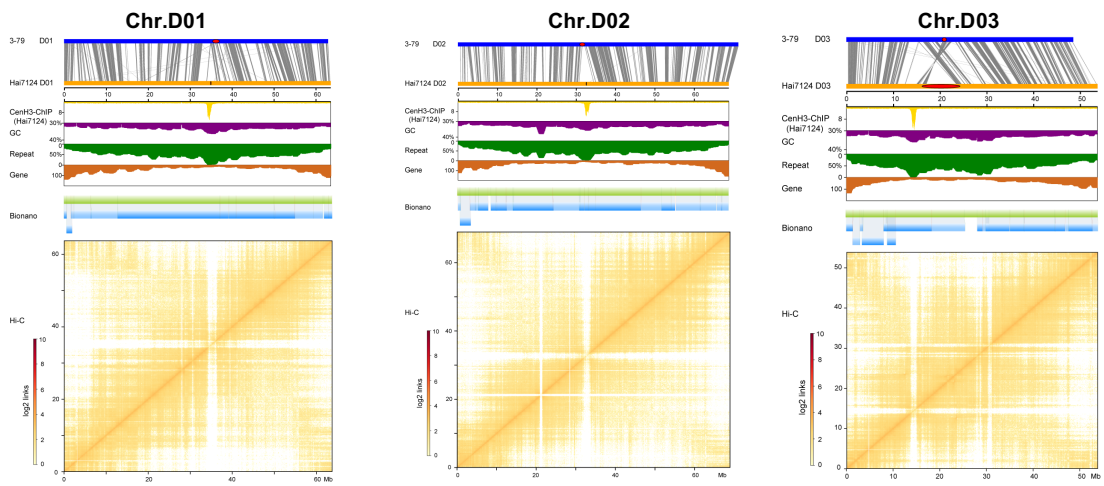
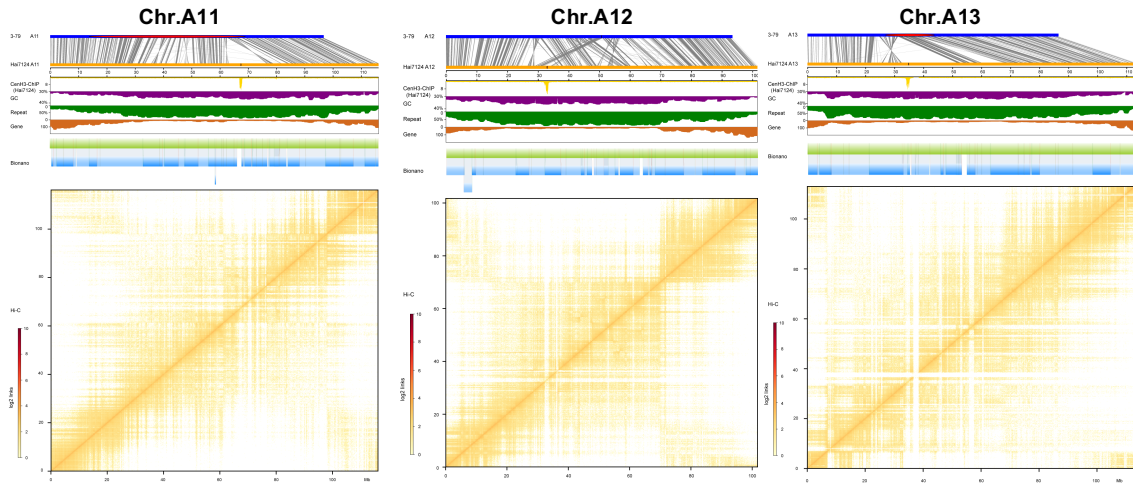


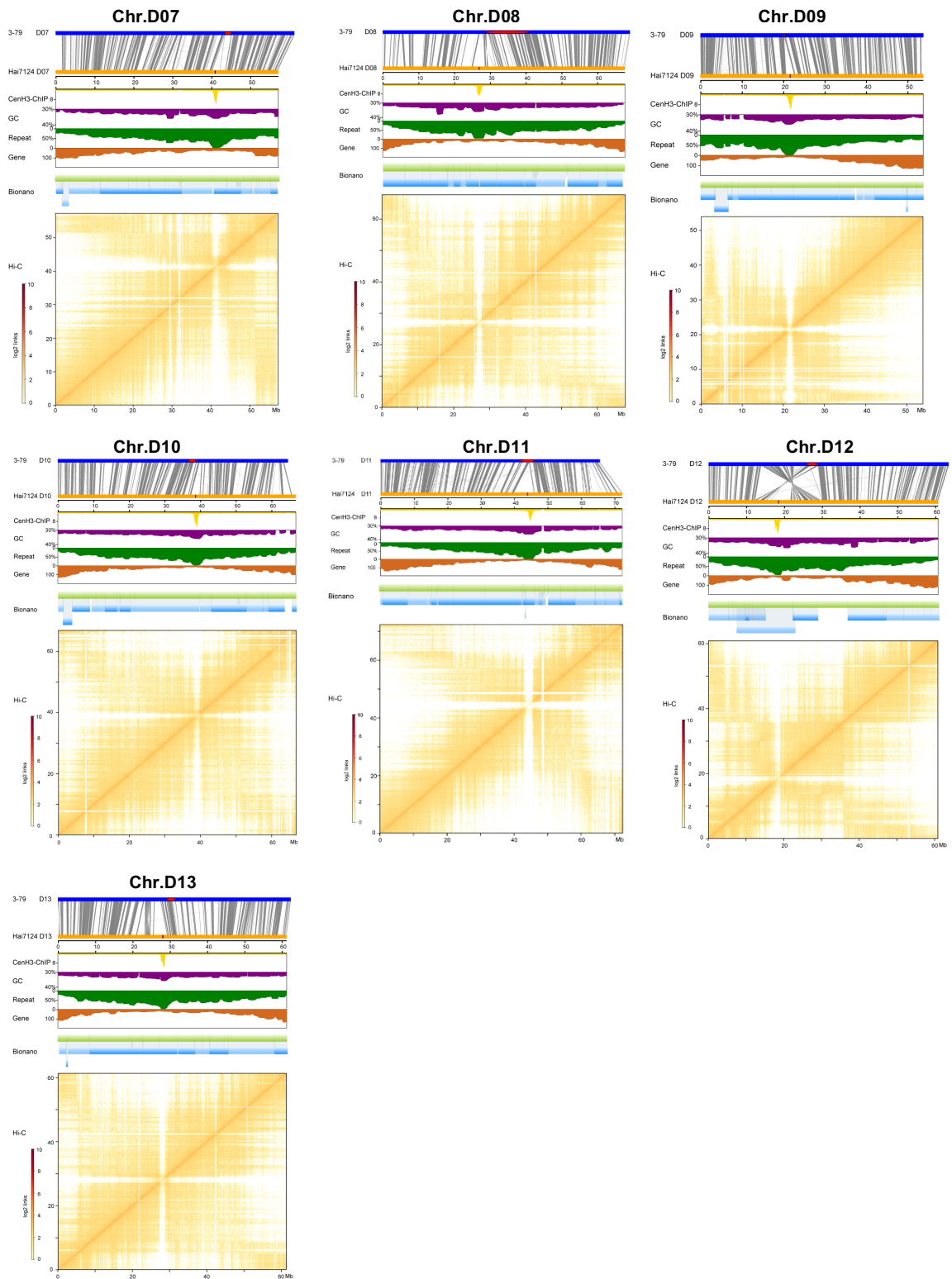
Supplementary Figure 7

Collinear comparison between remaining 25 chromosomes of TM-1(V1.1)⁴ and TM-1 (V2.1) genome.

Red region in the blue and orange bar indicates the centromere regions identified by the alignment of CRGs. Gray lines indicate the collinearity between two genomes. Each line of >5 kb unique mapping alignment. The peak on the yellow bar shows the CenH3 binding region on each chromosome of TM-1(V2.1) genome. Adjusted ChIP-seq reads was calculated in a 100-kb sliding window at 10 kb intervals. GC content, repeat distribution and gene density in a 1-Mb sliding window at 100-kb intervals. Comparison of TM-1 Bionano optical maps (blue) and TM-1(V2.1) genome (green). TM-1 Hi-C contact data mapped on the TM-1(V2.1) genome. The heat map represents the normalized contact matrix. Collinear comparison on each chromosome of TM-1(V1.1)⁴ and TM-1 (V2.1) genome.





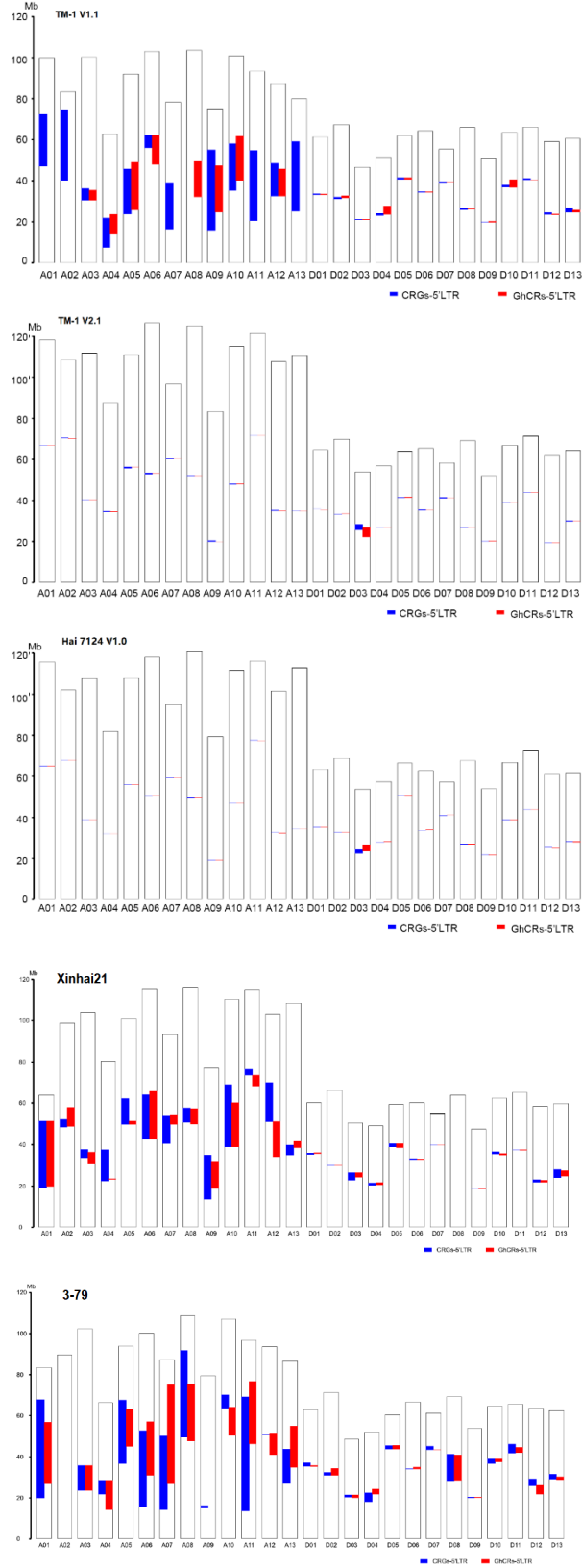


Supplementary Figure 8

Collinear comparison between remaining 25 chromosomes of Hai7124 (V1.1) and 3-79 genome⁷.

Red region in the blue and orange bar indicates the centromere regions identified by the alignment of CRGs. Gray lines indicate the collinearity between two genomes. Each line of >5 kb unique

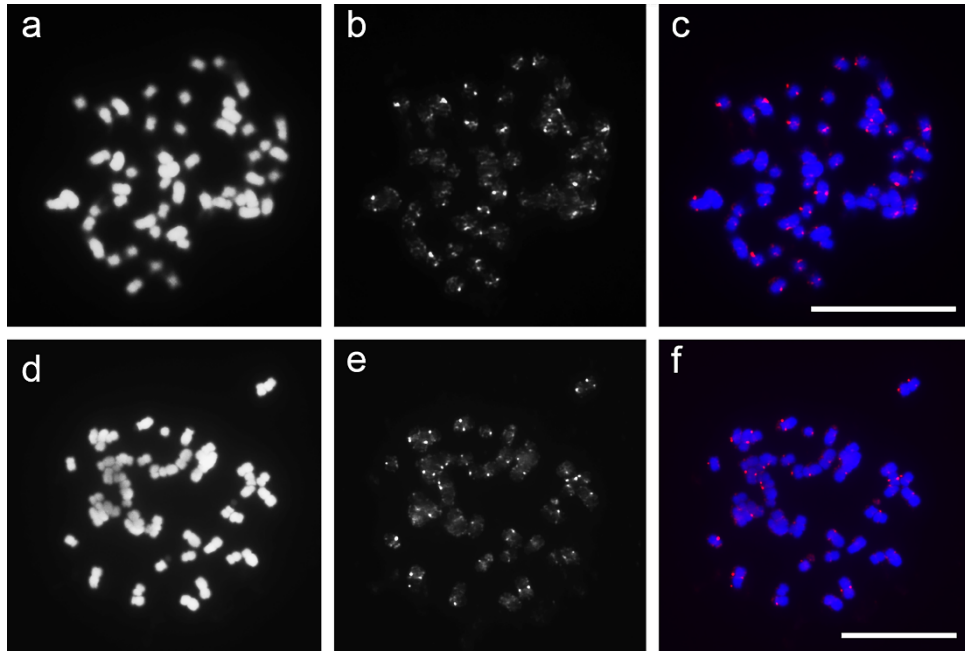
mapping alignment. The peak on the yellow bar shows the CenH3 binding region on each chromosome of Hai7124 (V1.1) genome. Adjusted ChIP-seq reads was calculated in a 100-kb sliding window at 10 kb intervals. GC content, repeat distribution and gene density in a 1-Mb sliding window at 100-kb intervals. Comparison of Hai7124 Bionano optical maps (blue) and Hai7124 (V1.1) genome (green). Hai7124 Hi-C contact data mapped on the Hai7124 (V1.1) genome. The heat map represents the normalized contact matrix. Collinear comparison on each chromosome of Hai7124 (V1.1) and 3-79 (ref. 7) genome.



Supplementary Figure 9

Schematic diagrams of centromeric regions of *G. hirsutum* acc. TM-1(V1.1 and V2.1), *G. barbadense* cv. Hai7124 (V1.1), Xinhai21(ref.⁵) and 3-79 (ref.⁷).

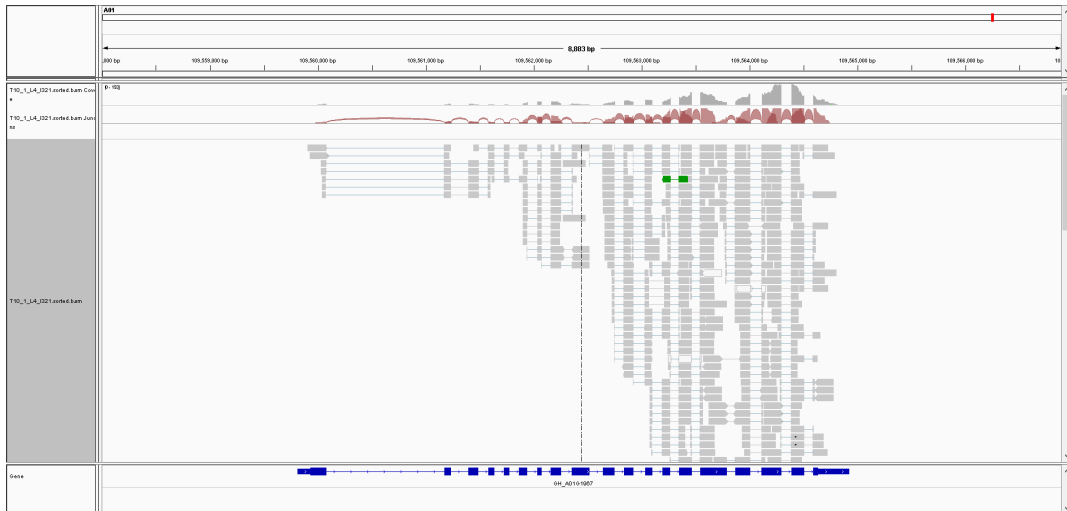
Centromeric regions of the A and D subgenome were identified by CRGs-5'LTR (blue) and GhCRs-5'LTR (red).



Supplementary Figure 10

FISH assay of the enrichment and localizations of TM-1 and Hai7124 CenH3 ChIP DNAs.

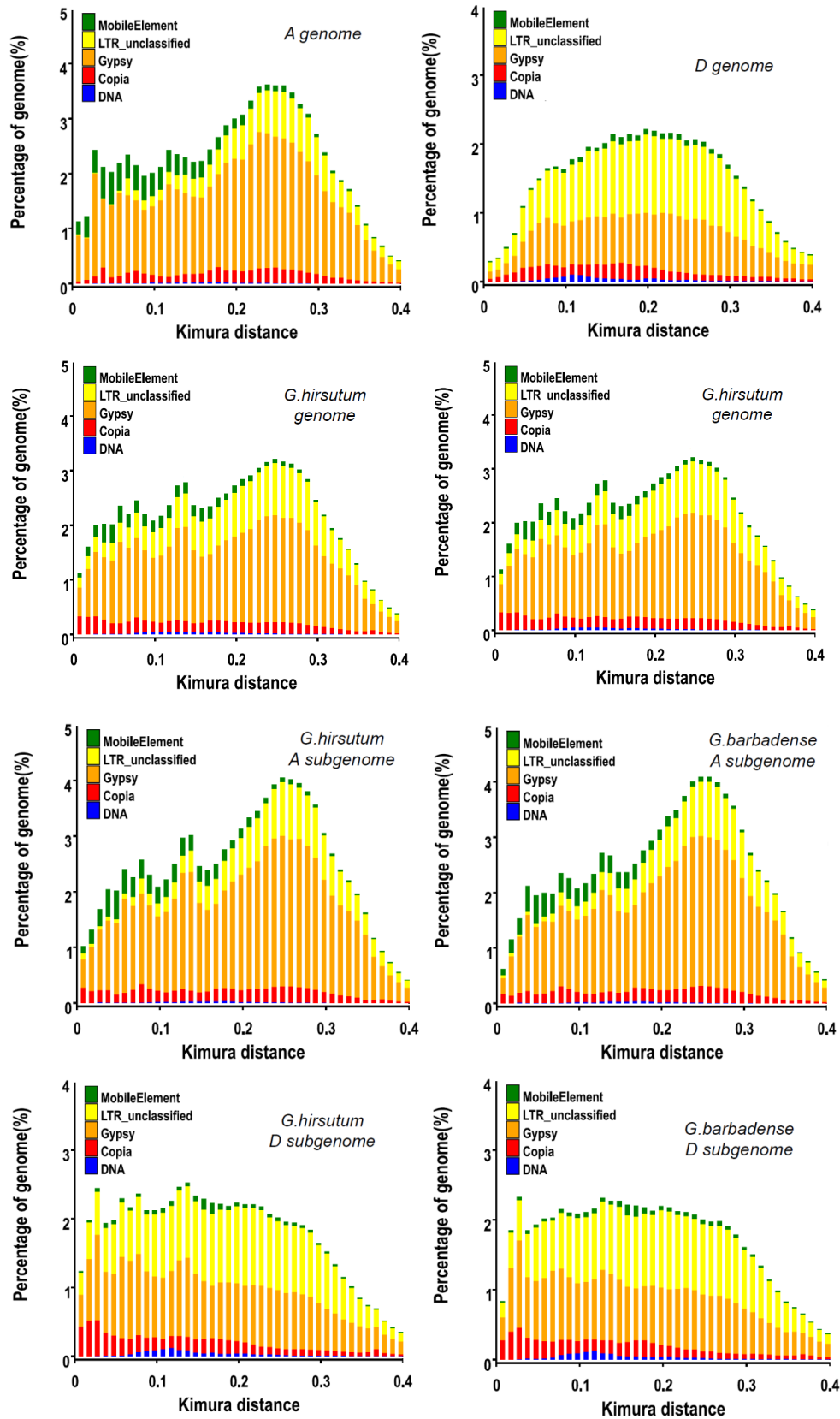
(a, d) Somatic chromosomes of TM-1 and Hai7124. **(b, e)** Somatic chromosomes were hybridized in FISH using labeled ChIP DNAs of TM-1 and Hai7124. **(c, f)** FISH signals were detected from the centromeres for the ChIP probes in TM-1 and Hai7124. Chromosomes were counterstained with DAPI **(c and f)**. Scale bar = 10 μm .



Supplementary Figure 11

An example for validating the accuracy of gene structural annotation using the mapped RNA-seq reads.

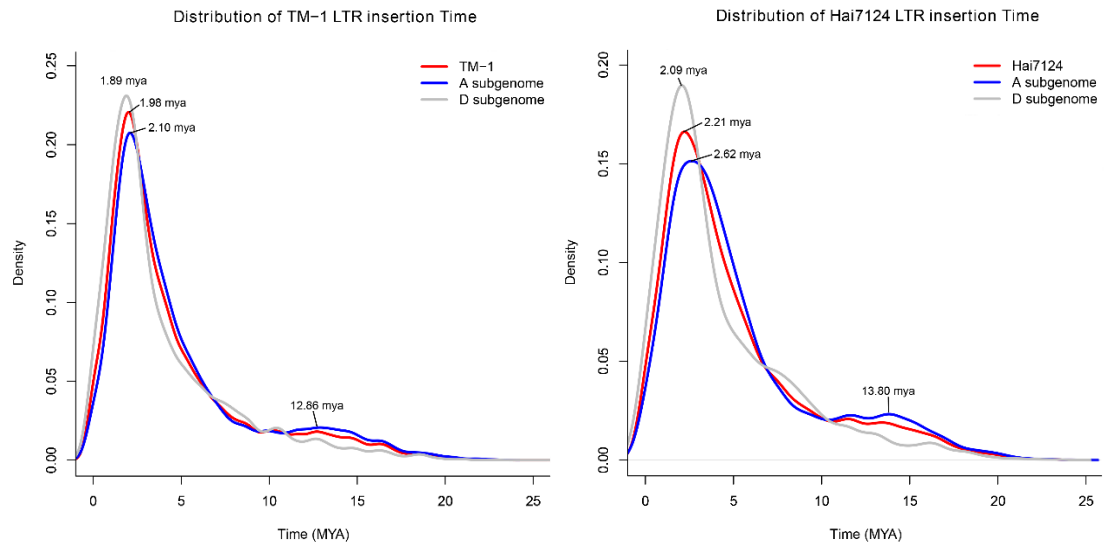
RNA-seq reads from TM-1 and Hai7124 were mapped to TM-1 and Hai7124 genome sequences, respectively. We checked a total of 100 genes (50 genes from TM-1 and 50 from Hai7124) covered by transcriptomic data by Integrative Genomics Viewer (IGV, <http://software.broadinstitute.org/software/igv/>). This figure took gene *GH_A01G1967* as an example.



Supplementary Figure 12

Distribution of Kimura distance of each type of TE in A-diploid, D-diploid, TM-1 and Hai7124 genome.

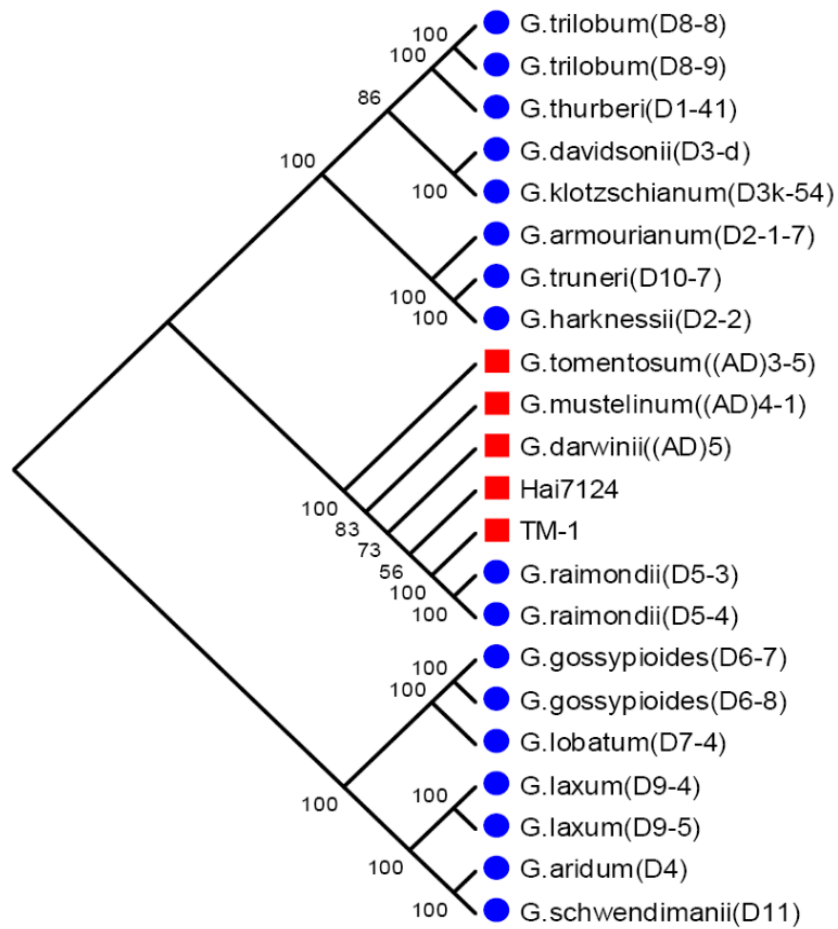
The Kimura distance was calculated between the repeat elements in the genome and the consensus sequence in the MIPS repeat database by RepeatMasker. Relative proportions of DNA transposons (blue), Copia elements (red), Gypsy elements (orange), unclassified LTR elements (yellow) and unclassified transposons (green) are displayed as stacked bar chart.



Supplementary Fig. 13

The insertion time of LTR retrotransposons in TM-1 and Hai7124.

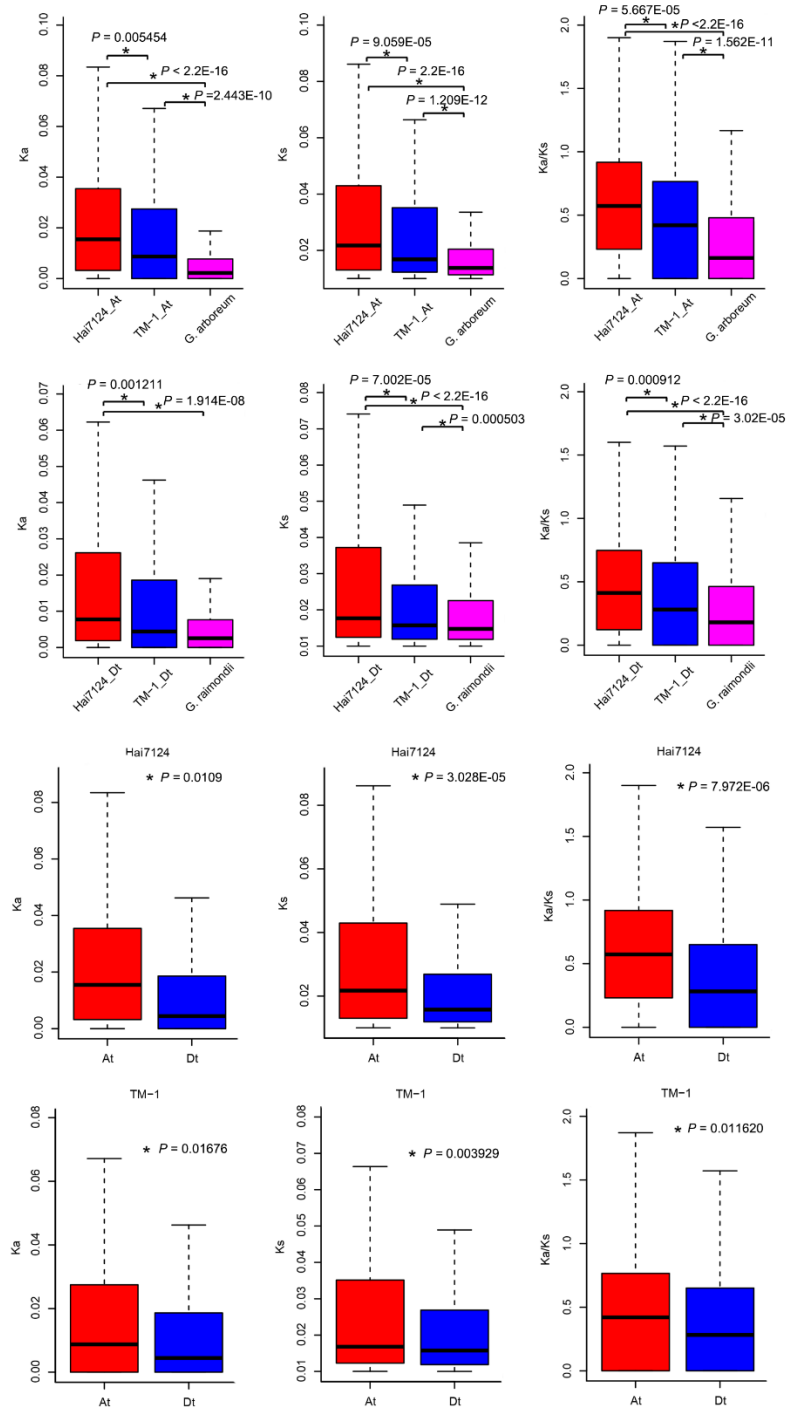
TM-1 appeared to have undergone a surge of retrotransposon amplification approximately 1.98 MYA (A subgenome: 2.10 MYA, D subgenome: 1.89 MYA), the second minor peak occurred at about 12.86 MYA. Hai7124 appeared to have undergone a surge of retrotransposon amplification approximately 2.21 MYA (A subgenome: 2.62 MYA, D subgenome: 2.09 MYA), the second minor peak occurred at about 13.80 MYA ago. The insertion time was estimated as $T = K/2\mu$ (K is the divergence rate, and μ is the neutral mutation rate; a neutral substitution rate of 2.6×10^{-9} was used for cotton) in the LTR_retriever package.



Supplementary Figure 14

Phylogenetic analysis of 17 accessions representing 13 D-genome diploid species and five AD-genome allotetraploids.

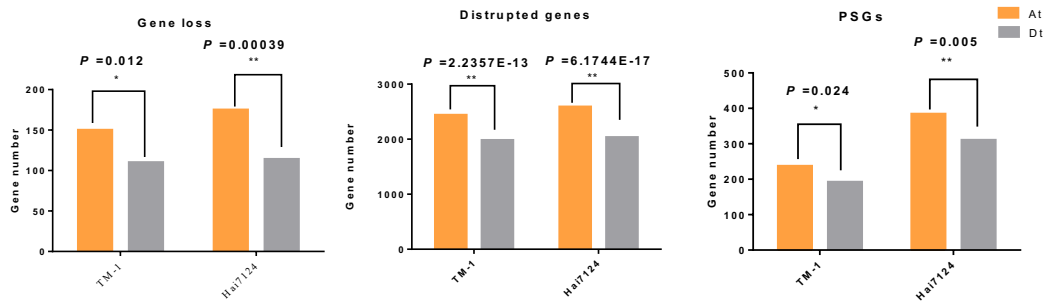
The phylogenetic tree was constructed using the Neighbor-Joining method. Bootstrap values are indicated on the nodes.



Supplementary Figure 15

Comparison of codon substitution rate distributions between two subgenomes of TM-1 and Hai7124 and their progenitors, *G. raimondii* and *G. arboreum*.

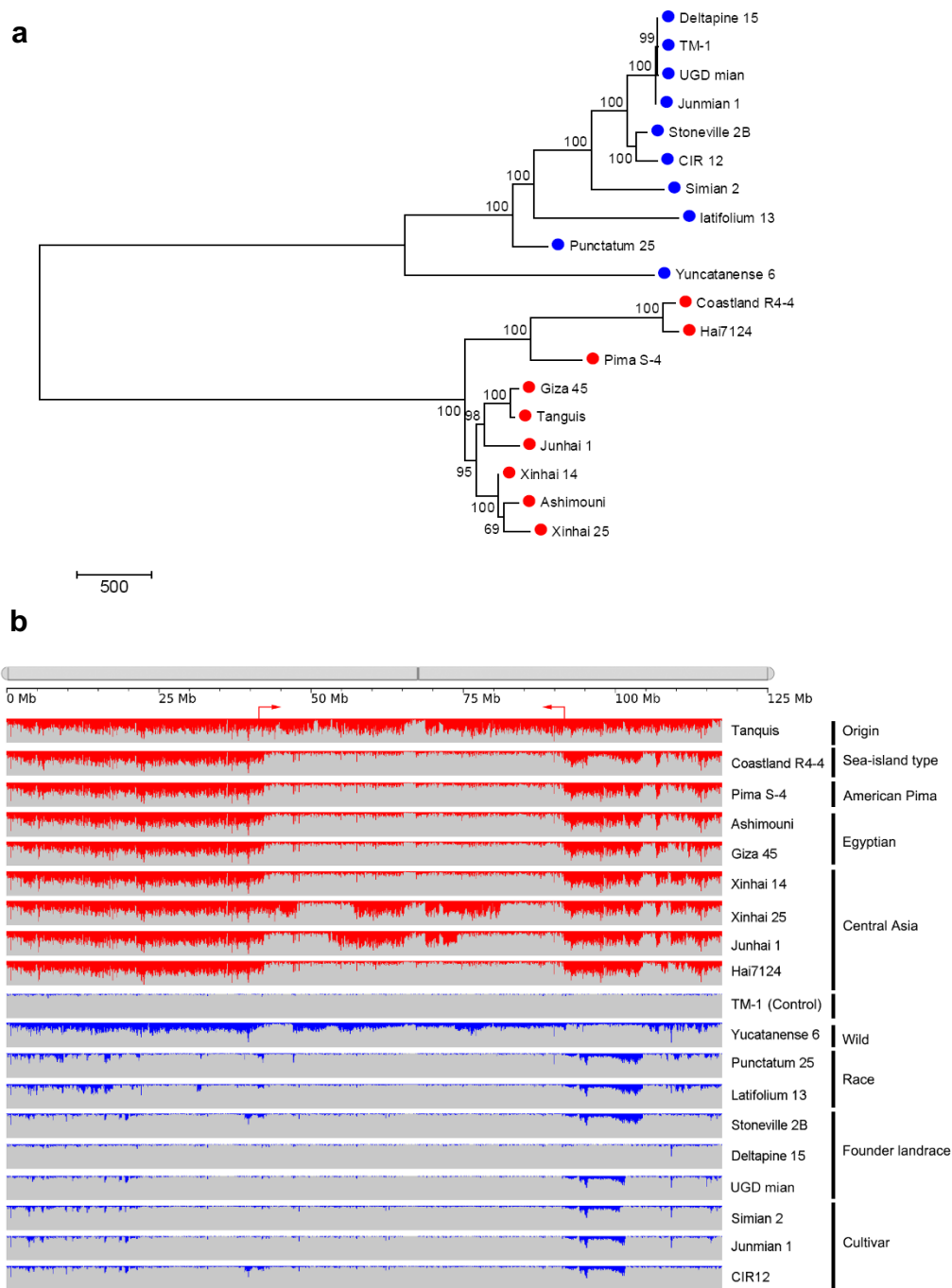
The comparison of K_a , K_s and K_a/K_s distribution between subgenomes and their corresponding progenitor genomes *G. raimondii*⁸ and *G. arboreum*¹⁰. All estimations with $K_s < 0.01$ were removed from the analysis. The central line for each box plot indicated median. The top and bottom edges of the box indicated the 25th and 75th percentiles and the whiskers extend 1.5 times of the interquartile range beyond the edges of the box. The asterisks represented significant difference (* Wilcoxon rank sum test, $P < 0.05$).



Supplementary Figure 16

Comparison of the number of gene loss, disrupted genes and positively selected genes (PSGs) on the A and D subgenome of TM-1(V2.1) and Hai7124 (V1.1).

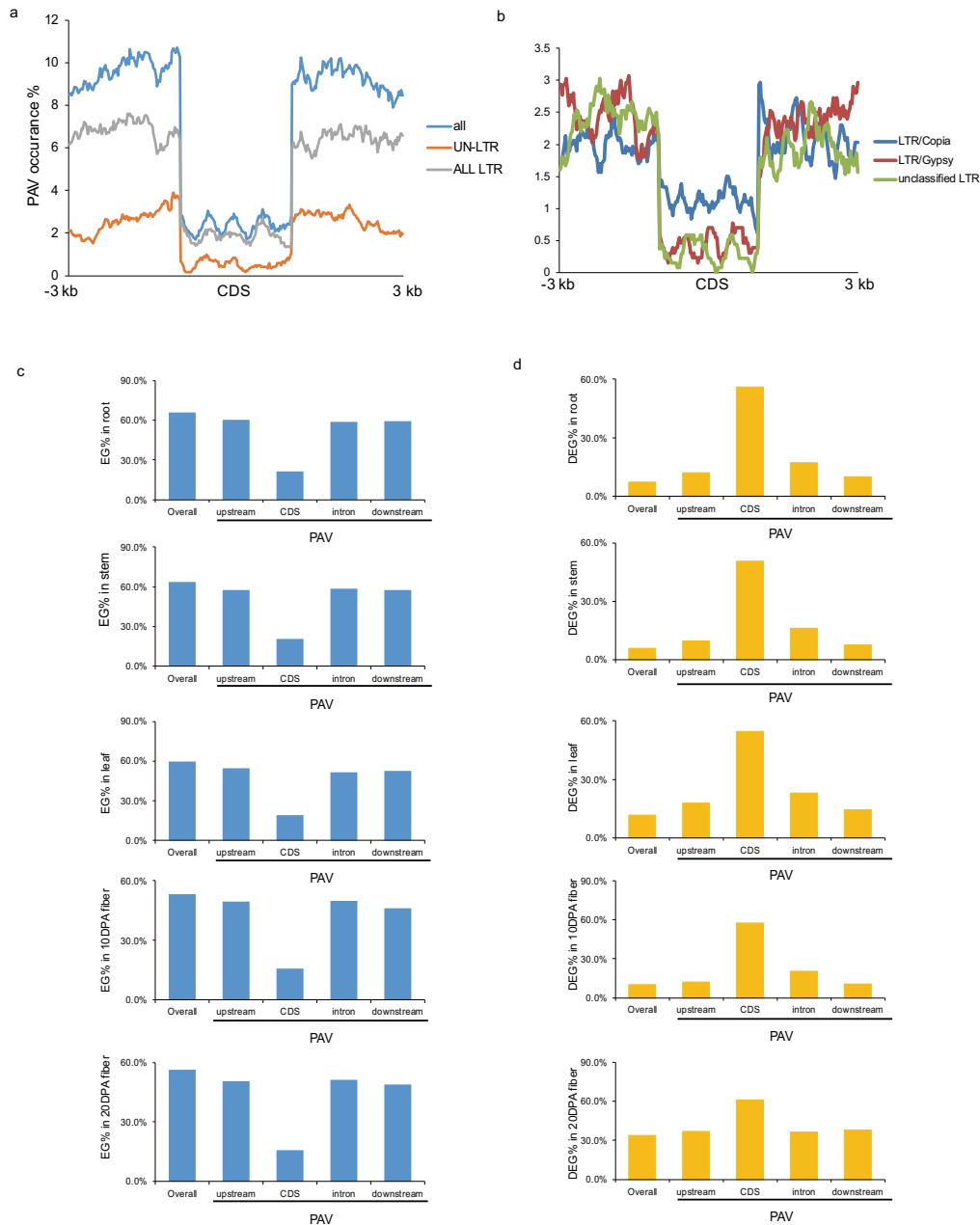
*P < 0.05, ** P<0.01, Fisher's exact tests.



Supplementary Figure 17

Phylogenetic relationships and introgression among 19 cotton accessions including 10 *G. hirsutum* and nine *G. barbadense*.

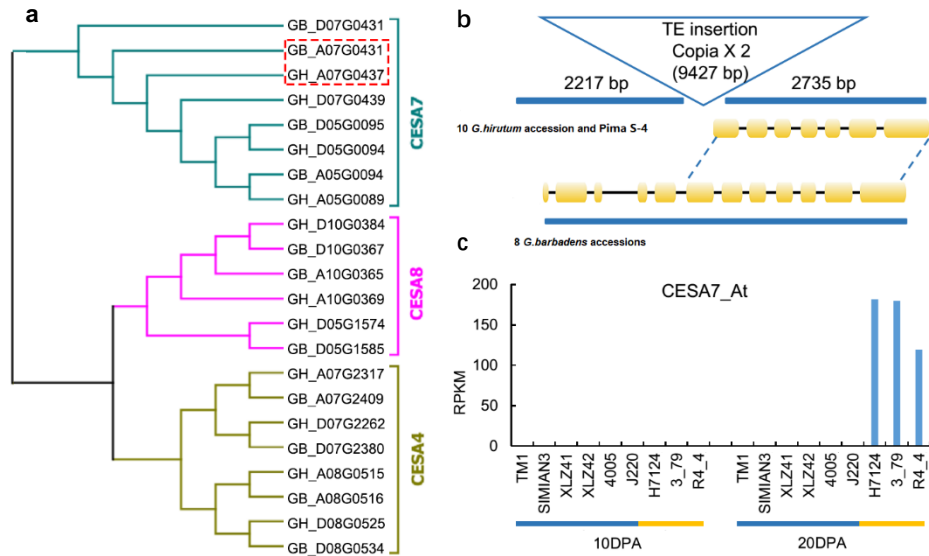
(a) A neighbor-joining tree of 10 *G. hirsutum* and nine *G. barbadense* accessions was constructed using 20,000 randomly selected whole-genome SNP data. Bootstrap values are indicated on the nodes. (b) SNP distributions in Chromosome A01 in cotton species. SNP numbers were calculated in each 100-kb window. Red and blue bar indicated SNP number called from the nine *G. barbadense* and 10 *G. hirsutum* accessions by mapped to TM-1 reference genome, respectively. The SNP distribution in TM-1 was used as control. The introgression region was between the red arrows.



Supplementary Fig. 19

Distribution of PAVs and their influences on genes expression.

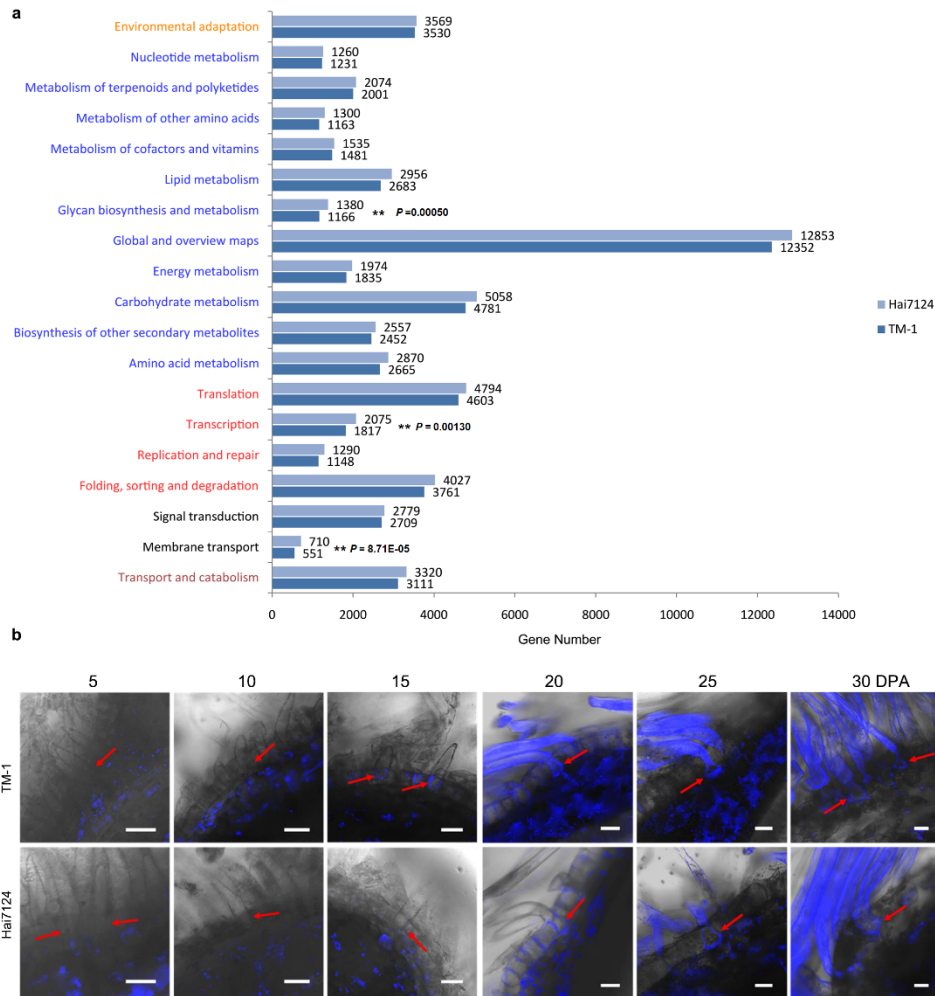
(a) Distribution of LTR across the gene body and its flanking regions (± 3 kb). **(b)** Distribution of Copia, Gypsy, and other LTRs across the gene body and flanking regions (± 3 kb). **(c)** Percentage of expressed genes (EGs) of all PCGs and genes with PAVs in different gene parts in root, stem, leaf, fiber at 10 DPA, and fiber at 20 DPA. **(d)** Percentage of differentially expressed genes (DEGs) between Hai7124 and TM-1 of all PCGs and genes with PAVs in different gene parts in root, stem, leaf, fiber at 10 DPA, and fiber at 20 DPA.



Supplementary Fig. 20

TE insertion in a CESA gene pair.

(a) Phylogenetic three of CESA genes related to SCW development, including CESA 4, 7 and 8 clades. Red dashed box indicates the homoeologous gene in TM-1 and Hai7124. **(b)** TE insertion in *GH_A07G0437* from all 10 tested *G.hirsutum* and Pima S-4 compared to the normal gene *GB_A07G0431* from *G. barbadense* (**Supplementary Table 29**). Blue line indicated the genomic sequence and the yellow box indicated the ORF. **(c)** The gene expression in different *G. hirsutum* (blue horizon line) and *G. barbadense* (yellow horizon line) species.

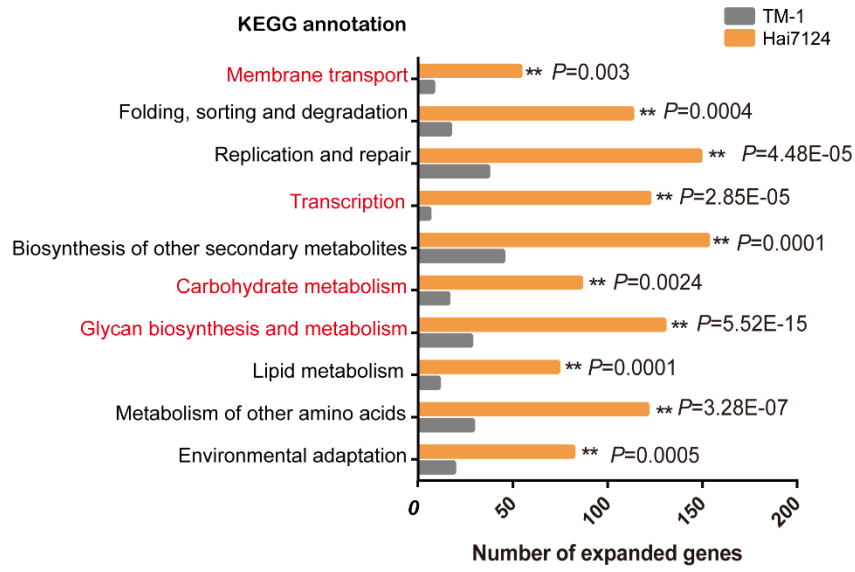


Supplementary Figure 21

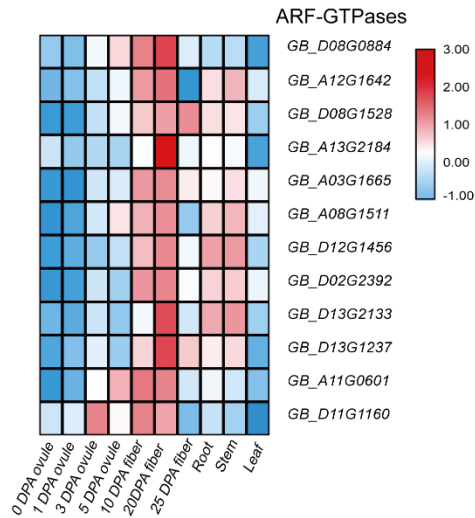
The molecular mechanism of fiber development between TM-1 and Hai7124.

(a) Comparison of KEGG annotation between TM-1 and Hai7124. The pathways in which the genes might be involved were assigned by BLAST against the KEGG databases (release 59.3), with an E-value cutoff of $1e-05$. The number of genes in each pathway were listed beside the bar. **, $P < 0.01$ with Fisher's exact test. **(b)** Aniline blue staining for callose at the fiber of TM-1 and Hai7124 plants. No aniline blue fluorescent signals for callose were detected at the 5-10 DPA fiber bases in TM-1 and 5-15 DPA fiber bases in Hai7124. By contrast, evident fluorescent signal of callose were observed at the 15-DPA fiber bases in TM-1, but at 20-DPA fiber bases in Hai7124. In addition, the whole fiber exhibited the extensive fluorescent signal of callose in 20-30 DPA of TM-1 and 25-30 DPA of Hai7124. For each stage, the imaging analysis was performed from three individual plants with a total of at least 15 optical sections. Scale bar = $20\mu\text{m}$.

a



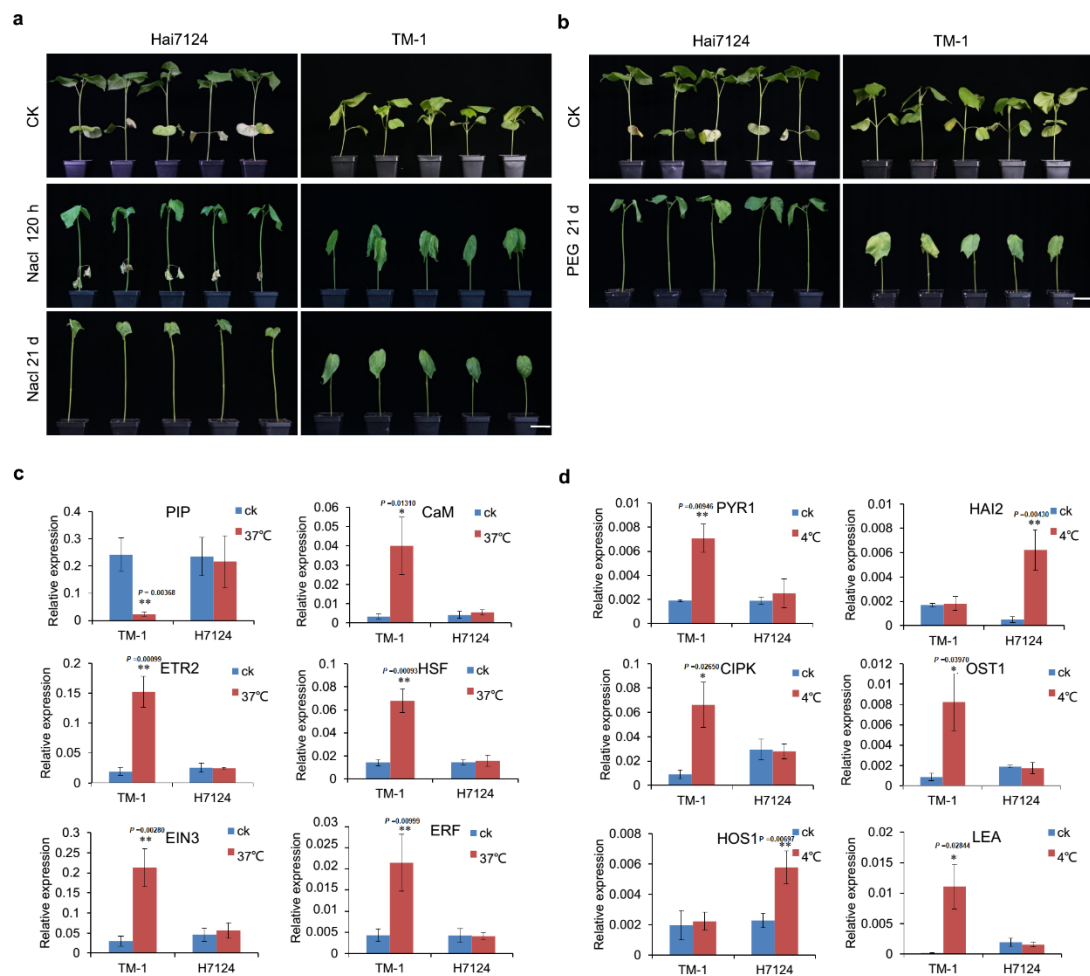
b



Supplementary Figure 22

Comparison of the expanded gene families in TM-1 and Hai7124.

(a) Comparison of KEGG annotation of expanded gene families in TM-1 and Hai7124). (b) The expression profile of *ARF-GTPases* genes which specifically expanded in Hai7124. ** P<0.01, Fisher's exact tests.



Supplementary Figure 23

Comparison of abiotic tolerance between TM-1 and Hai7124.

(a, b) Comparison of salt and drought tolerance between TM-1 and Hai7124. Photographs were taken under normal conditions and subsequent to treatment with NaCl and PEG for 120 hours or 21 days. Scale bar = 10 cm. (c) qRT-PCR analysis of *PIP*, *CaM*, *HSF*, *ETR2*, *EIN3* and *ERF* in the leaves of TM-1 and Hai7124 at 1 h after heat stress. Three biological replicates were performed per reaction, each with two technical replicates (using the same sample). Each value represents mean \pm SE (* $P < 0.05$, ** $P < 0.01$, Student's t-test). (d) qRT-PCR analysis of *PYR1*, *HAI2*, *CIPK*, *OST1*, *HOS1* and *LEA* in the leaves of TM-1 and Hai7124 at 24 h after cold stress. Three biological replicates were performed per reaction, each with two technical replicates (using the same sample). Each value represents mean \pm SE (* $P < 0.05$, ** $P < 0.01$, Student's t-test).

Supplementary Tables

Supplementary Table 1 Statistic of DNA-seq data from the Illumina technology

Species	Type of sequencing libraries	Insert Size(bp)	Read Length(bp)	Total Data(Gb)	Sequence depth (×)
<i>G. hirsutum</i> acc. TM-1	PCR-free library	470bp	265	174.25	73.78
		800bp	160	108.61	45.99
		2-4kb	150	139.29	58.98
	Mate Pair library	5-7kb	150	127.35	53.92
		8-10kb	150	119.17	50.46
	10× GemCode library		150	126.64	53.62
	Total			795.31	336.75
<i>G. barbadense</i> cv. Hai7124	PCR-free library	470bp	265	193.11	86.7
		800bp	160	139.91	62.81
		2-4kb	150	132.81	59.63
	Mate Pair library	5-7kb	150	111.43	50.03
		8-10kb	150	102.96	46.22
	10× GemCode library		150	120.65	54.17
	Total			800.87	359.56

Supplementary Table 2 The initial assembly of Hai7124 by software DenovoMAGIC

Hai7124	Contig		Scaffold	
	Size (bp)	Number	Size (bp)	Number
N90	14,387	32,755	2,133,805	236
N80	26,930	21,604	4,039,212	162
N70	40,755	14,942	5,939,532	118
N60	56,821	10,349	8,226,566	85
N50	76,802	6,990	10,811,328	61
Longest	1,536,444	-	36,820,227	-
Total Size	2,211,778,839	-	2,227,353,779	-
Number ($\geq 1\text{kb}$)	-	71,245	-	11,701
Number ($\geq 10\text{kb}$)	-	38,742	-	1,026
Number ($\geq 100\text{kb}$)	-	4,775	-	451
Number ($\geq 1\text{Mb}$)	-	6	-	301

Supplementary Table 3 Optical maps of TM-1 and Hai7124

Bionano data information						
Sample Name	Enzyme	Molecules Number	Total Length (Mb)	Molecule N50 (kb)	Average length (kb)	Label Density (/100kb)
TM-1	<i>BssSI</i>	1,796,868	457,130.03	268.91	254.40	13.91
Hai7124	<i>BssSI</i>	1,625,639	449,305.79	298.68	276.39	12.52
Optical maps information						
	Total Genome Map Length (Mb)	Avg. Genome Map Length (Mb)	Median Genome Map Length (Mb)	Genome Map N50(Mb)		
TM-1	2,521.22	1.07	0.45	2.33		
Hai7124	2,425.66	1.54	0.31	4.33		
Hybrid genome assembly						
	Scaffold Length(bp)	Scaffold Number	Max Scaffold Length(bp)	GC(%)		
TM-1	2,374,491,760	49,347	41,918,329	35.42		
Hai7124	2,225,652,196	11,425	101,653,628	34.29		

Supplementary Table 4 Summary of Hi-C data

	TM-1		Hai7124	
	Numbers	Percentage (%)	Numbers	Percentage (%)
Raw reads	1,825,615,872		1,860,974,502	
Filter unmapped reads	8,739,607	0.5	6,219,280	0.3
Filter PCR duplications	291,126,039	15.9	307,129,655	16.5
Filter low mapping quality reads (MQ < 10)	422,117,541	23.1	474,128,683	25.5
Filter reads outside 500bp of <i>HindIII</i> restriction site	23,296,206	1.3	31,486,706	1.7
Filter multiple-aligned reads	213,737,543	11.7	228,227,448	12.3
Valid reads	866,598,936	47.5	813,782,730	43.7
Pairs on same scaffold	334,180,244	77.1	330,403,741	81.2
Pairs on different scaffolds	99,119,224	22.9	76,487,624	18.8

Supplementary Table 5 Characteristics of the 26 linkage groups in allotetraploid cotton

Linkage group	No. bins	No. scaffolds	Size (Mb)	No. SNPs
A01	241	106	118.0	193,150
A02	248	163	108.9	338,582
A03	318	182	112.3	303,510
A04	224	158	88.3	264,400
A05	280	150	109.8	257,844
A06	316	236	126.9	441,942
A07	328	153	96.6	308,645
A08	271	186	125.5	380,384
A09	260	129	83.8	216,941
A10	339	180	116.1	353,458
A11	362	168	122.5	350,890
A12	333	222	108.1	342,139
A13	330	163	111.7	354,048
A subgenome	3,850	2196	1428.5	4,105,933
D01	263	113	65.5	167,720
D02	257	113	71.1	202,096
D03	187	94	54.0	157,617
D04	193	86	58.2	131,395
D05	225	102	65.6	127,298
D06	271	135	66.0	185,826
D07	230	107	59.4	150,273
D08	247	101	69.8	184,390
D09	229	94	53.1	124,256
D10	287	132	67.9	164,743
D11	287	118	72.2	162,722
D12	235	111	62.5	149,639
D13	188	90	64.7	151,149
D subgenome	3,099	1396	829.9	2,059,124
Total	6,949	3592	2258.4	6,165,057

*Genetic distance was calculated by MSTMap and TM-1 as the reference.

Supplementary Table 6 Possibly mis-assembled scaffolds in Hai7124 detected by BioNano optical, Hi-C and linkage maps

	No. of mis-assembled scaffolds	Size of mis-assembled scaffolds (Mb)	No. of mis-assembled locus	note
Scaffolds assembled by DenovoMAGIC3	54	582.2	63	checked by BioNano optical map
Scaffolds integrated with BioNano optical maps	23	819.3	52	checked by Hi-C map and linkage map

Supplementary Table 7 The final assembly of Hai7124 (V1.1)

Hai7124	Contig		Scaffold		Chromosome	
	Size (bp)	Number	Size (bp)	Number	Size (bp)	Number
N90	15,130	31,664	6,114,884	100	57,348,712	23
N80	27,765	21,106	9,671,034	71	62,791,478	20
N70	41,555	14,667	14,005,833	52	67,735,652	16
N60	57,558	10,190	19,339,146	39	79,226,513	13
N50	77,663	6,902	23,438,167	29	101,615,779	10
Longest	1,536,444	-	101,653,628	-	120,787,428	-
Total Size	2,192,522,227	-	2,224,979,100	-	2,226,679,100	-
Gap Size	0	-	32,456,873	-	34,156,873	-
Non-Gap Size	2,192,522,227	-	2,192,522,227	-	2,192,522,227	-

Supplementary Table 8 The initial assembly of TM-1 by DenovoMAGIC

TM-1	contig		Scaffold	
	Size (bp)	Number	Size (bp)	Number
N90	21,292	23,250	2,142,091	249
N80	40,162	15,406	4,111,221	174
N70	59,856	10,639	6,073,104	127
N60	83,435	7,313	7,625,263	92
N50	112,494	4,889	10,940,307	66
Longest	3,852,169	-	43,045,293	-
Total Size	2,344,123,929	-	2,361,663,414	-
Number (>=1kb)	-	57,291	-	16,448
Number (>=10kb)	-	31,492	-	1,289
Number (>=100kb)	-	5,799	-	470
Number (>=1Mb)	-	46	-	313

Supplementary Table 9 Possibly mis-assembled scaffolds in TM-1 detected by BioNano optical, Hi-C and linkage maps

	No. mis-assembled scaffolds	Size of mis-assembled scaffolds (Mb)	No. mis-assembled locus	note
Scaffolds assembled by DenovoMAGIC3	139	1163.2	206	checked by BioNano optical maps
Scaffolds integrated with BioNano optical maps	0	0	0	checked by Hi-C maps and linkage map

Supplementary Table 10 The final assembly of TM-1(V2.1)

TM-1	Contig		Scaffold		Chromosome	
	Size (bp)	Number	Size (bp)	Number	Size (bp)	Number
N90	24,062	21,703	3,881,943	158	58,417,686	23
N80	42,459	14,714	6,930,448	113	64,447,585	20
N70	61,713	10,293	9,515,247	85	69,080,421	16
N60	84,632	7,152	12,479,724	64	83,216,487	13
N50	113,327	4,831	15,510,801	48	107,588,319	10
Longest	2,103,187	-	41,918,329	-	126,488,190	-
Total	2,267,899,0	-	2,295,257,0	-	2,298,437,01	-
Size	98	-	19	-	9	-
Gap Size	0	-	27,357,921	-	30,537,921	-
Non-	2,267,899,0	-	2,267,899,0	-	2,267,899,09	-
Gap Size	98	-	98	-	8	-

Supplementary Table 11 Assessment of the TM-1 genome assembly (V2.1) using whole sequence of 36 BACs from *G. hirsutum*.

Type	BAC	Chr.	BAC length (bp)	Number of 1kb-mers	Number of mapped 1kb-mers	Number of 1kb-mers mapped to corresponding chromosome	Percentage of mapped 1kb-mers (%)	Percentage of 1kb-mers mapped to corresponding chromosome (%)
TM-1	HQ650107.1	A12	110,706	109,707	109,707	107,466	100	97.96
TM-1	HQ650105.1	A12	35,934	34,935	34,935	33,641	100	96.3
TM-1	HQ650108.1	D12	60,529	59,530	59,530	59,530	100	100
TM-1	HQ650106.1	D12	121,433	120,434	120,434	117,808	100	97.82
Maxxa	AC243164.1	D01	113,782	112,783	112,783	110,988	100	98.41
Maxxa	AC243162.1	A06	98,696	97,697	97,697	95,767	100	98.02
Maxxa	AC243160.1	A01	113,785	112,786	112,786	107,848	100	95.62
Maxxa	AC243158.1	D01	118,603	117,604	117,604	113,595	100	96.59
Maxxa	AC243156.1	A12	119,216	118,217	118,217	114,615	100	96.95
Maxxa	AC243154.1	D01	104,636	103,637	103,637	103,144	100	99.52
Maxxa	AC243152.1	D01	104,306	103,307	103,307	100,343	100	97.13
Maxxa	AC243150.1	D07	86,390	85,391	85,391	81,640	100	95.61
Maxxa	AC243148.1	A11	108,382	107,383	107,383	105,967	100	98.68
Maxxa	AC243145.1	D11	134,387	133,388	133,388	127,890	100	95.88
Maxxa	AC243143.1	A12	137,405	136,406	136,406	129,473	100	94.92
Maxxa	AC243141.1	D12	89,174	88,175	88,175	88,175	100	100
Maxxa	AC243139.1	D11	58,716	57,717	57,717	57,441	100	99.52
Maxxa	AC243137.1	A10	137,066	136,067	136,067	134,516	100	98.86
Maxxa	AC243133.1	A01	163,851	162,852	162,852	150,379	100	92.34
Maxxa	AC243131.1	D12	121,823	120,824	120,824	118,381	100	97.98
Maxxa	AC243165.1	A12	142,351	141,352	141,352	135,848	100	96.11
Maxxa	AC243163.1	A12	138,301	137,302	137,302	132,351	100	96.39
Maxxa	AC243161.1	A12	59,488	58,489	58,489	56,356	100	96.35
Maxxa	AC243159.1	D10	58,828	57,829	57,829	54,068	100	93.5
Maxxa	AC243157.1	A01	144,980	143,981	143,981	138,724	100	96.35
Maxxa	AC243155.1	A12	120,301	119,302	119,302	115,530	100	96.84
Maxxa	AC243153.1	D12	152,015	151,016	151,016	150,750	100	99.82
Maxxa	AC243151.1	D01	108,361	107,362	107,362	98,858	100	92.08
Maxxa	AC243149.1	D12	113,692	112,693	112,693	101,731	100	90.27
Maxxa	AC243146.1	D12	141,129	140,130	140,130	136,651	100	97.52
Maxxa	AC243144.1	D02	94,928	93,929	93,929	85,038	100	90.53
Maxxa	AC243142.1	D12	102,430	101,431	101,431	98,841	100	97.45
Maxxa	AC243140.1	D08	110,533	109,534	109,534	107,697	100	98.32
Maxxa	AC243138.1	D02	132,457	131,458	131,458	130,339	100	99.15
Maxxa	AY632359.1	A10	103,930	102,931	102,931	96,710	100	93.96
Maxxa	AY632360.1	D10	135,862	134,863	134,863	127,563	100	94.59
Total	NA	NA	3,998,4	3,962,442	3,962,442	3,825,662	100	96.55

Supplementary Table 12 Centromere mapping by CRGs and GhCRs in Hai7124 (V1.1)

Chromosome	CRGs-5'LTR			GhCRs-5'LTR		
	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)
A01	64.91	64.86-64.98	0.11	64.87	64.84-64.96	0.13
A02	67.77	67.68-67.93	0.24	67.8	67.71-67.93	0.22
A03	38.97	38.85-39.03	0.18	38.98	38.93-39.03	0.1
A04	32.03	31.98-32.14	0.16	32.03	31.93-32.10	0.17
A05	55.78	55.75-55.93	0.18	55.89	55.78-55.98	0.2
A06	53	52.84-53.10	0.26	52.94	52.86-53.09	0.23
A07	59.34	59.27-59.41	0.14	59.33	59.24-59.41	0.17
A08	49.47	49.35-49.63	0.29	49.41	49.34-49.53	0.19
A09	19.16	19.09-19.20	0.11	19.17	19.05-19.22	0.17
A10	46.78	46.68-46.89	0.21	46.78	46.69-46.89	0.2
A11	67.5	67.41-67.57	0.16	67.46	67.38-67.54	0.16
A12	33.05	32.92-33.11	0.19	33	32.93-33.09	0.17
A13	34.9	34.83-34.97	0.14	34.86	34.76-34.91	0.16
D01	34.99	34.92-35.09	0.17	35.09	35.00-35.19	0.19
D02	32.65	32.58-32.70	0.11	32.65	32.59-32.70	0.12
D03	17.34	16.18-24.33	8.16	24.98	23.33-26.63	3.31
D04	28.03	27.96-28.10	0.14	28.14	28.09-28.27	0.19
D05	42.17	41.99-42.29	0.3	42.14	41.98-42.26	0.28
D06	33.72	33.66-33.87	0.2	33.86	33.72-33.96	0.23
D07	40.99	40.93-41.08	0.15	41.05	40.97-41.12	0.15
D08	26.97	26.88-27.05	0.17	26.96	26.89-27.03	0.14
D09	21.69	21.62-21.75	0.14	21.63	21.55-21.70	0.15
D10	38.7	38.62-38.75	0.13	38.7	38.61-38.75	0.14
D11	43.94	43.88-44.05	0.17	43.87	43.79-43.94	0.15
D12	18.53	18.46-18.60	0.14	18.86	18.75-18.99	0.24
D13	28.08	27.99-28.23	0.24	28.08	27.98-28.23	0.26

CIM: Confidence Interval for the Median.

Supplementary Table 13 Centromere mapping by CRGs and GhCRs in TM-1(V2.1)

Chromosome	CRGs-5'LTR			GhCRs-5'LTR		
	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)
A01	66.8	66.74-66.95	0.2	66.71	66.66-66.82	0.16
A02	70.61	70.35-70.77	0.42	70.14	70.08-70.30	0.23
A03	40.42	40.31-40.56	0.25	40.35	40.29-40.48	0.19
A04	34.52	34.41-34.73	0.31	34.49	34.42-34.57	0.15
A05	55.74	55.68-56.33	0.65	56.35	56.30-56.42	0.12
A06	55.93	55.72-56.06	0.34	55.93	55.81-56.02	0.21
A07	60.3	60.18-60.40	0.21	60.38	60.30-60.43	0.13
A08	52.15	52.03-52.20	0.17	52.09	52.04-52.14	0.1
A09	20.09	19.89-20.37	0.48	19.96	19.92-20.07	0.15
A10	47.74	47.71-48.01	0.3	47.99	47.86-48.06	0.2
A11	71.39	71.33-71.54	0.2	71.45	71.36-71.53	0.17
A12	35.09	35.01-35.31	0.3	35	34.92-35.05	0.13
A13	34.81	34.72-34.94	0.22	34.7	34.61-34.79	0.17
D01	35.9	35.84-36.02	0.18	35.44	35.35-35.55	0.2
D02	33.44	33.38-33.50	0.12	33.51	33.45-33.60	0.15
D03	27.94	25.61-28.38	2.77	25.37	22.30-26.82	4.53
D04	27.01	26.94-27.17	0.23	27.02	26.95-27.10	0.15
D05	41.24	41.17-41.50	0.32	41.55	41.37-41.65	0.28
D06	35.35	35.16-35.60	0.43	35.35	35.25-35.41	0.16
D07	41.12	41.01-41.36	0.35	41.25	41.12-41.36	0.24
D08	26.97	26.82-27.10	0.28	27.03	26.95-27.13	0.18
D09	20.18	20.05-20.27	0.22	20.22	20.18-20.28	0.1
D10	38.82	38.76-38.93	0.17	39.08	38.94-39.18	0.23
D11	43.81	43.65-43.87	0.22	43.83	43.74-43.92	0.18
D12	19.29	19.13-19.33	0.2	19.22	19.16-19.31	0.15
D13	29.97	29.79-30.19	0.4	30.15	30.00-30.24	0.24

CIM: Confidence Interval for the Median.

Supplementary Table 14 Centromere mapping by CRGs and GhCRs in TM-1(V1.1)⁴

Chromosome	CRGs-5'LTR			GhCRs-5'LTR		
	Median (Mb)	95 % CIM (Mb)	Size of 95 % CIM (Mb)	Median (Mb)	95 % CIM (Mb)	Size of 95 % CIM (Mb)
A01	59.82	47.13-72.33	25.2	-	-	-
A02	60.17	40.25-74.48	34.23	-	-	-
A03	34.84	30.47-36.23	5.76	34.14	30.47-35.31	4.84
A04	13.93	7.53-21.80	14.28	22.5	13.93-23.57	9.64
A05	25.86	23.76-45.62	21.86	42.2	25.86-49.06	23.2
A06	56.66	56.13-62.13	6	56.66	47.92-62.13	14.21
A07	25.74	16.34-39.06	22.73	-	-	-
A08	-	-	-	36.58	32.11-49.30	17.19
A09	28.31	15.86-55.04	39.18	28.31	24.78-47.31	22.54
A10	51.09	35.14-58.08	22.94	51.09	40.26-61.61	21.34
A11	44.24	20.55-54.71	34.16	-	-	-
A12	37.71	32.52-48.47	15.95	38.01	32.52-45.67	13.15
A13	40.73	25.11-59.06	33.95	-	-	-
D01	33.36	33.10-33.62	0.51	33.39	33.28-33.61	0.34
D02	31.66	31.26-31.99	0.74	32.09	31.75-32.70	0.95
D03	21.14	21.10-21.19	0.09	21.14	21.09-21.19	0.1
D04	23.6	23.16-24.20	1.04	24.7	23.61-27.67	4.06
D05	41.03	40.81-41.40	0.59	41.08	40.81-41.43	0.62
D06	34.56	34.35-34.71	0.35	34.52	34.36-34.62	0.26
D07	39.37	39.16-39.56	0.4	39.39	39.36-39.58	0.22
D08	26.1	25.97-26.53	0.56	26.1	26.08-26.47	0.39
D09	19.86	19.77-19.98	0.2	19.95	19.76-20.30	0.54
D10	37.58	36.86-37.88	1.02	38.31	36.79-40.62	3.83
D11	40.42	40.36-41.16	0.8	40.41	40.35-40.45	0.1
D12	23.76	23.56-24.49	0.93	23.57	23.42-23.76	0.34
D13	25.72	24.60-26.56	1.95	25.63	24.61-25.73	1.13

CIM: Confidence Interval for the Median.

Supplementary Table 15 Centromere mapping by CRGs and GhCRs in Xinhai21(ref. ⁵)

Chromosome	CRGs-5'LTR			GhCRs-5'LTR		
	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)
A01	21.28	19.24-51.49	32.25	28.96	19.85-51.49	31.65
A02	48.99	48.70-52.28	3.58	49.75	48.99-57.95	8.95
A03	36.26	33.72-37.67	3.95	35.08	30.96-36.34	5.38
A04	23.49	22.40-37.58	15.18	23.3	23.24-23.52	0.28
A05	50.38	50.03-62.06	12.03	50.26	50.05-51.46	1.41
A06	42.8	42.79-64.02	21.23	46.19	42.79-65.62	22.84
A07	49.97	40.53-54.05	13.52	54.6	49.99-54.82	4.83
A08	56.84	51.02-57.53	6.51	54.61	50.13-57.23	7.09
A09	18.6	13.67-35.04	21.38	29.86	18.92-31.95	13.04
A10	49.29	39.07-69.05	29.98	48.24	39.07-60.01	20.94
A11	73.5	73.49-76.38	2.89	71.76	68.33-73.50	5.17
A12	60.18	51.41-69.92	18.51	38.39	34.13-51.41	17.28
A13	35.96	35.12-39.84	4.72	39.33	38.64-41.76	3.12
D01	35.79	35.45-36.09	0.64	35.99	35.74-36.14	0.4
D02	30.09	29.94-30.16	0.22	30.09	29.98-30.16	0.18
D03	24.6	22.82-26.47	3.65	25.44	24.32-26.47	2.15
D04	21.26	20.50-21.48	0.98	21.26	20.52-21.58	1.06
D05	39.97	39.01-40.65	1.64	39.65	38.52-40.56	2.04
D06	33.22	32.91-33.37	0.46	33.08	32.82-33.25	0.42
D07	39.99	39.93-40.02	0.09	39.99	39.93-40.04	0.1
D08	30.66	30.55-30.73	0.18	30.69	30.65-30.84	0.19
D09	18.75	18.58-18.80	0.23	18.56	18.45-18.69	0.23
D10	35.77	35.60-36.57	0.97	35.6	35.05-35.77	0.72
D11	37.59	37.38-37.62	0.25	37.37	37.32-37.58	0.27
D12	22.71	21.83-23.08	1.26	22.11	21.83-22.76	0.93
D13	26.13	24.07-27.91	3.84	26.13	24.83-27.56	2.73

CIM: Confidence Interval for the Median.

Supplementary Table 16 Centromere mapping by CRGs and GhCRs in 3-79 (ref.⁷)

Chromosome	CRGs-5'LTR			GhCRs-5'LTR		
	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)	Median (Mb)	95%CIM (Mb)	Size of 95%CIM (Mb)
A01	19.97	19.97-67.84	47.87	44.31	26.87-56.64	29.77
A02	-	-	-	-	-	-
A03	35.6	23.61-35.74	12.13	35.59	23.61-35.74	12.13
A04	27.74	21.73-28.49	6.76	22.79	14.25-28.49	14.25
A05	53.1	36.67-67.57	30.9	53.1	45.17-63.12	17.95
A06	43.5	15.90-52.77	36.87	43.5	31.00-57.13	26.13
A07	30.6	14.21-50.11	35.9	50.11	26.86-75.20	48.34
A08	75.55	49.64-91.66	42.02	49.81	47.74-75.56	27.81
A09	16.18	15.03-16.18	1.15	-	-	-
A10	64.09	63.57-70.25	6.69	56.21	50.45-64.09	13.64
A11	63.87	13.74-69.08	55.35	66.48	46.33-76.74	30.41
A12	50.76	50.68-50.77	0.09	47.39	41.10-51.10	9.99
A13	30.95	26.91-43.80	16.88	41.46	34.84-55.06	20.22
D01	36.42	35.43-37.03	1.6	35.34	35.15-35.70	0.55
D02	31.12	30.94-32.30	1.36	31.62	30.98-34.44	3.46
D03	20.51	20.50-21.43	0.93	20.93	20.00-21.43	1.43
D04	21.75	17.99-22.38	4.39	22.4	21.77-24.28	2.51
D05	44.79	43.71-45.44	1.73	45.43	43.74-45.60	1.85
D06	34.13	33.99-34.30	0.31	34.16	34.04-34.92	0.88
D07	43.43	43.40-45.16	1.76	43.44	43.42-43.48	0.07
D08	28.79	28.35-41.22	12.87	28.65	28.50-40.93	12.43
D09	20.39	20.05-20.45	0.41	20.2	20.11-20.42	0.31
D10	37.86	36.67-38.95	2.27	37.95	37.52-38.98	1.46
D11	43.94	41.73-46.18	4.45	44.05	42.19-44.66	2.48
D12	26.45	25.78-29.08	3.29	22.54	21.70-26.18	4.48
D13	30.1	29.11-31.54	2.43	29.27	28.81-30.10	1.29

CIM: Confidence Interval for the Median.

Supplementary Table 17 Characterization of CenH3-binding domains in TM-1(V2.1) and Hai7124(V1.1)

TM-1 V2.1	CenH3-binding domains (TM-1)			Hai7124 V1.1	CenH3-binding domains (Hai7124)		
	Start (Mb)	End (Mb)	Length (Mb)		Start (Mb)	End (Mb)	Length (Mb)
A01	66.02	67.53	1.51	A01	64.06	65.89	1.83
A02	69.28	70.86	1.58	A02	67.16	68.38	1.22
A03	39.86	41.19	1.33	A03	38.07	39.88	1.81
A04	33.98	35.54	1.56	A04	31.12	33.00	1.88
A05	55.42	56.71	1.29	A05	54.98	56.49	1.51
A06	55.02	57.09	2.07	A06	52.22	53.86	1.64
A07	59.52	60.67	1.15	A07	58.48	60.26	1.78
A08	51.31	52.53	1.22	A08	48.74	50.39	1.65
A09	19.01	20.24	1.23	A09	18.26	19.92	1.66
A10	47.21	48.46	1.25	A10	46.06	47.40	1.34
A11	71.38	72.85	1.47	A11	66.88	68.38	1.50
A12	34.26	35.33	1.07	A12	31.75	33.88	2.13
A13	33.53	34.90	1.37	A13	33.81	35.82	2.01
D01	34.04	35.06	1.02	D01	33.75	35.72	1.97
D02	-	-	-	D02	31.15	34.00	2.85
D03	13.35	14.90	1.55	D03	13.55	15.49	1.94
D04	25.58	26.52	0.94	D04	26.71	28.62	1.91
D05	41.44	42.49	1.05	D05	42.07	43.76	1.69
D06	34.12	35.69	1.57	D06	32.81	34.25	1.44
D07	41.13	41.99	0.86	D07	40.15	42.18	2.03
D08	-	-	-	D08	25.83	28.38	2.55
D09	20.21	21.03	0.82	D09	20.48	23.02	2.54
D10	38.47	39.09	0.62	D10	37.82	40.11	2.29
D11	43.61	44.55	0.94	D11	44.17	45.93	1.76
D12	18.04	18.81	0.77	D12	17.29	19.40	2.11
D13	30.13	31.23	1.10	D13	27.43	28.99	1.56

Supplementary Table 18 General statistics of predicted protein-coding genes in the TM-1 genome assembly (V2.1).

	Gene set	Number	Average gene length (bp)	Average mRNA length (bp)	Average exons per gene	Average exon length (bp)	Average intron length (bp)
De novo	Augustus	90,590	2,694	1,503	5	288	283
	Genescan	112,194	11,844	1,301	5	260	2,633
	GlimmerHMM	138,730	1,678	962	3	316	350
	SNAP	149,052	1,103	706	3	275	253
	GeneID	158,488	4,383	865	4	224	1,230
Homo	<i>A. thaliana</i>	69,890	2,496	1,173	4	276	408
	<i>B. papaya</i>	89,543	2,065	963	3	281	455
	<i>T. cacao</i>	110,952	2,547	1,087	4	305	570
	<i>G. arboreum</i>	140,728	2,899	1,040	4	286	707
	<i>G. raimondii</i>	119,063	2,855	955	4	262	717
	<i>H. syriacus</i>	127,692	1,318	623	3	241	437
	<i>V. vinifera</i>	69,122	2,967	1,162	5	252	499
	RNA-seq	55,702	4,558	2,109	6	358	500
	Pacbio transcriptome	71,304	7,180	2,531	8	332	702
	TM-1(V2.1)	72,761	2,970	1,183	5	242	460
	A subgenome	36,063	3,018	1,182	5	242	473
	D subgenome	36,319	2,943	1,190	5	242	447
	<i>G. arboreum</i> ¹⁰	41,330	2,533	1,083	5	236	368
	<i>G. raimondii</i> ⁸	37,505	3,177	1,199	5	237	341
	TM-1(V1.1) ⁴	70,478	3,126	1,179	5	236	486

Notes: Data from corresponding genomic articles.

Supplementary Table 19 General statistics of predicted protein-coding genes in the Hai7124 genome assembly (V1.1)

Gene set	Number	Average gene length (bp)	Average mRNA length (bp)	Average exons per gene	Average exon length (bp)	Average intron length (bp)
Augustus	83,071	2,361	1,143	5	227	301
Genescan	92,187	13,409	1,097	5	201	2,760
De novo GlimmerHMM	90,759	2,041	957	4	235	353
SNAP	107,742	1,187	647	3	208	256
GeneID	142,027	4,652	729	4	183	1,313
<i>A. thaliana</i>	60,032	2,719	1,215	5	254	398
<i>C. papaya</i>	78,457	2,219	986	4	262	447
<i>T. cacao</i>	108,262	2,666	1,090	4	294	582
Homo <i>G. arboreum</i>	130,951	2,990	1,042	4	271	684
<i>G. raimondii</i>	108,706	2,984	964	4	247	697
<i>G. hirsutum</i>	110,532	2,610	1,043	4	266	538
<i>H. syriacus</i>	120,110	1,351	628	3	234	429
<i>V. vinifera</i>	59,407	3,295	1,207	5	232	498
RNA-seq	55,873	4,903	2,194	6	362	535
Pacbio transcriptome	72,729	6,818	2,614	8	340	629
<i>G. barbadense</i> (V1.1)	75,071	3,034	1,178	5	238	470
A subgenome	37,292	3,068	1,174	5	237	478
D subgenome	37,232	3,026	1,187	5	238	461
<i>G. arboreum</i> ¹⁰	41,330	2,533	1,083	5	236	368
<i>G. raimondii</i> ⁸	37,505	3,177	1,199	5	237	341
<i>G. hirsutum</i> ⁴	70,478	3,126	1,179	5	236	486
<i>G. barbadense</i> ⁷	80,876	3,253	1,484	5	290	429

Notes: *Data from corresponding genomic articles.

Supplementary Table 20 Statistic of the manually checked 100 genes

Cotton	GeneID	Number of annotated exons	Number of annotated introns	Number of exons supported by transcriptomic data	Number of splice junctions (IGV)	Integrity ratio of gene
				(IGV)		
	<i>GH_A01G0448</i>	24	23	24	23	100.00%
	<i>GH_A01G1967</i>	19	18	19	18	100.00%
	<i>GH_A01G2504</i>	10	9	10	9	100.00%
	<i>GH_A02G0011</i>	6	5	6	5	100.00%
	<i>GH_A02G0716</i>	8	7	8	7	100.00%
	<i>GH_A02G1554</i>	4	3	4	3	100.00%
	<i>GH_A03G1190</i>	6	5	6	5	100.00%
	<i>GH_A03G1510</i>	15	14	15	14	100.00%
	<i>GH_A04G0121</i>	7	6	7	6	100.00%
	<i>GH_A04G0847</i>	6	5	6	5	100.00%
	<i>GH_A05G3936</i>	19	18	19	18	100.00%
	<i>GH_A06G2387</i>	6	5	6	5	100.00%
	<i>GH_A07G0318</i>	5	4	5	4	100.00%
	<i>GH_A07G0332</i>	4	3	4	3	100.00%
	<i>GH_A07G0861</i>	13	12	13	12	100.00%
	<i>GH_A08G0458</i>	4	3	10	9	40.00%
	<i>GH_A08G2285</i>	4	3	4	3	100.00%
	<i>GH_A08G2302</i>	7	6	7	6	100.00%
	<i>GH_A09G2602</i>	7	6	7	6	100.00%
TM-1	<i>GH_A10G0206</i>	2	1	2	1	100.00%
	<i>GH_A10G0220</i>	14	13	14	13	100.00%
	<i>GH_A11G0185</i>	8	7	8	7	100.00%
	<i>GH_A11G2963</i>	9	8	9	8	100.00%
	<i>GH_A12G0016</i>	12	11	12	11	100.00%
	<i>GH_A12G0054</i>	5	4	5	4	100.00%
	<i>GH_A13G0015</i>	8	7	8	7	100.00%
	<i>GH_A13G0136</i>	11	10	11	10	100.00%
	<i>GH_A13G1741</i>	9	8	9	8	100.00%
	<i>GH_D01G0010</i>	10	9	10	9	100.00%
	<i>GH_D01G1582</i>	18	17	18	17	100.00%
	<i>GH_D02G2312</i>	4	3	4	3	100.00%
	<i>GH_D03G0045</i>	7	6	7	6	100.00%
	<i>GH_D04G0064</i>	9	8	9	8	100.00%
	<i>GH_D04G0217</i>	22	21	22	21	100.00%
	<i>GH_D05G1147</i>	9	8	9	8	100.00%
	<i>GH_D05G1157</i>	10	9	12	11	83.33%
	<i>GH_D05G1907</i>	6	5	6	5	100.00%
	<i>GH_D06G0011</i>	2	1	2	1	100.00%
	<i>GH_D07G0019</i>	13	12	13	12	100.00%

<i>GH_D07G2305</i>	11	10	11	10	100.00%
<i>GH_D08G0049</i>	9	8	9	8	100.00%
<i>GH_D08G2063</i>	3	2	3	2	100.00%
<i>GH_D09G0032</i>	4	3	4	3	100.00%
<i>GH_D09G2598</i>	6	5	6	5	100.00%
<i>GH_D10G0144</i>	11	10	11	10	100.00%
<i>GH_D11G0291</i>	13	12	13	12	100.00%
<i>GH_D12G0006</i>	14	13	14	13	100.00%
<i>GH_D12G2213</i>	9	8	9	8	100.00%
<i>GH_D13G0096</i>	10	9	11	10	90.91%
<i>GH_D13G2601</i>	4	3	4	3	100.00%
<i>GB_A01G0057</i>	5	4	5	4	100.00%
<i>GB_A01G2610</i>	6	5	6	5	100.00%
<i>GB_A02G1730</i>	5	4	5	4	100.00%
<i>GB_A03G0068</i>	7	6	7	6	100.00%
<i>GB_A03G1763</i>	4	3	4	3	100.00%
<i>GB_A03G1858</i>	19	18	19	18	100.00%
<i>GB_A04G1652</i>	4	3	4	3	100.00%
<i>GB_A05G0063</i>	3	2	3	2	100.00%
<i>GB_A05G0695</i>	3	2	3	2	100.00%
<i>GB_A06G0025</i>	1	0	2	1	50.00%
<i>GB_A06G0106</i>	3	2	3	2	100.00%
<i>GB_A06G1761</i>	6	5	6	5	100.00%
<i>GB_A07G0084</i>	3	2	3	2	100.00%
<i>GB_A07G2586</i>	3	2	3	2	100.00%
<i>GB_A07G2618</i>	6	5	6	5	100.00%
<i>GB_A08G0069</i>	9	8	9	8	100.00%
Hai7124 <i>GB_A08G0418</i>	10	9	10	9	100.00%
<i>GB_A08G2417</i>	7	6	7	6	100.00%
<i>GB_A09G0494</i>	5	4	5	4	100.00%
<i>GB_A10G0153</i>	16	15	16	15	100.00%
<i>GB_A10G2158</i>	3	2	3	2	100.00%
<i>GB_A11G0182</i>	4	3	4	3	100.00%
<i>GB_A12G0254</i>	12	11	12	11	100.00%
<i>GB_A13G0701</i>	5	4	5	4	100.00%
<i>GB_A13G2022</i>	14	13	14	13	100.00%
<i>GB_D01G0033</i>	6	5	6	5	100.00%
<i>GB_D01G2105</i>	4	3	4	3	100.00%
<i>GB_D01G2230</i>	2	1	2	1	100.00%
<i>GB_D02G0014</i>	6	5	6	5	100.00%
<i>GB_D02G1780</i>	14	13	14	13	100.00%
<i>GB_D03G0016</i>	2	1	2	1	100.00%
<i>GB_D03G0084</i>	3	2	3	2	100.00%
<i>GB_D04G0126</i>	10	9	10	9	100.00%

<i>GB_D04G0355</i>	3	2	3	2	100.00%
<i>GB_D04G2038</i>	4	3	4	3	100.00%
<i>GB_D05G0121</i>	5	4	5	4	100.00%
<i>GB_D05G1178</i>	5	4	5	4	100.00%
<i>GB_D06G2518</i>	9	8	9	8	100.00%
<i>GB_D07G2484</i>	12	11	12	11	100.00%
<i>GB_D08G0088</i>	3	2	4	3	75.00%
<i>GB_D08G0213</i>	25	24	25	24	100.00%
<i>GB_D08G2292</i>	6	5	6	5	100.00%
<i>GB_D09G0142</i>	10	9	10	9	100.00%
<i>GB_D10G0265</i>	15	14	15	14	100.00%
<i>GB_D11G0020</i>	7	6	7	6	100.00%
<i>GB_D11G3730</i>	5	4	5	4	100.00%
<i>GB_D12G1208</i>	11	10	11	10	100.00%
<i>GB_D12G2459</i>	15	14	15	14	100.00%
<i>GB_D13G2633</i>	7	6	7	6	100.00%
<i>GB_D13G2718</i>	6	5	6	5	100.00%

Note: (Integrity ratio of gene) = (Number of annotated exons) / (Number of exons supported by transcriptomic data).

Supplementary Table 21 Comparison of genome annotations by BUSCO analysis of the two new assemblies and the previously published assemblies of allotetraploid cotton genomes

	<i>G. hirsutum</i>			<i>G. barbadense</i>		
	TM-1(V2.1)	TM-1(V1.1) ⁴	TM-1(CRI) ⁶	Hai724 (V1.1)	Xinhai21 (ref. ⁵)	3-79 (ref. ⁷)
Annotated protein-coding genes	72,761	70,478	76,943	75,071	77,526	80,876
Complete BUSCOs*(%)	97.50	97.43	96.04	97.29	96.04	87.22
Single copy (%)	3.89	4.44	21.53	3.47	16.81	36.94
Duplicated copy (%)	93.61	92.99	74.51	93.82	79.24	50.28
Fragmented (%)	0.35	0.49	0.90	0.42	1.11	0.97
Missing (%)	2.15	2.08	3.06	2.29	2.85	11.81

*BUSCOs analysis included 1440 embryophyta genes, the A-subgenome, D-subgenome and Un-anchored scaffolds were assessed by BUSCO independently, and the duplicated copy (%) was calculated as the ratio of complete genes occurred more than once in the whole genome.

Supplementary Table 22 Summary of the TFs identified in the two allotetraploid assemblies and *G. raimondii*⁸

Family	TM-1(V2.1)				Hai712(V1.1)				<i>G. raimondii</i> ⁸
	A _t	D _t	Unlocated	Total	A _t	D _t	Unlocated	Total	D genome
bHLH	226	230	3	459	231	236	1	468	243
ERF	226	217	0	443	218	226	1	445	228
MYB	218	223	0	441	219	218	2	439	226
C2H2	162	160	1	323	159	164	0	323	162
NAC	150	150	1	301	151	151	4	306	154
WRKY	116	116	0	232	118	117	1	236	120
bZIP	113	116	0	229	119	111	0	230	115
MYB_related	115	114	0	229	114	115	0	229	120
B3	108	113	1	222	110	106	7	223	101
C3H	97	101	0	198	97	102	0	199	103
HD-ZIP	77	79	2	158	75	81	2	158	80
GRAS	77	80	0	157	78	75	0	153	82
LBD	66	65	0	131	67	74	0	141	72
MIKC	59	69	0	128	64	65	0	129	68
G2-like	62	62	0	124	61	65	1	127	65
Dof	59	59	0	118	59	59	0	118	60
Trihelix	54	53	0	107	53	52	0	105	56
GATA	45	44	0	89	44	45	0	89	46
TALE	45	44	0	89	44	42	0	86	45
HSF	41	41	0	82	40	43	0	83	42
ARF	40	41	0	81	40	41	0	81	40
NF-YB	37	35	1	73	41	37	0	78	38
TCP	37	35	1	73	38	36	0	74	37
NF-YC	36	36	0	72	36	36	1	73	38
FAR1	35	33	0	68	34	35	1	70	35
M-type	22	44	0	66	23	44	0	67	64
AP2	32	32	0	64	31	32	1	64	32
SBP	29	29	0	58	29	30	0	59	30
CO-like	28	28	0	56	29	29	0	58	29
ARR-B	27	27	0	54	27	27	0	54	26
GRF	24	25	0	49	24	25	0	49	24
YABBY	24	25	0	49	25	24	0	49	24
ZF-HD	23	24	0	47	23	25	0	48	27
WOX	20	21	0	41	21	20	0	41	20
DBB	21	18	0	39	19	18	0	37	19
CPP	17	17	1	35	16	21	0	37	20
Nin-like	16	19	0	34	18	17	0	35	14
NF-YA	16	17	0	33	16	18	0	34	17

HB-other	15	15	0	30	15	16	0	31	14
SRS	15	15	0	30	15	15	0	30	15
E2F/DP	10	14	0	24	12	11	0	23	11
BES1	12	11	0	22	9	14	0	23	10
S1Fa-like	13	9	0	22	11	11	1	23	15
BBR-BPC	9	8	1	18	11	9	0	20	9
LSD	9	9	0	18	9	10	0	19	13
NZZ/SPL	10	8	0	18	8	11	0	19	11
EIL	8	9	0	17	9	10	0	19	15
RAV	8	9	0	17	10	8	0	18	8
SAP	9	8	0	17	8	9	0	17	11
CAMTA	8	8	0	16	7	9	0	16	10
GeBP	7	8	0	15	8	6	0	14	9
LFY	4	5	0	9	4	4	0	8	4
HB-PHD	4	4	0	8	3	4	0	7	5
VOZ	4	4	0	8	4	3	0	7	4
HRT-like	3	1	1	5	3	2	1	6	2
NF-X1	2	2	0	4	2	2	0	4	2
STAT	1	3	0	4	2	2	0	4	2
Whirly	2	2	0	4	1	2	0	3	2
Total	2,753	2,794	13	5,558	2,762	2,820	24	5,606	2,894

Supplementary Table 23 Comparison of transposable elements between two subgenomes of TM-1(V2.1)

Type	No. TE in TM-1*	Total (Mb)**	TM-1 (%)***	A _t (Mb)	A _t (%)	D _t (Mb)	D _t (%)
Mobile element	2,634,087	1,460.46	63.89	979.05	69.04	443.23	53.85
Class I: Retroelement	2,429,905	1,402.35	61.35	938.40	66.18	429.91	52.23
LTR Retroelement	2,399,786	1,390.29	60.82	929.98	65.58	427.64	51.96
copia	212,839	163.86	7.17	106.57	7.52	54.78	6.66
gypsy	1,402,429	946.42	41.40	709.74	50.05	213.36	25.92
unclassified LTR	784,518	443.24	19.39	224.07	15.80	209.74	25.48
non-LTR retrotransposon	11,550	5.20	0.23	2.59	0.18	1.56	0.19
unclassified retroelement	18,569	14.93	0.65	12.39	0.87	1.95	0.24
Class II: DNA transposon	22,339	17.09	0.75	7.56	0.53	9.41	1.14
DNA transposon superfamily	22,074	17.00	0.74	7.53	0.53	9.36	1.14
CACTA superfamily	5,421	2.79	0.12	1.10	0.08	1.66	0.20
hAT superfamily	602	0.40	0.02	0.22	0.02	0.17	0.02
mutator superfamily	16,035	13.83	0.60	6.22	0.44	7.54	0.92
other	16	0.0045	0.0002	0.0011	0.0001	0.0027	0.0003
unclassified DNA transposon	265	0.0953	0.0042	0.034	0.0024	0.0511	0.0062
Unclassified element	181,843	91.69	4.01	72.77	5.13	12.06	1.46

1. A_t and D_t indicate the A subgenome and D subgenome, respectively.
2. * most repeats fragmented by insertions or deletions have been counted as one element.
3. ** some elements may partly include other element domain.
4. *** The non-gap assembly size 2.29 Gb (A_t: 1.42Gb, D_t: 0.82Gb) was used to calculate the percentage of transposable elements in TM-1.

Supplementary Table 24 Comparison of transposable elements between two subgenomes of Hai7124(V1.1)

Type	No. TE in Hai7124 *	Total (Mb)**	Hai7124 (%)***	A _t (Mb)	A _t (%)	D _t (Mb)	D _t (%)
Mobile element	2,476,366	1,374.61	62.15	922.04	67.50	420.66	52.15
Class I: Retroelement	2,271,929	1,316.84	59.54	881.59	64.54	407.21	50.49
LTR Retroelement	2,239,127	1,304.24	58.97	872.81	63.89	404.86	50.19
copia	197,985	150.64	6.81	97.97	7.17	50.33	6.24
gypsy	1,278,455	874.23	39.53	659.42	48.27	196.73	24.39
unclassified LTR	762,687	435.45	19.69	221.66	16.23	205.85	25.52
non-LTR retotransposon	12,864	5.69	0.26	2.96	0.22	1.61	0.20
unclassified retroelement	19,938	15.09	0.68	12.49	0.91	1.99	0.25
Class II: DNA transposon	22,224	17.13	0.77	7.56	0.55	9.45	1.17
DNA transposon superfamily	21,971	17.05	0.77	7.53	0.55	9.41	1.17
CACTA superfamily	5,268	2.73	0.12	1.08	0.08	1.63	0.20
hAT superfamily	633	0.41	0.02	0.23	0.02	0.17	0.02
mutator superfamily	16,053	13.92	0.63	6.23	0.46	7.62	0.94
other	17	0.0062	0.0003	0.0025	0.0002	0.0031	0.0004
unclassified DNA transposon	253	0.0871	0.0039	0.0397	0.0029	0.0418	0.0052
Unclassified element	182,213	90.80	4.11	73.42	5.37	11.47	1.42

A_t and D_t indicate the A subgenome and D subgenome, respectively.

* most repeats fragmented by insertions or deletions have been counted as one element.

** some elements may partly include other element domain.

*** The non-gap assembly size 2.22 Gb (A_t: 1.37Gb, D_t: 0.81Gb) was used to calculate the percentage of transposable elements in Hai7124.

Supplementary Table 25 Resequencing data of 17 accessions representing 13 D-genome diploid species and five AD-genome allotetraploids.

Sample	Accession	Reads number(M)	Bases number (G)	GC%	Q20 %	Q30 %	Mapped reads(M)	mapping rate%
TM-1	(AD) ₁	62.89	6.16	39.26	98.75	93.09	56.31	89.54
Hai7124	(AD) ₂	70.31	6.41	37.64	98.70	94.29	60.03	85.39
<i>G. tomentosum</i>	(AD) ₃₋₅	67.51	6.60	38.48	98.81	93.69	60.75	89.99
<i>G. mustelinum</i>	(AD) ₄₋₁	25.11	2.46	35.44	98.96	94.45	23.03	91.71
<i>G. darwinii</i>	(AD) ₅	66.20	6.08	36.44	97.45	90.53	58.67	88.62
<i>G. thurberi</i>	D ₁₋₄₁	25.87	2.53	37.41	98.70	93.11	25.15	97.21
<i>G. armourianum</i>	D ₂₋₁₋₇	26.52	2.60	38.07	98.72	93.20	26.05	98.24
<i>G. harknessii</i>	D ₂₋₂	59.40	5.82	40.98	98.98	94.15	57.69	97.12
<i>G. davidsonii</i>	D _{3-d}	58.05	5.65	37.90	98.69	92.99	55.85	96.21
<i>G. klotzschianum</i>	D _{3k-54}	30.08	2.94	37.32	98.69	93.10	29.33	97.51
<i>G. aridum</i>	D ₄₋₁₃	30.26	2.96	37.24	98.69	93.14	29.60	97.83
<i>G. raimondii</i>	D ₅₋₃	24.70	2.41	37.59	98.42	92.06	24.21	97.98
<i>G. raimondii</i>	D ₅₋₄	52.51	5.14	37.09	98.67	93.03	51.36	97.81
<i>G. gossypoides</i>	D ₆₋₇	27.84	2.73	37.84	99.11	94.93	27.24	97.83
<i>G. gossypoides</i>	D ₆₋₈	35.35	3.44	37.19	98.57	92.68	34.60	97.88
<i>G. lobatum</i>	D ₇₋₄	20.00	1.96	35.94	98.80	93.68	19.53	97.65
<i>G. trilobum</i>	D ₈₋₈	43.86	4.28	37.45	98.62	92.87	43.06	98.17
<i>G. trilobum</i>	D ₈₋₉	20.70	2.03	34.66	98.82	93.77	20.33	98.18
<i>G. laxum</i>	D ₉₋₄	34.86	3.41	36.27	98.98	94.35	33.71	96.69
<i>G. laxum</i>	D ₉₋₅	37.06	3.64	37.06	99.03	94.56	36.29	97.91
<i>G. turneri</i>	D ₁₀₋₇	55.99	5.50	37.07	99.18	95.22	54.96	98.16
<i>G. schwendimanii</i>	D ₁₁₋₁	55.09	5.40	38.18	99.07	94.68	53.58	97.25

Supplementary Table 26 Detailed information of the translocations between the A subgenomes of *G. hirsutum* and *G. barbadense* and its corresponding diploid progenitor genomes *G. arboreum* and *G. raimondii*

Species	Chr.	Start (Mb)	End (Mb)	Length (Mb)	Species	Chr.	Start (Mb)	End (Mb)	Length (Mb)	Orientation
<i>G. hirsutum</i>	A01	88.7	97.8	9.1	<i>G. arboreum</i>	Chr02	55.7	69.0	13.3	-
<i>G. hirsutum</i>	A01	98.6	117.3	18.7	<i>G. arboreum</i>	Chr02	79.0	98.6	19.6	+
<i>G. hirsutum</i>	A02	0.1	39.8	39.7	<i>G. arboreum</i>	Chr03	0.0	47.2	47.1	+
<i>G. hirsutum</i>	A02	40.3	50.8	10.5	<i>G. arboreum</i>	Chr01	77.1	87.2	10.1	-
<i>G. hirsutum</i>	A03	0.0	0.8	0.8	<i>G. arboreum</i>	Chr01	111.5	112.2	0.8	-
<i>G. hirsutum</i>	A03	1.6	2.9	1.2	<i>G. arboreum</i>	Chr01	110.3	111.4	1.1	-
<i>G. hirsutum</i>	A03	3.1	4.0	0.9	<i>G. arboreum</i>	Chr01	109.3	110.2	0.9	+
<i>G. hirsutum</i>	A03	4.0	5.1	1.1	<i>G. arboreum</i>	Chr01	108.1	109.1	1.1	+
<i>G. hirsutum</i>	A03	11.5	23.5	12.0	<i>G. arboreum</i>	Chr01	88.8	101.1	12.3	-
<i>G. hirsutum</i>	A04	1.1	1.7	0.6	<i>G. arboreum</i>	Chr05	94.9	95.6	0.7	+
<i>G. hirsutum</i>	A04	8.1	12.9	4.8	<i>G. arboreum</i>	Chr05	82.8	88.2	5.4	-
<i>G. hirsutum</i>	A05	78.8	87.9	9.2	<i>G. arboreum</i>	Chr04	69.2	82.3	13.1	-
<i>G. hirsutum</i>	A05	95.6	110.7	15.1	<i>G. arboreum</i>	Chr04	82.6	98.5	15.9	+
<i>G. hirsutum</i>	D04	34.0	43.5	9.5	<i>G. raimondii</i>	Chr09	57.1	66.5	9.3	+
<i>G. barbadense</i>	A01	86.4	95.4	9.0	<i>G. arboreum</i>	Chr02	55.7	69.0	13.3	-
<i>G. barbadense</i>	A01	96.0	114.8	18.8	<i>G. arboreum</i>	Chr02	79.0	98.6	19.6	+
<i>G. barbadense</i>	A02	0.0	38.6	38.6	<i>G. arboreum</i>	Chr03	0.0	47.2	47.1	+
<i>G. barbadense</i>	A02	40.4	50.9	10.4	<i>G. arboreum</i>	Chr01	77.1	87.2	10.1	-
<i>G. barbadense</i>	A03	0.0	0.8	0.8	<i>G. arboreum</i>	Chr01	111.5	112.2	0.8	-
<i>G. barbadense</i>	A03	1.6	2.7	1.0	<i>G. arboreum</i>	Chr01	110.3	111.4	1.1	-
<i>G. barbadense</i>	A03	2.8	3.7	0.9	<i>G. arboreum</i>	Chr01	109.3	110.2	0.9	+
<i>G. barbadense</i>	A03	3.7	4.8	1.1	<i>G. arboreum</i>	Chr01	108.1	109.1	1.1	+
<i>G. barbadense</i>	A03	11.0	23.0	12.0	<i>G. arboreum</i>	Chr01	88.8	101.1	12.3	-
<i>G. barbadense</i>	A04	1.1	1.8	0.6	<i>G. arboreum</i>	Chr05	94.9	95.6	0.7	+
<i>G. barbadense</i>	A04	4.5	6.9	2.4	<i>G. arboreum</i>	Chr05	89.6	92.4	2.8	-
<i>G. barbadense</i>	A04	8.8	12.2	3.4	<i>G. arboreum</i>	Chr05	82.8	86.7	3.9	-
<i>G. barbadense</i>	A05	74.6	83.4	8.8	<i>G. arboreum</i>	Chr04	69.2	82.2	13.0	-
<i>G. barbadense</i>	A05	93.0	107.5	14.6	<i>G. arboreum</i>	Chr04	82.6	98.5	15.9	+
<i>G. barbadense</i>	D04	34.7	44.0	9.4	<i>G. raimondii</i>	Chr09	57.1	66.5	9.3	+
<i>G. arboreum</i>	Chr01	78.0	87.2	9.2	<i>G. raimondii</i>	Chr03	17.2	21.8	4.6	-
<i>G. arboreum</i>	Chr01	88.8	95.9	7.1	<i>G. raimondii</i>	Chr03	29.5	33.0	3.5	-
<i>G. arboreum</i>	Chr01	96.1	100.4	4.3	<i>G. raimondii</i>	Chr03	33.0	35.7	2.7	+
<i>G. arboreum</i>	Chr01	108.1	109.1	1.1	<i>G. raimondii</i>	Chr03	41.3	42.3	0.9	-
<i>G. arboreum</i>	Chr01	109.3	110.2	0.9	<i>G. raimondii</i>	Chr03	42.3	43.1	0.8	-
<i>G. arboreum</i>	Chr01	110.3	111.4	1.1	<i>G. raimondii</i>	Chr03	43.2	44.2	1.0	+
<i>G. arboreum</i>	Chr01	111.5	112.2	0.8	<i>G. raimondii</i>	Chr03	45.1	45.8	0.6	+
<i>G. arboreum</i>	Chr02	60.0	78.0	18.0	<i>G. raimondii</i>	Chr02	43.7	48.2	4.4	-
<i>G. arboreum</i>	Chr02	83.8	98.6	14.8	<i>G. raimondii</i>	Chr02	52.1	62.1	10.0	+
<i>G. arboreum</i>	Chr04	18.6	34.0	15.4	<i>G. raimondii</i>	Chr09	59.8	66.5	6.7	-

Supplementary Table 27 Peaks of each *Ks* distribution of orthologs in cotton genomes

Orthologs	<i>Ks</i> peak value	Divergence time (MYA)
<i>G. arboreum</i> vs <i>G. raimondii</i>	0.034	6.538
<i>G. barbadense</i> A _t vs <i>G. barbadense</i> D _t	0.032	6.154
<i>G. hirsutum</i> A _t vs <i>G. hirsutum</i> D _t	0.037	7.115
<i>G. hirsutum</i> A _t vs <i>G. barbadense</i> A _t	0.002	0.385
<i>G. hirsutum</i> D _t vs <i>G. barbadense</i> D _t	0.003	0.577
<i>G. barbadense</i> A _t vs <i>G. arboreum</i>	0.004	0.769
<i>G. hirsutum</i> A _t vs <i>G. arboreum</i>	0.005	0.962
<i>G. barbadense</i> D _t vs <i>G. raimondii</i>	0.009	1.731
<i>G. hirsutum</i> D _t vs <i>G. raimondii</i>	0.010	1.923

Note: The formula “ $t = Ks/2r$ ” was used to estimate the divergence time between species, where “*r*” is the neutral substitution rate. A neutral substitution rate of 2.6×10^{-9} was used in the current study.

Supplementary Table 28 Wilcoxon rank sum test of Ka , Ks and Ka/Ks value between datasets

Wilcoxon rank sum test	Ka	Ks	Ka/Ks
TM-1.A _t vs Hai7124.A _t	0.005454	9.059e-05	5.667e-05
TM-1.A _t vs <i>G. arboreum</i>	2.443e-10	1.209e-12	1.562e-11
TM-1. A _t vs TM-1.D _t	0.01676	0.003929	0.01162
TM-1.D _t vs Hai7124.D _t	0.001511	7.002e-05	0.0009119
TM-1.D _t vs <i>G. raimondii</i>	0.08579	0.0005033	3.02e-05
Hai7124. A _t vs <i>G. arboreum</i>	< 2.2e-16	< 2.2e-16	< 2.2e-16
Hai7124.A _t vs Hai7124.D _t	0.0109	3.028e-05	7.972e-06
Hai7124.D _t vs <i>G. raimondii</i>	1.914e-08	< 2.2e-16	< 2.2e-16

Supplementary Table 29 Summary of cotton samples for deep-sequencing

Species	Sample ID	Type	Origin	Reads	GC %	Q30	Mapped reads(M)	Mapping rate%	Accession
				number (M)					
<i>G. barbadense</i>	Tanguis	Landrace	Peru	801	34	93.9	779	97.3	SRR7887412
	Coastland R4-4	Cultivar	America	993	33.5	95	979	98.6	SRR7887420
	Pima S-4	Cultivar	America	1064	33.5	95.5	1048	98.5	SRR7887413
	Ashimouni	Landrace	Egypt	911	34.5	96.3	899	98.7	SRR7887421
	Giza 45	Cultivar	Egypt	956	39.4	94.1	945	98.9	SRR7887423
	Xinhai 14#	Cultivar	China	830	35	95.6	820	98.8	SRR7887411
	Xinhai 25#	Cultivar	China	1053	33.9	95.6	1034	98.2	SRR7887410
	Junhai 1#	Cultivar	China	978	34.6	95.8	964	98.6	SRR7887422
Hai7124	Cultivar	China	1620	36.5	87.2	1589	98.1	SRR8090394	
<i>G. hirsutum</i>	TM-1	Cultivar	America	1356	38.5	90	1308	96.5	SRR7910059
	Yucatanense 6	Race	Mexico	937	34.7	95.5	932	99.4	SRR7887415
	Punctatum 25	Race	Egypt	995	35.4	95.2	988	99.4	SRR7887417
	latifolium 13	Race	Mexico	936	36.4	93.6	930	99.3	SRR7887419
	Stoneville 2B	Cultivar	America	635	34.9	94.6	628	99	SRR5512449
	Deltapine 15	Cultivar	America	872	35.2	95.9	866	99.3	SRR5512448
	UGD mian	Cultivar	China	1072	33.8	96.5	1061	99	SRR5512442
	Simian 2#	Cultivar	China	1078	34.7	96.2	1068	99	SRR5512445
	Junmian 1#	Cultivar	China	863	34	96.4	853	98.9	SRR7887422
CIR12	Cultivar	China	911	34.6	95.3	903	99.1	SRR7887414	

Supplementary Table 30 *G. hirsutum*-introgressed regions in nine *G. barbadense* accessions

Sample ID	Start position 1 (Mb)	End Position 1 (Mb)	Start position 2 (Mb)	End Position 2 (Mb)
Tanguis	-	-	-	-
Coastland R4-4	43.10	92.00	-	-
Pima S-4	43.10	92.00	-	-
Ashimouni	43.10	92.00	-	-
Giza 45	43.10	92.00	-	-
Xinhai 14#	43.10	92.00	-	-
Xinhai 25#	48.70	57.90	81.60	92.00
Junhai 1#	48.40	54.50	75.00	92.00
Hai7124	43.10	92.00	-	-

Supplementary Table 31 The annotation of identified SNPs and InDels between TM-1 and Hai7124

	TM-1 annotation		Hai7124 annotation	
	SNP	InDel	SNP	InDel
Total	13,845,619	1,358,773	13,845,619	1,358,773
Intergenic	12,616,750	1,112,480	12,543,350	1,103,970
Upstream	319,702	90,061	334,154	90,338
Downstream	253,511	64,883	268,768	65,359
Intronic	454,381	83,274	484,839	90,483
Splicing	747	149	874	170
Exonic	200,528	7,926	213,634	8,453
Stop gain	2,865	152	2,968	176
Stop loss	901	50	994	49
Nonsynonymous SNV	125,613	-	134,314	-
Synonymous SNV	69,976	-	73,404	-
Non-frameshift	-	3,392	-	3,473
Frameshift	-	4,302	-	4,710
Unknown	1,173	30	1,954	45

Supplementary Table 32 Nonrandom distribution of SNP polymorphic regions between TM-1 and Hai7124.

Chr.	Start (Mb)	End (Mb)	Size (Mb)	SNP frequency (SNP/kb)	ID	Type
A01	42.7	92	49.3	0.14	LSPR1	SNP-poor
A01	105.1	108.1	3	0.11	LSPR2	SNP-poor
A01	109.3	110.5	1.2	0.09	LSPR3	SNP-poor
A01	112	112.5	0.5	0.23	LSPR4	SNP-poor
A02	70.8	71.9	1.1	0.07	LSPR5	SNP-poor
A02	102.9	103.4	0.5	0.08	LSPR6	SNP-poor
A03	6.4	6.9	0.5	0.39	LSPR7	SNP-poor
A05	13.1	16.4	3.3	0.59	LSPR8	SNP-poor
A05	18.6	19.7	1.1	0.34	LSPR9	SNP-poor
A05	55.9	57	1.1	0.03	LSPR10	SNP-poor
A05	59.9	61.2	1.3	0.13	LSPR11	SNP-poor
A06	18.8	22.4	3.6	0.25	LSPR12	SNP-poor
A06	80.1	80.9	0.8	0.08	LSPR13	SNP-poor
A08	94.8	101.6	6.8	0.51	LSPR14	SNP-poor
A08	110.3	110.8	0.5	0.15	LSPR15	SNP-poor
A10	89	89.9	0.9	0.24	LSPR16	SNP-poor
A11	9.8	10.3	0.5	0.08	LSPR17	SNP-poor
A11	41.4	42	0.6	0.01	LSPR18	SNP-poor
A11	71.1	71.8	0.7	0.10	LSPR19	SNP-poor
A11	94.9	95.5	0.6	0.10	LSPR20	SNP-poor
A11	111.9	112.6	0.7	0.06	LSPR21	SNP-poor
A12	58.2	58.9	0.7	0.18	LSPR22	SNP-poor
A12	62.8	63.6	0.8	0.05	LSPR23	SNP-poor
D01	16.4	16.9	0.5	0.22	LSPR24	SNP-poor
D01	59.3	59.9	0.6	0.74	LSPR25	SNP-poor
D02	63.5	64.2	0.7	0.18	LSPR26	SNP-poor
D03	47.5	48.2	0.7	0.12	LSPR27	SNP-poor
D04	3.9	4.4	0.5	0.35	LSPR28	SNP-poor
D04	10.1	12.5	2.4	0.11	LSPR29	SNP-poor
D08	25.8	27.4	1.6	0.59	LSPR30	SNP-poor
D09	45.8	46.6	0.8	0.14	LSPR31	SNP-poor
D09	50.6	51.4	0.8	0.36	LSPR32	SNP-poor
D11	10.2	11.3	1.1	0.18	LSPR33	SNP-poor
D11	60.3	60.9	0.6	0.13	LSPR34	SNP-poor
D09	24.5	25.3	0.8	19.73	HSPR1	SNP-rich
D09	28.7	31.5	2.8	17.21	HSPR2	SNP-rich
D10	22.3	23.3	1	20.10	HSPR3	SNP-rich
D10	54.1	54.7	0.6	19.07	HSPR4	SNP-rich
D13	41.4	42.4	1	19.96	HSPR5	SNP-rich

Supplementary Table 33 List of 1.10,366 genes with sequence variations (GSVs).
(included as a separate EXCEL file).

Supplementary Table 34 Detection of GSVs in 19 deep-sequenced accessions.
(included as a separate EXCEL file).

Supplementary Table 35 Summary of GSVs in 19 deep-sequenced accessions.
(included as a separate EXCEL file).

Supplementary Table 36 10 inversions and 32 translocations identified between TM-1 and Hai7124.

Variation ID	Variation Type	TM-1				Hai7124				Orientation
		Chr.	Start (bp)	End (bp)	Length (bp)	Chr.	Start (bp)	End (bp)	Length (bp)	
CGSV00133	Inversion	A01	12,576,710	12,578,523	1,814	A01	12,655,743	12,657,545	1,803	-
CGSV00848	Inversion	A02	11,753,662	11,761,458	7,797	A02	12,069,098	12,077,036	7,939	-
CGSV01275	Inversion	A02	48,577,129	48,579,071	1,943	A02	48,455,244	48,457,188	1,945	-
CGSV02697	Inversion	A03	70,500,819	70,518,316	17,498	A03	67,542,266	67,559,791	17,526	-
CGSV11599	Inversion	A11	31,138,932	31,154,353	15,422	A11	31,127,104	31,142,518	15,415	-
CGSV12942	Inversion	A12	18,997,467	19,002,901	5,435	A12	19,342,474	19,347,919	5,446	+
CGSV18521	Inversion	D06	37,364,243	37,365,353	1,111	D06	35,258,240	35,259,344	1,105	-
CGSV20220	Inversion	D09	11,204,286	11,248,964	44,679	D09	13,004,839	13,034,264	29,426	-
CGSV21551	Inversion	D11	23,165,713	23,190,526	24,814	D11	23,227,314	23,252,969	25,656	-
CGSV23239	Inversion	D13	56,843,771	56,844,934	1,164	D13	53,642,768	53,643,919	1,152	-
CGSV01245	Translocation	A02	45,914,710	45,917,937	3,228	A06	31,323,154	31,326,388	3,235	-
CGSV02695	Translocation	A03	70,283,571	70,287,013	3,443	D13	50,545,440	50,548,875	3,436	+
CGSV03251	Translocation	A04	4,362,753	4,365,396	2,644	A11	108,664,918	108,667,565	2,648	-
CGSV03257	Translocation	A04	7,602,676	7,605,780	3,105	D09	1,723,882	1,726,982	3,101	+
CGSV04391	Translocation	A05	27,555,506	27,559,271	3,766	D05	14,010,397	14,014,161	3,765	-
CGSV04880	Translocation	A05	79,421,108	79,432,363	11,256	A08	3,653,370	3,664,525	11,156	-
CGSV05258	Translocation	A06	4,418,204	4,421,221	3,018	D06	53,148,712	53,151,752	3,041	+
CGSV05687	Translocation	A06	43,221,991	43,224,876	2,886	D05	10,079,367	10,082,252	2,886	+
CGSV06576	Translocation	A06	123,817,833	123,823,074	5,242	D10	1,647,579	1,652,823	5,245	+
CGSV06805	Translocation	A07	20,143,334	20,149,463	6,130	A06	103,777,333	103,783,461	6,129	+
CGSV08873	Translocation	A08	89,659,375	89,667,579	8,205	A01	109,201,998	109,207,257	5,260	+
CGSV09175	Translocation	A09	3,590,974	3,597,096	6,123	A01	6,587,249	6,593,371	6,123	+
CGSV09903	Translocation	A09	70,768,435	70,773,785	5,351	A11	112,036,056	112,041,408	5,353	+
CGSV10035	Translocation	A10	195,559	199,377	3,819	D08	4,197,433	4,201,214	3,782	+

	Transloca									
CGSV10206	tion	A10	16,968,584	16,971,416	2,833	D12	39,653,817	39,656,652	2,836	-
	Transloca									
CGSV10230	tion	A10	18,942,774	18,950,026	7,253	A10	19,066,715	19,073,998	7,284	+
	Transloca									
CGSV11423	tion	A11	14,314,418	14,319,649	5,232	A06	1,256,677	1,261,910	5,234	-
	Transloca									
CGSV12981	tion	A12	22,129,730	22,141,168	11,439	A01	102,174,762	102,186,203	11,442	+
	Transloca									
CGSV14108	tion	A13	6,388,058	6,393,550	5,493	D07	12,590,662	12,596,152	5,491	+
	Transloca									
CGSV15156	tion	D01	5,017,108	5,022,059	4,952	A05	39,143,279	39,148,226	4,948	-
	Transloca									
CGSV15658	tion	D01	63,068,315	63,071,188	2,874	D07	50,861,671	50,864,545	2,875	+
	Transloca									
CGSV15837	tion	D02	13,897,463	13,900,565	3,103	A06	19,652,560	19,655,435	2,876	+
	Transloca									
CGSV17403	tion	D04	50,666,408	50,668,937	2,530	D07	54,839,230	54,841,760	2,531	+
	Transloca									
CGSV17821	tion	D05	31,395,555	31,398,558	3,004	D13	46,508,750	46,511,761	3,012	+
	Transloca									
CGSV18522	tion	D06	37,470,950	37,475,247	4,298	D08	21,469,206	21,473,370	4,165	-
	Transloca									
CGSV18820	tion	D07	177,102	180,212	3,111	A12	98,670,212	98,673,304	3,093	+
	Transloca									
CGSV18847	tion	D07	1,593,367	1,597,258	3,892	D13	12,200,218	12,204,142	3,925	+
	Transloca									
CGSV19300	tion	D07	47,972,994	47,978,265	5,272	A12	79,860,473	79,865,737	5,265	+
	Transloca									
CGSV19706	tion	D08	24,341,106	24,344,297	3,192	A10	20,950,850	20,954,044	3,195	+
	Transloca									
CGSV19714	tion	D08	24,845,176	24,849,756	4,581	D03	24,799,158	24,803,478	4,321	+
	Transloca									
CGSV20795	tion	D10	8,417,614	8,422,949	5,336	D10	55,620,840	55,626,183	5,344	+
	Transloca									
CGSV22549	tion	D12	49,477,047	49,482,386	5,340	A07	17,806,815	17,812,155	5,341	-

Supplementary Table 37 SVs in the genes and genotyping in the 19 deep-sequenced accessions.
(included as a separate EXCEL file).

Supplementary Table 38 Summary of SVs in 19 deep-sequenced accessions.
(included as a separate EXCEL file).

Supplementary Table 39 List of loss-of-function genes by SVs in TM-1 and Hai7124

TM-1		Hai7124	
A _t	D _t	A _t	D _t
GH_A01G0184	GH_D01G0047	GB_A01G0191	GB_D01G0206
GH_A01G0232	GH_D01G0103	GB_A01G0326	GB_D01G0207
GH_A01G0233	GH_D01G0207	GB_A01G0398	GB_D01G0208
GH_A01G0396	GH_D01G0316	GB_A01G0416	GB_D01G0312
GH_A01G0404	GH_D01G0398	GB_A01G0420	GB_D01G0327
GH_A01G0425	GH_D01G0518	GB_A01G0422	GB_D01G0422
GH_A01G0428	GH_D01G0619	GB_A01G0444	GB_D01G0708
GH_A01G0452	GH_D01G0645	GB_A01G0477	GB_D01G0709
GH_A01G0453	GH_D01G0646	GB_A01G0502	GB_D01G0780
GH_A01G0471	GH_D01G0763	GB_A01G0580	GB_D01G0791
GH_A01G0472	GH_D01G0787	GB_A01G0598	GB_D01G1220
GH_A01G0484	GH_D01G1075	GB_A01G0604	GB_D01G1314
GH_A01G0594	GH_D01G1076	GB_A01G0851	GB_D01G1315
GH_A01G0599	GH_D01G1247	GB_A01G0857	GB_D01G1368
GH_A01G0855	GH_D01G1386	GB_A01G0936	GB_D01G1575
GH_A01G0940	GH_D01G1442	GB_A01G0971	GB_D01G1707
GH_A01G0965	GH_D01G1692	GB_A01G1054	GB_D01G1995
GH_A01G1144	GH_D01G1693	GB_A01G1172	GB_D01G2049
GH_A01G1258	GH_D01G1694	GB_A01G1173	GB_D01G2119
GH_A01G1298	GH_D01G1695	GB_A01G1303	GB_D01G2301
GH_A01G1711	GH_D01G1696	GB_A01G1312	GB_D01G2309
GH_A01G1712	GH_D01G2181	GB_A01G1346	GB_D01G2324
GH_A01G1746	GH_D01G2399	GB_A01G2224	GB_D01G2437
GH_A01G1747	GH_D02G0090	GB_A01G2299	GB_D01G2561
GH_A01G1748	GH_D02G0100	GB_A01G2536	GB_D02G0107
GH_A01G1908	GH_D02G0101	GB_A01G2578	GB_D02G0108
GH_A01G2274	GH_D02G0284	GB_A02G0055	GB_D02G0262
GH_A01G2275	GH_D02G0318	GB_A02G0085	GB_D02G0729
GH_A01G2276	GH_D02G0421	GB_A02G0179	GB_D02G0972
GH_A01G2447	GH_D02G0559	GB_A02G0298	GB_D02G0977
GH_A01G2470	GH_D02G0560	GB_A02G0327	GB_D02G1053
GH_A02G0060	GH_D02G0725	GB_A02G0335	GB_D02G1130
GH_A02G0090	GH_D02G0783	GB_A02G0390	GB_D02G1235
GH_A02G0182	GH_D02G0898	GB_A02G0488	GB_D02G1273
GH_A02G0188	GH_D02G0924	GB_A02G0504	GB_D02G1375
GH_A02G0205	GH_D02G1316	GB_A02G0549	GB_D02G1475
GH_A02G0243	GH_D02G1367	GB_A02G0719	GB_D02G1501
GH_A02G0334	GH_D02G1874	GB_A02G0744	GB_D02G1510
GH_A02G0383	GH_D02G2018	GB_A02G0792	GB_D02G1538
GH_A02G0393	GH_D02G2183	GB_A02G0793	GB_D02G1638
GH_A02G0492	GH_D02G2184	GB_A02G0835	GB_D02G1672
GH_A02G0534	GH_D02G2315	GB_A02G0839	GB_D02G1681
GH_A02G0535	GH_D02G2562	GB_A02G0933	GB_D02G1739
GH_A02G0536	GH_D03G0072	GB_A02G0934	GB_D02G1878
GH_A02G0537	GH_D03G0170	GB_A02G1124	GB_D02G1901
GH_A02G0539	GH_D03G0197	GB_A02G1265	GB_D02G1975
GH_A02G0548	GH_D03G0232	GB_A02G1266	GB_D02G1976
GH_A02G0549	GH_D03G0244	GB_A02G1271	GB_D02G2084
GH_A02G0602	GH_D03G0279	GB_A02G1280	GB_D02G2187
GH_A02G0626	GH_D03G0344	GB_A02G1380	GB_D02G2244
GH_A02G0636	GH_D03G0508	GB_A02G1395	GB_D02G2491
GH_A02G0637	GH_D03G0588	GB_A02G1456	GB_D02G2532
GH_A02G0658	GH_D03G0621	GB_A02G1461	GB_D03G0189
GH_A02G0721	GH_D03G0664	GB_A02G1490	GB_D03G0206
GH_A02G0750	GH_D03G0668	GB_A02G1505	GB_D03G0207

GH_A02G0844	GH_D03G0788	GB_A02G1536	GB_D03G0227
GH_A02G0848	GH_D03G0791	GB_A02G1728	GB_D03G0316
GH_A02G0929	GH_D03G0794	GB_A02G1857	GB_D03G0317
GH_A02G0990	GH_D03G0858	GB_A02G1860	GB_D03G0409
GH_A02G1110	GH_D03G0890	GB_A02G1861	GB_D03G0647
GH_A02G1189	GH_D03G0949	GB_A02G1884	GB_D03G0688
GH_A02G1245	GH_D03G1350	GB_A02G1963	GB_D03G0693
GH_A02G1253	GH_D03G1410	GB_A02G1974	GB_D03G0769
GH_A02G1255	GH_D03G1473	GB_A03G0365	GB_D03G0806
GH_A02G1288	GH_D03G1662	GB_A03G0526	GB_D03G0962
GH_A02G1296	GH_D03G1763	GB_A03G0529	GB_D03G1124
GH_A02G1305	GH_D04G0169	GB_A03G0531	GB_D03G1232
GH_A02G1307	GH_D04G0389	GB_A03G0568	GB_D03G1442
GH_A02G1392	GH_D04G0510	GB_A03G0576	GB_D03G1503
GH_A02G1427	GH_D04G0545	GB_A03G0628	GB_D03G1663
GH_A02G1463	GH_D04G0854	GB_A03G0700	GB_D03G1794
GH_A02G1470	GH_D04G0877	GB_A03G0712	GB_D04G0032
GH_A02G1471	GH_D04G0878	GB_A03G0863	GB_D04G0173
GH_A02G1472	GH_D04G1017	GB_A03G0876	GB_D04G0322
GH_A02G1595	GH_D04G1538	GB_A03G0934	GB_D04G0536
GH_A02G1828	GH_D04G1680	GB_A03G0962	GB_D04G0755
GH_A02G1850	GH_D04G1784	GB_A03G1293	GB_D04G0891
GH_A02G1858	GH_D05G0070	GB_A03G1445	GB_D04G0931
GH_A02G1885	GH_D05G0101	GB_A03G1601	GB_D04G0990
GH_A02G1901	GH_D05G0226	GB_A03G1605	GB_D04G1329
GH_A02G1949	GH_D05G0784	GB_A03G1610	GB_D04G1466
GH_A02G2046	GH_D05G0803	GB_A03G1632	GB_D04G1601
GH_A02G2047	GH_D05G0923	GB_A03G1640	GB_D04G1796
GH_A03G0057	GH_D05G0979	GB_A03G1748	GB_D04G1797
GH_A03G0212	GH_D05G1031	GB_A03G1924	GB_D05G0137
GH_A03G0336	GH_D05G1173	GB_A03G2007	GB_D05G0138
GH_A03G0346	GH_D05G1281	GB_A03G2141	GB_D05G0222
GH_A03G0378	GH_D05G1348	GB_A03G2179	GB_D05G0369
GH_A03G0380	GH_D05G1599	GB_A03G2402	GB_D05G0568
GH_A03G0543	GH_D05G1671	GB_A03G2409	GB_D05G0778
GH_A03G0603	GH_D05G1755	GB_A03G2495	GB_D05G1171
GH_A03G0671	GH_D05G1834	GB_A04G0068	GB_D05G1184
GH_A03G0705	GH_D05G2206	GB_A04G0102	GB_D05G1191
GH_A03G0854	GH_D05G2426	GB_A04G0142	GB_D05G1345
GH_A03G0877	GH_D05G2478	GB_A04G0408	GB_D05G1346
GH_A03G0925	GH_D05G2562	GB_A04G0429	GB_D05G1351
GH_A03G0933	GH_D05G2933	GB_A04G0452	GB_D05G1371
GH_A03G0944	GH_D05G2989	GB_A04G0453	GB_D05G1523
GH_A03G0951	GH_D05G3012	GB_A04G0475	GB_D05G2093
GH_A03G0995	GH_D05G3057	GB_A04G0499	GB_D05G2225
GH_A03G1018	GH_D05G3058	GB_A04G0502	GB_D05G2335
GH_A03G1097	GH_D05G3284	GB_A04G0545	GB_D05G2336
GH_A03G1113	GH_D05G3389	GB_A04G0558	GB_D05G2419
GH_A03G1161	GH_D05G3555	GB_A04G0660	GB_D05G2575
GH_A03G1201	GH_D05G3616	GB_A04G0758	GB_D05G2901
GH_A03G1265	GH_D05G3668	GB_A04G0759	GB_D05G3064
GH_A03G1385	GH_D05G3875	GB_A04G0791	GB_D05G3065
GH_A03G1406	GH_D05G4025	GB_A04G0792	GB_D05G3117
GH_A03G1447	GH_D06G0180	GB_A04G0846	GB_D05G3162
GH_A03G1521	GH_D06G0302	GB_A04G0942	GB_D05G3225
GH_A03G1526	GH_D06G0387	GB_A04G1024	GB_D05G3267
GH_A03G1650	GH_D06G0479	GB_A04G1219	GB_D05G3701
GH_A03G1651	GH_D06G0549	GB_A04G1281	GB_D05G3749
GH_A03G1677	GH_D06G0683	GB_A04G1479	GB_D06G0129
GH_A03G1790	GH_D06G0897	GB_A04G1602	GB_D06G0317
GH_A03G1847	GH_D06G1089	GB_A04G1655	GB_D06G0401

GH_A03G1898	GH_D06G1160	GB_A04G1656	GB_D06G0422
GH_A03G1998	GH_D06G1166	GB_A04G1727	GB_D06G0525
GH_A03G2136	GH_D06G1172	GB_A04G1740	GB_D06G0632
GH_A03G2258	GH_D06G1447	GB_A05G0285	GB_D06G0715
GH_A03G2409	GH_D06G1455	GB_A05G0286	GB_D06G0747
GH_A04G0032	GH_D06G1714	GB_A05G0403	GB_D06G0828
GH_A04G0066	GH_D06G1733	GB_A05G0431	GB_D06G0829
GH_A04G0360	GH_D06G1739	GB_A05G0471	GB_D06G0921
GH_A04G0468	GH_D06G2150	GB_A05G0898	GB_D06G0932
GH_A04G0491	GH_D06G2167	GB_A05G1349	GB_D06G0951
GH_A04G0624	GH_D06G2234	GB_A05G1397	GB_D06G1132
GH_A04G0625	GH_D07G0014	GB_A05G1416	GB_D06G1164
GH_A04G0652	GH_D07G0047	GB_A05G1428	GB_D06G1215
GH_A04G0736	GH_D07G0098	GB_A05G1876	GB_D06G1314
GH_A04G0829	GH_D07G0164	GB_A05G2299	GB_D06G1315
GH_A04G0830	GH_D07G0397	GB_A05G2389	GB_D06G1323
GH_A04G0890	GH_D07G0431	GB_A05G2661	GB_D06G1353
GH_A04G0922	GH_D07G0573	GB_A05G2672	GB_D06G1369
GH_A04G1199	GH_D07G0613	GB_A05G2839	GB_D06G1484
GH_A04G1557	GH_D07G0737	GB_A05G2841	GB_D06G1485
GH_A04G1559	GH_D07G0738	GB_A05G2856	GB_D06G1782
GH_A04G1598	GH_D07G0798	GB_A05G2857	GB_D06G1810
GH_A04G1620	GH_D07G1093	GB_A05G2858	GB_D06G1890
GH_A04G1661	GH_D07G1237	GB_A05G2957	GB_D06G2077
GH_A04G1683	GH_D07G1270	GB_A05G3101	GB_D06G2104
GH_A04G1745	GH_D07G1622	GB_A05G3107	GB_D06G2166
GH_A05G0143	GH_D07G1803	GB_A05G3127	GB_D06G2194
GH_A05G0275	GH_D07G1955	GB_A05G3139	GB_D06G2240
GH_A05G0302	GH_D07G2108	GB_A05G3249	GB_D06G2327
GH_A05G0466	GH_D07G2223	GB_A05G3347	GB_D06G2333
GH_A05G0823	GH_D08G0064	GB_A05G3367	GB_D06G2376
GH_A05G1164	GH_D08G0164	GB_A05G3500	GB_D07G0042
GH_A05G1388	GH_D08G0241	GB_A05G3503	GB_D07G0692
GH_A05G1397	GH_D08G0649	GB_A05G3513	GB_D07G0734
GH_A05G1507	GH_D08G0650	GB_A05G3552	GB_D07G0739
GH_A05G2395	GH_D08G0725	GB_A05G3611	GB_D07G0826
GH_A05G2397	GH_D08G0827	GB_A05G3772	GB_D07G0872
GH_A05G2560	GH_D08G0910	GB_A05G3807	GB_D07G0926
GH_A05G2655	GH_D08G0978	GB_A05G3896	GB_D07G0972
GH_A05G2656	GH_D08G0984	GB_A05G3956	GB_D07G1119
GH_A05G2657	GH_D08G0995	GB_A05G3961	GB_D07G1212
GH_A05G2658	GH_D08G1035	GB_A05G4172	GB_D07G1243
GH_A05G2671	GH_D08G1066	GB_A05G4181	GB_D07G1376
GH_A05G2806	GH_D08G1110	GB_A05G4256	GB_D07G1377
GH_A05G2835	GH_D08G1186	GB_A05G4260	GB_D07G1419
GH_A05G2938	GH_D08G1405	GB_A05G4329	GB_D07G1581
GH_A05G3050	GH_D08G1450	GB_A05G4411	GB_D07G1768
GH_A05G3064	GH_D08G1604	GB_A05G4417	GB_D07G1845
GH_A05G3111	GH_D08G1675	GB_A06G0029	GB_D07G1927
GH_A05G3115	GH_D08G1803	GB_A06G0081	GB_D07G1947
GH_A05G3150	GH_D08G1805	GB_A06G0461	GB_D07G2144
GH_A05G3225	GH_D08G1997	GB_A06G0538	GB_D07G2222
GH_A05G3226	GH_D08G2059	GB_A06G0604	GB_D07G2249
GH_A05G3266	GH_D08G2064	GB_A06G0605	GB_D07G2437
GH_A05G3267	GH_D08G2468	GB_A06G0686	GB_D07G2438
GH_A05G3440	GH_D08G2805	GB_A06G0692	GB_D07G2439
GH_A05G3522	GH_D09G0009	GB_A06G0695	GB_D07G2440
GH_A05G3627	GH_D09G0028	GB_A06G0696	GB_D08G0234
GH_A05G3802	GH_D09G0064	GB_A06G0697	GB_D08G0271
GH_A05G3803	GH_D09G0124	GB_A06G0698	GB_D08G0418
GH_A05G3871	GH_D09G0184	GB_A06G0699	GB_D08G0428

GH_A05G3923	GH_D09G0211	GB_A06G0700	GB_D08G0503
GH_A05G3924	GH_D09G0283	GB_A06G0701	GB_D08G0681
GH_A05G3925	GH_D09G0296	GB_A06G0702	GB_D08G1125
GH_A05G4061	GH_D09G0368	GB_A06G0703	GB_D08G1165
GH_A05G4083	GH_D09G0383	GB_A06G0704	GB_D08G1267
GH_A05G4107	GH_D09G0408	GB_A06G0801	GB_D08G1426
GH_A05G4165	GH_D09G0625	GB_A06G0961	GB_D08G1506
GH_A05G4207	GH_D09G0644	GB_A06G0975	GB_D08G1864
GH_A05G4245	GH_D09G0654	GB_A06G0983	GB_D08G2059
GH_A05G4345	GH_D09G0671	GB_A06G1040	GB_D08G2079
GH_A06G0002	GH_D09G0725	GB_A06G1046	GB_D08G2143
GH_A06G0003	GH_D09G0728	GB_A06G1102	GB_D08G2363
GH_A06G0027	GH_D09G0729	GB_A06G1157	GB_D08G2384
GH_A06G0076	GH_D09G0731	GB_A06G1191	GB_D08G2462
GH_A06G0135	GH_D09G0732	GB_A06G1202	GB_D08G2527
GH_A06G0270	GH_D09G0733	GB_A06G1244	GB_D08G2568
GH_A06G0446	GH_D09G0735	GB_A06G1330	GB_D08G2930
GH_A06G0609	GH_D09G0782	GB_A06G1338	GB_D09G0121
GH_A06G0647	GH_D09G1106	GB_A06G1363	GB_D09G0179
GH_A06G0648	GH_D09G1107	GB_A06G1364	GB_D09G0182
GH_A06G0877	GH_D09G1145	GB_A06G1568	GB_D09G0215
GH_A06G0911	GH_D09G1284	GB_A06G1637	GB_D09G0250
GH_A06G0929	GH_D09G1393	GB_A06G1734	GB_D09G0554
GH_A06G0943	GH_D09G1510	GB_A06G1807	GB_D09G0627
GH_A06G1029	GH_D09G1527	GB_A06G1976	GB_D09G0666
GH_A06G1097	GH_D09G1590	GB_A06G2373	GB_D09G0684
GH_A06G1105	GH_D09G1627	GB_A06G2404	GB_D09G0714
GH_A06G1153	GH_D09G1735	GB_A07G0011	GB_D09G0982
GH_A06G1170	GH_D09G2359	GB_A07G0037	GB_D09G1288
GH_A06G1216	GH_D09G2542	GB_A07G0054	GB_D09G1378
GH_A06G1229	GH_D10G0626	GB_A07G0431	GB_D09G1379
GH_A06G1272	GH_D10G0638	GB_A07G0477	GB_D09G1401
GH_A06G1294	GH_D10G0687	GB_A07G0704	GB_D09G1464
GH_A06G1317	GH_D10G0694	GB_A07G0813	GB_D09G1545
GH_A06G1328	GH_D10G0921	GB_A07G0836	GB_D09G1560
GH_A06G1332	GH_D10G1252	GB_A07G0934	GB_D09G1606
GH_A06G1349	GH_D10G1253	GB_A07G1110	GB_D09G1755
GH_A06G1390	GH_D10G1381	GB_A07G1154	GB_D09G1783
GH_A06G1396	GH_D10G1459	GB_A07G1268	GB_D09G2070
GH_A06G1408	GH_D10G1516	GB_A07G1269	GB_D09G2366
GH_A06G1423	GH_D10G1667	GB_A07G1270	GB_D09G2638
GH_A06G1477	GH_D10G1684	GB_A07G1392	GB_D10G0234
GH_A06G1478	GH_D10G1685	GB_A07G1586	GB_D10G0595
GH_A06G1483	GH_D10G1686	GB_A07G1593	GB_D10G0616
GH_A06G1488	GH_D10G1912	GB_A07G1645	GB_D10G0643
GH_A06G1498	GH_D10G2163	GB_A07G1956	GB_D10G0673
GH_A06G1527	GH_D10G2166	GB_A07G1963	GB_D10G0679
GH_A06G1567	GH_D10G2242	GB_A07G2074	GB_D10G0781
GH_A06G1641	GH_D10G2263	GB_A07G2088	GB_D10G0985
GH_A06G1700	GH_D10G2277	GB_A07G2126	GB_D10G1239
GH_A06G1768	GH_D10G2295	GB_A07G2381	GB_D10G1240
GH_A06G1923	GH_D10G2376	GB_A07G2390	GB_D10G1321
GH_A06G1948	GH_D10G2430	GB_A07G2391	GB_D10G1401
GH_A06G2056	GH_D10G2435	GB_A07G2613	GB_D10G1621
GH_A06G2276	GH_D10G2491	GB_A07G2669	GB_D10G1668
GH_A06G2347	GH_D10G2706	GB_A08G0065	GB_D10G1774
GH_A07G0037	GH_D11G0176	GB_A08G0605	GB_D10G1901
GH_A07G0365	GH_D11G0204	GB_A08G0636	GB_D10G1924
GH_A07G0435	GH_D11G0420	GB_A08G0739	GB_D10G2038
GH_A07G0437	GH_D11G0681	GB_A08G0916	GB_D10G2039
GH_A07G0536	GH_D11G0732	GB_A08G1020	GB_D10G2175

GH_A07G0713	GH_D11G0733	GB_A08G1025	GB_D10G2258
GH_A07G0725	GH_D11G0823	GB_A08G1125	GB_D10G2354
GH_A07G0766	GH_D11G0939	GB_A08G1127	GB_D10G2355
GH_A07G0771	GH_D11G0992	GB_A08G1190	GB_D10G2366
GH_A07G0862	GH_D11G1157	GB_A08G1240	GB_D10G2487
GH_A07G0938	GH_D11G1351	GB_A08G1247	GB_D10G2513
GH_A07G1023	GH_D11G1393	GB_A08G1257	GB_D10G2544
GH_A07G1088	GH_D11G1403	GB_A08G1297	GB_D10G2611
GH_A07G1147	GH_D11G1602	GB_A08G1408	GB_D10G2612
GH_A07G1357	GH_D11G1640	GB_A08G1532	GB_D10G2676
GH_A07G1364	GH_D11G1727	GB_A08G1670	GB_D10G2710
GH_A07G1420	GH_D11G1881	GB_A08G1867	GB_D10G2722
GH_A07G1575	GH_D11G2016	GB_A08G2002	GB_D11G0085
GH_A07G1582	GH_D11G2017	GB_A08G2131	GB_D11G0099
GH_A07G1630	GH_D11G2018	GB_A08G2473	GB_D11G0346
GH_A07G1635	GH_D11G2029	GB_A08G2475	GB_D11G0420
GH_A07G1652	GH_D11G2117	GB_A08G2607	GB_D11G0422
GH_A07G1726	GH_D11G2184	GB_A08G2608	GB_D11G0625
GH_A07G1735	GH_D11G2293	GB_A08G2645	GB_D11G0685
GH_A07G1776	GH_D11G2304	GB_A08G2647	GB_D11G0730
GH_A07G1817	GH_D11G2366	GB_A08G2777	GB_D11G0761
GH_A07G1836	GH_D11G2503	GB_A08G2785	GB_D11G1622
GH_A07G1844	GH_D11G2532	GB_A08G2897	GB_D11G1744
GH_A07G1962	GH_D11G2533	GB_A08G2952	GB_D11G1944
GH_A07G2018	GH_D11G2662	GB_A09G0081	GB_D11G2050
GH_A07G2051	GH_D11G3041	GB_A09G0225	GB_D11G2051
GH_A07G2062	GH_D11G3042	GB_A09G0232	GB_D11G2052
GH_A07G2525	GH_D11G3054	GB_A09G0233	GB_D11G2066
GH_A07G2630	GH_D11G3137	GB_A09G0236	GB_D11G2100
GH_A08G0041	GH_D11G3158	GB_A09G0244	GB_D11G2106
GH_A08G0068	GH_D11G3356	GB_A09G0367	GB_D11G2116
GH_A08G0156	GH_D11G3476	GB_A09G0380	GB_D11G2157
GH_A08G0214	GH_D12G0110	GB_A09G0551	GB_D11G2400
GH_A08G0401	GH_D12G0130	GB_A09G0555	GB_D11G2408
GH_A08G0576	GH_D12G0161	GB_A09G0641	GB_D11G2454
GH_A08G0799	GH_D12G0275	GB_A09G0642	GB_D11G2587
GH_A08G0822	GH_D12G0615	GB_A09G0643	GB_D11G2853
GH_A08G0910	GH_D12G0675	GB_A09G0655	GB_D11G2915
GH_A08G0965	GH_D12G0676	GB_A09G0742	GB_D11G3093
GH_A08G0968	GH_D12G0706	GB_A09G0779	GB_D11G3171
GH_A08G0975	GH_D12G0759	GB_A09G0966	GB_D11G3176
GH_A08G1042	GH_D12G0777	GB_A09G1136	GB_D11G3205
GH_A08G1081	GH_D12G1365	GB_A09G1485	GB_D11G3220
GH_A08G1092	GH_D12G1405	GB_A09G1570	GB_D11G3221
GH_A08G1108	GH_D12G1563	GB_A09G1661	GB_D11G3249
GH_A08G1114	GH_D12G1608	GB_A09G1764	GB_D11G3338
GH_A08G1132	GH_D12G1615	GB_A09G1966	GB_D11G3400
GH_A08G1168	GH_D12G1622	GB_A09G2017	GB_D11G3599
GH_A08G1197	GH_D12G2005	GB_A09G2148	GB_D11G3620
GH_A08G1202	GH_D12G2128	GB_A09G2161	GB_D11G3669
GH_A08G1279	GH_D12G2131	GB_A09G2162	GB_D11G3775
GH_A08G1324	GH_D12G2510	GB_A09G2377	GB_D12G0130
GH_A08G1334	GH_D12G2564	GB_A09G2574	GB_D12G0418
GH_A08G1404	GH_D12G2670	GB_A09G2808	GB_D12G0469
GH_A08G1623	GH_D12G2815	GB_A10G0144	GB_D12G0507
GH_A08G2020	GH_D12G2977	GB_A10G0464	GB_D12G0818
GH_A08G2022	GH_D13G0140	GB_A10G0484	GB_D12G0895
GH_A08G2080	GH_D13G0508	GB_A10G0779	GB_D12G0926
GH_A08G2319	GH_D13G0537	GB_A10G0823	GB_D12G0961
GH_A08G2332	GH_D13G0629	GB_A10G0840	GB_D12G0966
GH_A08G2355	GH_D13G0721	GB_A10G0906	GB_D12G1010

GH_A08G2364	GH_D13G0742	GB_A10G1126	GB_D12G1213
GH_A08G2707	GH_D13G0925	GB_A10G1188	GB_D12G1451
GH_A08G2732	GH_D13G1264	GB_A10G1305	GB_D12G1577
GH_A08G2786	GH_D13G1318	GB_A10G1314	GB_D12G1645
GH_A08G2841	GH_D13G1488	GB_A10G1351	GB_D12G1687
GH_A09G0020	GH_D13G1548	GB_A10G1372	GB_D12G1818
GH_A09G0076	GH_D13G1892	GB_A10G1373	GB_D12G2051
GH_A09G0086	GH_D13G2204	GB_A10G1403	GB_D12G2111
GH_A09G0212	GH_D13G2413	GB_A10G1470	GB_D12G2754
GH_A09G0224	GH_D13G2508	GB_A10G1472	GB_D13G0163
GH_A09G0304		GB_A10G1637	GB_D13G0224
GH_A09G0412		GB_A10G1641	GB_D13G0315
GH_A09G0527		GB_A10G1688	GB_D13G0530
GH_A09G0528		GB_A10G1730	GB_D13G0531
GH_A09G0538		GB_A10G1846	GB_D13G0740
GH_A09G0593		GB_A10G1886	GB_D13G0788
GH_A09G0598		GB_A10G2004	GB_D13G0801
GH_A09G0648		GB_A10G2043	GB_D13G0802
GH_A09G0682		GB_A10G2066	GB_D13G0806
GH_A09G0683		GB_A10G2116	GB_D13G0879
GH_A09G0684		GB_A10G2197	GB_D13G0969
GH_A09G0744		GB_A10G2403	GB_D13G1121
GH_A09G0757		GB_A10G2552	GB_D13G1191
GH_A09G0883		GB_A10G2554	GB_D13G1283
GH_A09G1049		GB_A10G2574	GB_D13G1396
GH_A09G1125		GB_A10G2603	GB_D13G1451
GH_A09G1261		GB_A10G2604	GB_D13G1452
GH_A09G1373		GB_A10G2646	GB_D13G1472
GH_A09G1396		GB_A10G2717	GB_D13G1484
GH_A09G1624		GB_A10G2727	GB_D13G1577
GH_A09G1662		GB_A10G2728	GB_D13G1632
GH_A09G1896		GB_A10G2732	GB_D13G1633
GH_A09G1960		GB_A10G2775	GB_D13G1670
GH_A09G2049		GB_A11G0045	GB_D13G1683
GH_A09G2196		GB_A11G0104	GB_D13G1690
GH_A09G2197		GB_A11G0300	GB_D13G1778
GH_A09G2198		GB_A11G0458	GB_D13G1951
GH_A09G2199		GB_A11G0481	GB_D13G1994
GH_A09G2200		GB_A11G0504	GB_D13G1996
GH_A09G2465		GB_A11G0745	GB_D13G2073
GH_A09G2468		GB_A11G0838	GB_D13G2111
GH_A09G2556		GB_A11G1026	GB_D13G2219
GH_A09G2566		GB_A11G1192	GB_D13G2309
GH_A10G0021		GB_A11G1217	GB_D13G2434
GH_A10G0303		GB_A11G1315	GB_D13G2597
GH_A10G0947		GB_A11G1602	
GH_A10G1079		GB_A11G1831	
GH_A10G1202		GB_A11G1908	
GH_A10G1203		GB_A11G1961	
GH_A10G1206		GB_A11G2064	
GH_A10G1231		GB_A11G2090	
GH_A10G1248		GB_A11G2123	
GH_A10G1276		GB_A11G2181	
GH_A10G1340		GB_A11G2242	
GH_A10G1408		GB_A11G2247	
GH_A10G1434		GB_A11G2315	
GH_A10G1530		GB_A11G2372	
GH_A10G1595		GB_A11G2408	
GH_A10G1626		GB_A11G2533	
GH_A10G1629		GB_A11G2734	
GH_A10G1740		GB_A11G2781	

GH_A10G1743	GB_A11G2840
GH_A10G1933	GB_A11G2919
GH_A10G1991	GB_A11G2961
GH_A10G2178	GB_A11G2985
GH_A10G2219	GB_A11G2986
GH_A10G2298	GB_A11G3032
GH_A10G2330	GB_A11G3037
GH_A10G2402	GB_A11G3116
GH_A10G2412	GB_A11G3141
GH_A10G2438	GB_A11G3171
GH_A10G2439	GB_A11G3512
GH_A10G2475	GB_A11G3515
GH_A10G2491	GB_A11G3602
GH_A10G2544	GB_A11G3642
GH_A10G2546	GB_A11G3669
GH_A10G2564	GB_A11G3670
GH_A10G2568	GB_A11G3698
GH_A10G2570	GB_A11G3710
GH_A10G2577	GB_A11G3739
GH_A10G2685	GB_A12G0069
GH_A11G0157	GB_A12G0087
GH_A11G0451	GB_A12G0116
GH_A11G0508	GB_A12G0167
GH_A11G0648	GB_A12G0199
GH_A11G0649	GB_A12G0270
GH_A11G0736	GB_A12G0404
GH_A11G0820	GB_A12G0429
GH_A11G0903	GB_A12G0568
GH_A11G1030	GB_A12G0627
GH_A11G1154	GB_A12G0629
GH_A11G1164	GB_A12G0648
GH_A11G1201	GB_A12G0652
GH_A11G1244	GB_A12G0733
GH_A11G1276	GB_A12G0850
GH_A11G1283	GB_A12G0866
GH_A11G1357	GB_A12G0926
GH_A11G1658	GB_A12G0935
GH_A11G1696	GB_A12G0953
GH_A11G1697	GB_A12G0979
GH_A11G1722	GB_A12G0985
GH_A11G1925	GB_A12G1030
GH_A11G1935	GB_A12G1175
GH_A11G1985	GB_A12G1198
GH_A11G2060	GB_A12G1216
GH_A11G2099	GB_A12G1265
GH_A11G2159	GB_A12G1333
GH_A11G2164	GB_A12G1402
GH_A11G2181	GB_A12G1665
GH_A11G2195	GB_A12G1947
GH_A11G2198	GB_A12G1948
GH_A11G2286	GB_A12G1950
GH_A11G2349	GB_A12G2061
GH_A11G2371	GB_A12G2140
GH_A11G2380	GB_A12G2147
GH_A11G2408	GB_A12G2600
GH_A11G2424	GB_A12G2631
GH_A11G2509	GB_A12G2731
GH_A11G2567	GB_A12G2814
GH_A11G2568	GB_A12G2815
GH_A11G2605	GB_A13G0077
GH_A11G2610	GB_A13G0085

GH_A11G2662	GB_A13G0112
GH_A11G2722	GB_A13G0158
GH_A11G2723	GB_A13G0174
GH_A11G2738	GB_A13G0619
GH_A11G2802	GB_A13G0622
GH_A11G2832	GB_A13G0672
GH_A11G2837	GB_A13G0683
GH_A11G3021	GB_A13G0699
GH_A11G3123	GB_A13G0707
GH_A11G3239	GB_A13G1035
GH_A11G3240	GB_A13G1047
GH_A11G3241	GB_A13G1062
GH_A11G3242	GB_A13G1092
GH_A11G3243	GB_A13G1397
GH_A11G3244	GB_A13G1498
GH_A11G3245	GB_A13G1570
GH_A11G3337	GB_A13G1769
GH_A11G3345	GB_A13G1979
GH_A11G3350	GB_A13G1991
GH_A11G3387	GB_A13G2009
GH_A11G3388	GB_A13G2035
GH_A11G3428	GB_A13G2145
GH_A11G3529	GB_A13G2170
GH_A11G3531	GB_A13G2266
GH_A11G3609	GB_A13G2398
GH_A11G3663	GB_A13G2691
GH_A12G0071	
GH_A12G0117	
GH_A12G0264	
GH_A12G0320	
GH_A12G0321	
GH_A12G0385	
GH_A12G0397	
GH_A12G0409	
GH_A12G0600	
GH_A12G0601	
GH_A12G0624	
GH_A12G0646	
GH_A12G0771	
GH_A12G0783	
GH_A12G0784	
GH_A12G0850	
GH_A12G0851	
GH_A12G0935	
GH_A12G1060	
GH_A12G1062	
GH_A12G1095	
GH_A12G1141	
GH_A12G1152	
GH_A12G1266	
GH_A12G1323	
GH_A12G1431	
GH_A12G1445	
GH_A12G1593	
GH_A12G1685	
GH_A12G1757	
GH_A12G1775	
GH_A12G1806	
GH_A12G1891	
GH_A12G1918	
GH_A12G2102	

GH_A12G2340
GH_A12G2930
GH_A13G0059
GH_A13G0087
GH_A13G0112
GH_A13G0153
GH_A13G0196
GH_A13G0611
GH_A13G0660
GH_A13G0769
GH_A13G0856
GH_A13G0951
GH_A13G0986
GH_A13G1021
GH_A13G1042
GH_A13G1129
GH_A13G1247
GH_A13G1274
GH_A13G1325
GH_A13G1398
GH_A13G1461
GH_A13G1462
GH_A13G1471
GH_A13G1472
GH_A13G1512
GH_A13G1867
GH_A13G2068
GH_A13G2542
GH_A13G2557

Supplementary Table 40 Summary of all identified CNV gene in 19 deep-sequenced cotton accessions.

CNV Types Accessions	Copy number loss	Copy number gain	Copy number loss/gain	Total
Hai7124	1170	615		1785
Giza 45	1146	501		1647
Xinhai 14#	1145	633		1778
Tanguis	1218	698		1916
Coastland R4-4	1169	698		1867
Ashimouni	1066	671		1737
Xinhai 25#	1145	665		1810
Junhai 1#	1168	676		1844
Pima S-4	1139	782		1921
CIR7	64	122		186
Simian 2#	85	72		157
Yucatanense 6	434	325		759
Punctatum 25	148	125		273
Latifolium 13	270	236		506
Deltapine 15	39	42		81
Stoneville 2B	66	66		132
Junmian 1#	75	67		142
CIR12	64	33		97
All accessions	2145	1624	136	3905

Supplementary Table 41 Primers for qRT-PCR analysis of the mentioned genes in this study.

Gene name	Gene ID	Sequences 5'-3'
<i>TST1</i>	<i>GH_A01G0997</i>	
	<i>GB_A01G0993</i>	F: TGCAATCTGTGCTTTGGTTTAT
	<i>GH_D01G1026</i>	R: AAGTGGCATTTCCTTTGGTCTCT
	<i>GB_D01G1066</i>	
<i>ALMT16</i>	<i>GH_A12G1872</i>	
	<i>GB_scaffold7397_objG0001</i>	F: ATTGGAAGAATCTGGGAGCA
	<i>GH_D12G1875</i>	R: CAGTAAACGAGAGACGGGGC
	<i>GB_D12G1947</i>	
<i>NHX1</i>	<i>GH_A11G2946</i>	
	<i>GB_A11G3014</i>	F: GATGCTACTGATATCCCCAGC
	<i>GH_D11G2977</i>	R: GAACAAACCCTCTTCTCCAA
	<i>GB_D11G3012</i>	
<i>VIN1</i>	<i>GH_A12G2191</i>	
	<i>GB_A12G2280</i>	F: TCAACAATGCAAGTGGAGTGAA
	<i>GH_D12G2209</i>	R: AAGGGGAAAGGACGAATAAAAAG
	<i>GB_D12G2282</i>	
<i>PIP</i>	<i>GH_A04G0928</i>	F: ACATTGAGGTTGGTGGTGAG
	<i>GB_A04G0967</i>	R: TAGCAAGGTCGGTCTGGGTC
<i>CaM</i>	<i>GH_A02G0944</i>	F: CTAAACTCACTCCGTCTGCG
	<i>GB_A02G0941</i>	R: CCCGACTTTATGAACGACCT
<i>HSF</i>	<i>GH_A02G1830</i>	F: TTCCAGCTTTGTCCGTCAG
	<i>GB_A02G1863</i>	R: CGATGTTGTCACCGTCTTCC
<i>ETR2</i>	<i>GH_A02G0389</i>	F: AATACTGGAGGAGGGACTGT
	<i>GB_A02G0385</i>	R: TCGGCTTCTACTGCTTGATA
<i>EIN3</i>	<i>GH_A05G1024</i>	F: TAGTGAACTTCGCCTGGGTT
	<i>GB_A05G1032</i>	R: GGTGTTGGAGCAGATGTAAT
<i>ERF</i>	<i>GH_D11G3146</i>	F: CCGCCGCCGTAGCTGAGAAA
	<i>GB_D11G3185</i>	R: TCCGCCGAATCGAAAGTGCC
<i>PYR1</i>	<i>GH_A11G0283</i>	F: AAGGTTTCCAAATGGTCGTG
	<i>GB_A11G0289</i>	R: CTCGCTCATCGTCCAGAATA
<i>HAI2</i>	<i>GH_A05G0933</i>	F: AAGCGGGAGGTCGGGTTATT
	<i>GB_A05G0942</i>	R: AAACCGTCACTCGCCAGAAT
<i>CIPK</i>	<i>GH_A11G1046</i>	F: TATTCTTGATCCAAACCCTC
	<i>GB_A11G1056</i>	R: ATTTACATCCTCATCCTGCT
<i>OST1</i>	<i>GH_D11G0571</i>	F: TGGCGACCTTTATGCTGTCA
	<i>GB_D11G0567</i>	R: GGCAAGATGGGTTGGTGTTA
<i>HOS1</i>	<i>GH_A03G0113</i>	F: GGTAGTTCTGATTCGGTGGT
	<i>GB_A03G0116</i>	R: CCTGTAAAGCCTCATCTGTG
<i>LEA</i>	<i>GH_A09G1035</i>	F: CAGAGGAGCCGCCGAGACAA
	<i>GB_A09G1142</i>	R: ACCGCATCAGTCGCACCTTG

Supplementary Table 42 Expanded gene families of Hai7124.
In separate EXCEL file.

Supplementary Table 43 Expanded gene families of TM-1.
(included as a separate EXCEL file).

Supplementary Table 44 The statistic of response genes under four stress conditions

Stress condition	TM-1		Hai7124		P-value
	Induced	NO.	Induced	NO.	
37°C	16,029	56,732	5,270	69,802	0
4°C	8,725	64,036	12,475	62,596	5.079E-142
Abiotic stress	27,694	45,067	17,957	57,114	0

Supplementary Table 45 Summary of the genes participating in the ethylene and plasma membrane signaling pathways under heat stress in TM-1 and Hai7124.

	TM-1 (gene id)	Heat	Hai7124 (gene id)	Heat
PIPK	<i>GH_D05G0375</i>	1	<i>GB_D05G0376</i>	
	<i>GH_D06G1687</i>	1	<i>GB_D06G1751</i>	
PIP	<i>GH_A01G2320</i>	-1		
	<i>GH_A01G2321</i>	-1		
			<i>GB_A01G2425</i>	-1
			<i>GB_A01G2426</i>	-1
	<i>GH_A03G0269</i>	-1	<i>GB_A03G0263</i>	-1
	<i>GH_A03G0443</i>	-1	<i>GB_A03G0428</i>	
	<i>GH_A03G2291</i>	-1	<i>GB_A03G2378</i>	
	<i>GH_A04G0928</i>	-1	<i>GB_A04G0967</i>	
	<i>GH_A05G1039</i>		<i>GB_A05G1051</i>	-1
	<i>GH_A06G1894</i>	-1	<i>GB_A06G1934</i>	
	<i>GH_A08G2252</i>		<i>GB_A08G2361</i>	-1
	<i>GH_A09G1670</i>	-1	<i>GB_A09G1795</i>	
	<i>GH_A09G1802</i>	-1	<i>GB_A09G1921</i>	
	<i>GH_A10G2271</i>	-1	<i>GB_A10G2425</i>	1
	<i>GH_A10G2583</i>	-1		
	<i>GH_A10G2584</i>	-1		
	<i>GH_A12G0388</i>	-1		
			<i>GB_A12G0396</i>	-1
	<i>GH_D01G0022</i>		<i>GB_D01G0023</i>	-1
	<i>GH_D01G2400</i>	-1		
	<i>GH_D03G1534</i>	-1	<i>GB_D03G1563</i>	
	<i>GH_D03G1698</i>	-1	<i>GB_D03G1732</i>	-1
	<i>GH_D04G1262</i>	-1	<i>GB_D04G1335</i>	
	<i>GH_D06G1923</i>	-1	<i>GB_D06G2002</i>	
	<i>GH_D08G2257</i>		<i>GB_D08G2348</i>	-1
	<i>GH_D09G1616</i>	-1	<i>GB_D09G1633</i>	
	<i>GH_D09G1752</i>	-1	<i>GB_D09G1766</i>	
			<i>GB_D10G2380</i>	-1
	<i>GH_D10G2360</i>	-1	<i>GB_D10G2480</i>	
	<i>GH_D10G2686</i>	-1		
	<i>GH_D10G2687</i>	-1		
	<i>GH_D10G2688</i>	-1		
	<i>GH_D11G3773</i>	-1	<i>GB_D11G3843</i>	
	<i>GH_D12G0314</i>	-1	<i>GB_D12G0320</i>	
	<i>GH_D13G0177</i>	-1	<i>GB_D13G0171</i>	
	<i>GH_scaffold18810_objG0001</i>	-1		
CaM	<i>GH_A02G0367</i>	1	<i>GB_A02G0362</i>	
	<i>GH_A02G0944</i>	1	<i>GB_A02G0941</i>	

	<i>GH_A03G1352</i>	1	<i>GB_A03G1421</i>	
	<i>GH_A05G0774</i>	1	<i>GB_A05G0779</i>	
	<i>GH_A06G0120</i>	1	<i>GB_A06G0122</i>	
	<i>GH_A11G1306</i>	1	<i>GB_A11G1313</i>	
	<i>GH_D02G0392</i>	1	<i>GB_D02G0402</i>	1
	<i>GH_D02G1161</i>	1	<i>GB_D02G1208</i>	
	<i>GH_D05G0772</i>	1	<i>GB_D05G0767</i>	
	<i>GH_D11G1335</i>	1	<i>GB_D11G1349</i>	
HSF	<i>GH_A02G1664</i>	1	<i>GB_A02G1691</i>	
	<i>GH_A02G1830</i>	1	<i>GB_A02G1863</i>	
	<i>GH_A08G2524</i>	1	<i>GB_A08G2639</i>	
ETR1	<i>GH_A01G0366</i>	1	<i>GB_A01G0352</i>	
	<i>GH_D01G0349</i>	1	<i>GB_D01G0372</i>	
ERS1	<i>GH_A11G1179</i>	1	<i>GB_A11G1184</i>	
	<i>GH_D11G1209</i>	1	<i>GB_D11G1218</i>	
ETR2	<i>GH_A02G0389</i>	1	<i>GB_A02G0385</i>	
	<i>GH_A09G2452</i>	1	<i>GB_A09G2559</i>	
	<i>GH_D02G0410</i>	1	<i>GB_D02G0420</i>	
EIN4	<i>GH_A11G2822</i>	1	<i>GB_A11G2889</i>	
	<i>GH_D11G2854</i>	1	<i>GB_D11G2874</i>	
CTR1	<i>GH_A05G2902</i>	-1	<i>GB_A05G2947</i>	
	<i>GH_A05G2903</i>	-1		
	<i>GH_D05G2927</i>	-1	<i>GB_D05G2934</i>	-1
EIN2	<i>GH_D04G1251</i>	1	<i>GB_D04G1323</i>	
	<i>GH_D09G1609</i>	1	<i>GB_D09G1626</i>	
EIN3	<i>GH_A03G0454</i>	1		
	<i>GH_A05G1024</i>	1	<i>GB_A05G1032</i>	
	<i>GH_D03G1523</i>	1		
	<i>GH_D05G1011</i>	1	<i>GB_D05G1008</i>	
	<i>GH_D08G0326</i>	1		
	<i>GH_D08G2246</i>	1	<i>GB_D08G2333</i>	
	<i>GH_D13G2571</i>	1	<i>GB_D13G2665</i>	
ERF	<i>GH_A02G0409</i>	1	<i>GB_A02G0405</i>	
	<i>GH_A07G2403</i>	1	<i>GB_A07G2488</i>	
	<i>GH_A11G1356</i>	1	<i>GB_A11G1363</i>	
	<i>GH_A11G2829</i>	1	<i>GB_A11G2896</i>	
	<i>GH_A11G3118</i>	1	<i>GB_A11G3190</i>	
	<i>GH_A13G1961</i>	1		
	<i>GH_D02G0425</i>	1	<i>GB_D02G0435</i>	
	<i>GH_D11G2860</i>	1		
	<i>GH_D11G3146</i>	1	<i>GB_D11G3185</i>	

Note: The number 1 and -1 represent the up-regulated and down-regulated genes, respectively.

Supplementary Table 46 Summary of the genes participating in the ABA and Calcium Signals under cold stress in TM-1 and Hai7124.

	TM-1 (gene id)	Cold	Hai7124 (gene id)	Cold
ABAR/CHLH	<i>GH_A09G1384</i>	1	<i>GB_A09G1502</i>	
	<i>GH_D09G1334</i>	1	<i>GB_D09G1339</i>	
PYR1	<i>GH_A11G0283</i>	1	<i>GB_A11G0289</i>	
	<i>GH_A12G2288</i>	1	<i>GB_A12G2377</i>	
PYL2	<i>GH_A05G4338</i>		<i>GB_A05G4431</i>	1
	<i>GH_A08G1516</i>	1	<i>GB_A08G1598</i>	
PYL3	<i>GH_A01G2484</i>	1	<i>GB_A01G2592</i>	
PYL6	<i>GH_A11G0999</i>	1	<i>GB_A11G1011</i>	1
	<i>GH_A12G2707</i>		<i>GB_A12G2805</i>	1
PYL8	<i>GH_D08G1529</i>	1	<i>GB_D08G1593</i>	
PYL9	<i>GH_D11G1029</i>	1	<i>GB_D11G1040</i>	1
PYL11	<i>GH_D01G2559</i>	1	<i>GB_D01G2651</i>	
PYL12	<i>GH_D04G0034</i>	1	<i>GB_D04G0038</i>	1
SNRK2.3	<i>GH_A12G0703</i>		<i>GB_A12G0731</i>	1
	<i>GH_D12G0710</i>	1	<i>GB_D12G0711</i>	1
SNRK2.6/	<i>GH_A11G0543</i>	1	<i>GB_A11G0552</i>	1
	<i>GH_A11G2142</i>	1	<i>GB_A11G2179</i>	
	<i>GH_D08G1901</i>		<i>GB_D08G1980</i>	1
	<i>GH_D11G0571</i>	1	<i>GB_D11G0567</i>	
ABI1	<i>GH_D06G0701</i>		<i>GB_D06G0736</i>	1
ABI2	<i>GH_A08G2793</i>	1	<i>GB_A08G2905</i>	1
	<i>GH_D08G2786</i>	1	<i>GB_D08G2895</i>	1
AHG1	<i>GH_A12G2829</i>		<i>GB_A12G2929</i>	1
HAB1	<i>GH_A05G0407</i>		<i>GB_A05G0412</i>	1
	<i>GH_D05G0409</i>		<i>GB_D05G0413</i>	1
	<i>GH_D04G1991</i>		<i>GB_D04G2079</i>	1
PP2CA	<i>GH_A10G2460</i>	1	<i>GB_A10G2630</i>	1
	<i>GH_D10G2569</i>	1	<i>GB_D10G2585</i>	1
			<i>GB_D13G0192</i>	1
	<i>GH_D13G0199</i>	1		
HAI2	<i>GH_A05G0933</i>		<i>GB_A05G0942</i>	1
	<i>GH_D05G0924</i>		<i>GB_D05G0924</i>	1
CBF	<i>GH_A05G0849</i>	1	<i>GB_A05G0856</i>	1
	<i>GH_A11G0086</i>	1	<i>GB_A11G0090</i>	1
	<i>GH_A12G2805</i>	1	<i>GB_A12G2906</i>	1
	<i>GH_D05G0843</i>	1	<i>GB_D05G0838</i>	
	<i>GH_D09G0479</i>	1	<i>GB_D09G0478</i>	
	<i>GH_D11G0092</i>	1	<i>GB_D11G0089</i>	
	<i>GH_D12G2833</i>	1	<i>GB_D12G2916</i>	1
LEA	<i>GH_A01G2197</i>	1	<i>GB_A01G2298</i>	1
	<i>GH_A03G0456</i>	1	<i>GB_A03G0441</i>	
	<i>GH_A03G2308</i>	1	<i>GB_A03G2394</i>	
	<i>GH_A03G2449</i>	1		
	<i>GH_A05G0802</i>	1		
	<i>GH_A05G1023</i>	1	<i>GB_A05G1031</i>	1

	<i>GH_A07G1210</i>	1	<i>GB_A07G1192</i>	1
	<i>GH_A07G1518</i>		<i>GB_A07G1526</i>	1
	<i>GH_A09G1035</i>	1	<i>GB_A09G1142</i>	
			<i>GB_A09G1632</i>	1
	<i>GH_A10G0591</i>	1	<i>GB_A10G0595</i>	
	<i>GH_A11G0963</i>	1	<i>GB_A11G0974</i>	1
	<i>GH_A13G1424</i>	1	<i>GB_A13G1524</i>	
	<i>GH_D01G0718</i>		<i>GB_D01G0749</i>	1
	<i>GH_D01G2279</i>	1	<i>GB_D01G2382</i>	1
	<i>GH_D03G1521</i>	1	<i>GB_D03G1549</i>	
	<i>GH_D05G1010</i>	1	<i>GB_D05G1007</i>	1
	<i>GH_D07G1520</i>	1	<i>GB_D07G1539</i>	1
	<i>GH_D09G1277</i>	1		
			<i>GB_D09G1281</i>	1
	<i>GH_D10G0627</i>	1	<i>GB_D10G0620</i>	
	<i>GH_D11G0994</i>	1	<i>GB_D11G1005</i>	1
	<i>GH_D11G1182</i>	1	<i>GB_D11G1192</i>	1
	<i>GH_D12G0992</i>		<i>GB_D12G0757</i>	1
CSP	<i>GH_A01G0030</i>	1		
			<i>GB_A01G0029</i>	1
	<i>GH_D01G0030</i>	1		
CBL	<i>GH_A06G0967</i>		<i>GB_A06G0999</i>	1
	<i>GH_A11G0271</i>	1	<i>GB_A11G0278</i>	
	<i>GH_D11G0281</i>	1	<i>GB_D11G0279</i>	1
CIPK	<i>GH_A02G0371</i>		<i>GB_A02G0366</i>	1
	<i>GH_A05G0742</i>	1	<i>GB_A05G0744</i>	1
	<i>GH_A08G0969</i>	1	<i>GB_A08G1015</i>	
	<i>GH_A09G0855</i>	1	<i>GB_A09G0964</i>	1
	<i>GH_A10G0377</i>	1	<i>GB_A10G0374</i>	1
	<i>GH_A11G1046</i>	1	<i>GB_A11G1056</i>	
	<i>GH_D02G0395</i>	1	<i>GB_D02G0405</i>	
	<i>GH_D05G0598</i>	1	<i>GB_D05G0599</i>	
	<i>GH_D05G0738</i>	1	<i>GB_D05G0732</i>	1
	<i>GH_D08G0946</i>	1	<i>GB_D08G1010</i>	
	<i>GH_D09G0811</i>	1	<i>GB_D09G0816</i>	1
	<i>GH_D10G0393</i>	1	<i>GB_D10G0375</i>	
	<i>GH_D11G1075</i>	1	<i>GB_D11G1085</i>	1
HOS1	<i>GH_A03G0113</i>		<i>GB_A03G0116</i>	1
	<i>GH_D03G1850</i>		<i>GB_D03G1876</i>	1

Note: The number 1 and -1 represent the up-regulated and down-regulated genes, respectively.

Supplementary Table 47 Statistics of RNAseq libraries in this study.
(included as a separate EXCEL file).