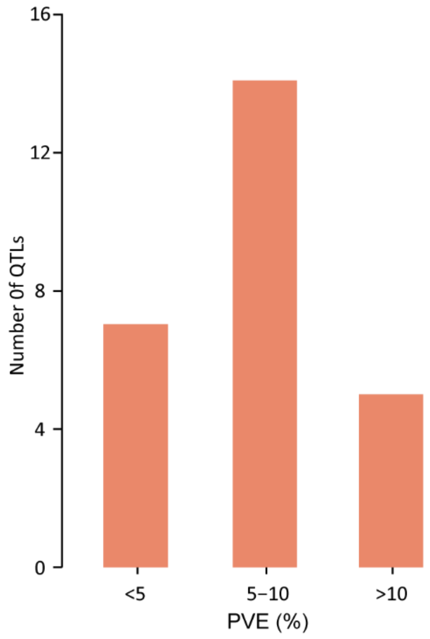


Figure S1. Number of QTLs for starch content identified via the SLM method sorted according to PVE value.

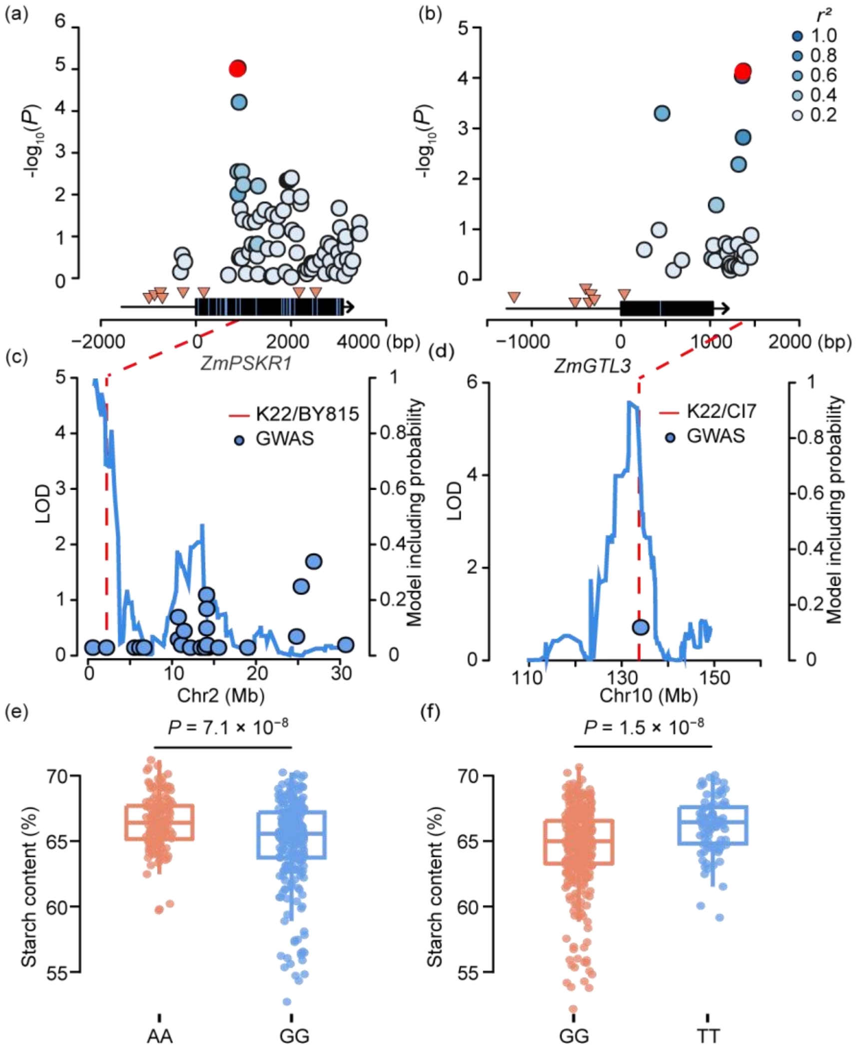


Figure S2 Associations between starch content and *ZmPSKR1* and *ZmGTL3*. (a,b) Associations between the SNPs at the *ZmPSKR1* (a) and *ZmGTL3* (b) loci and starch content. The red dots show the lead SNP with the most significant association. Color coding of the remaining SNPs reflects the extent of their linkage disequilibrium (r^2) with the lead SNP. The black and gray boxes above the x axis represent exons and UTRs, respectively. The InDels (orange triangles) and nonsynonymous SNPs (blue vertical lines) in the promoter and exons are shown. (c, d) Co-localization between candidate genes and the QTL or SNPs identified via SLM and RIL-based GWAS for *ZmPSKR1* (c) and *ZmGTL3* (d). The blue line shows the LOD profile of the QTL identified via SLM for the indicated RILs, whereas the blue dots represent the results of the RIL-GWAS. The red dashed lines indicate the position of candidate genes. (e, f) The genetic effects of the lead SNPs at the *ZmPSKR1* (e) and *ZmGTL3* (f) loci on starch content. The *P*-values were calculated based on a two-tailed Student's *t*-test.

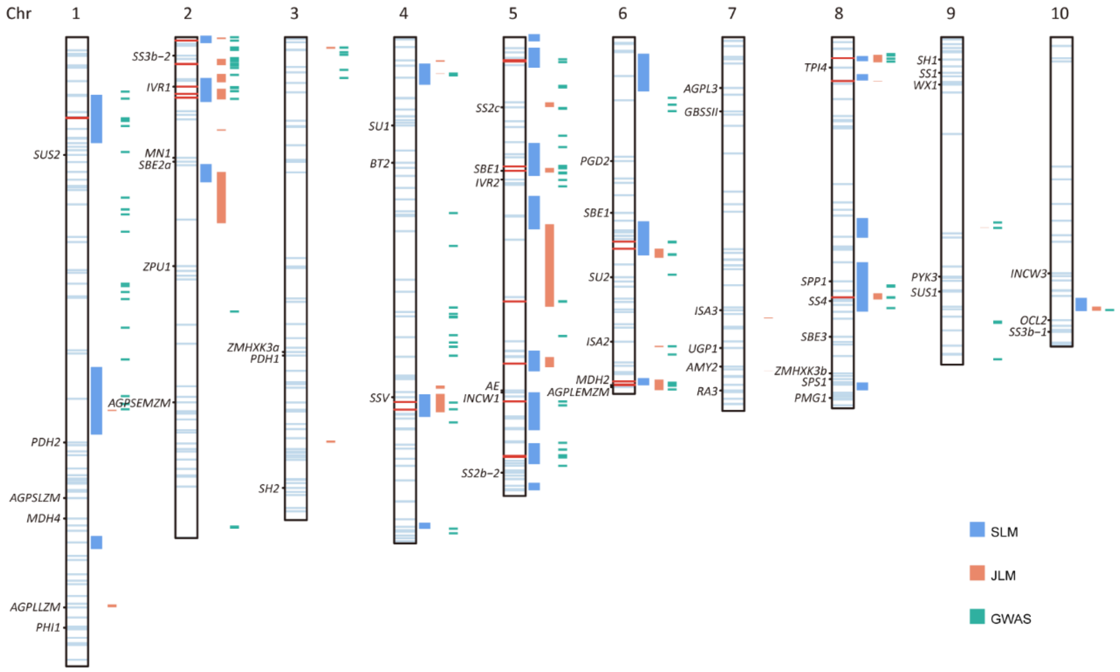


Figure S3. Co-localization of the identified QTLs and 471 genes involved in carbohydrate metabolism. The QTLs identified by SLM, JLM, and GWAS are represented as vertical rectangles of different colors next to each chromosome. The horizontal light blue lines on each chromosome show the positions of the 471 identified genes. The red horizontal lines denote those identified genes that co-localized with the QTLs identified by at least two methods.

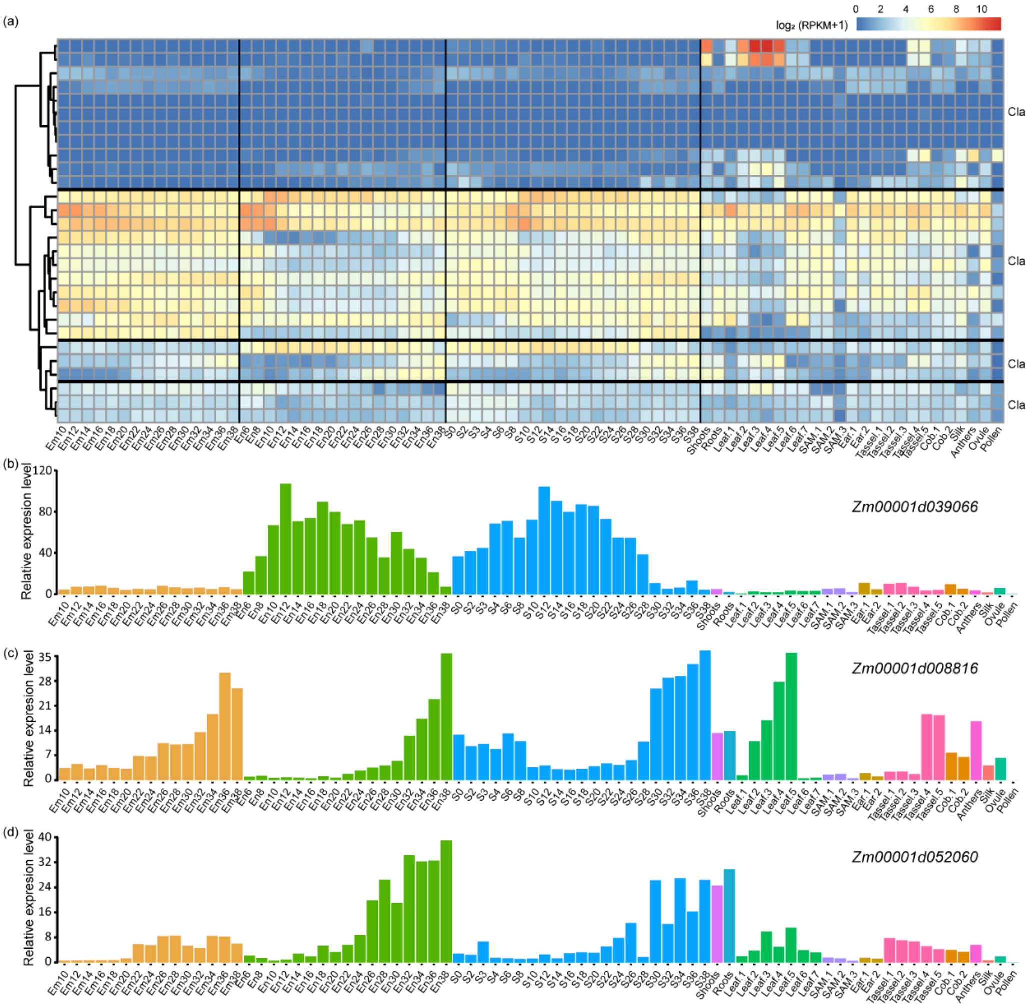
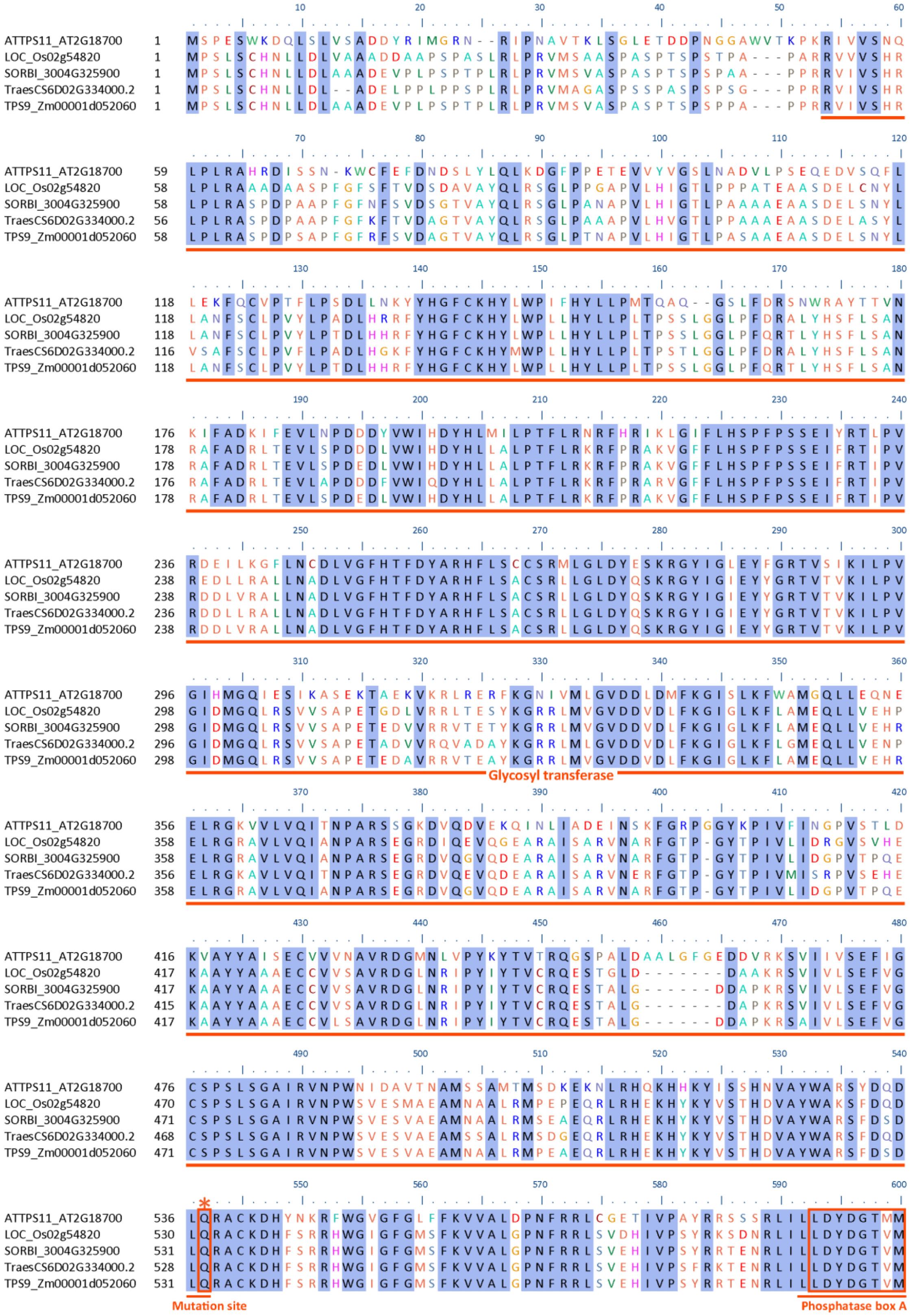


Figure S4. Expression patterns of 28 pathway genes co-localized with the detected QTLs. (a) Hierarchical clustering of 28 genes based on their expressional levels in different tissues from the public transcriptome data (**Dataset S3**; Chen *et al.*, 2014). The log$_2$-based values of one plus the normalized gene transcription level as reads per kilobase per million (RPKM) were used to build the heatmap. The rows in the heat map indicate the genes, and the columns indicate the tissues. Red represents high abundance, and blue represents low abundance. (b–d) Expression pattern of *Zm00001d039066* (b), *Zm00001d008816* (c), and *Zm00001d052060* (d) in various tissues at different stages of development. Em: embryo; En: endosperm; S: seed; SAM: shoot apical meristem.


ATTPS11_AT2G18700
LOC_Os02g54820
SORBI_3004G325900
TraesCS6D02G334000.2
TPS9_Zm00001d052060
Glycosyl transferase
*
Mutation site
Phosphatase box A

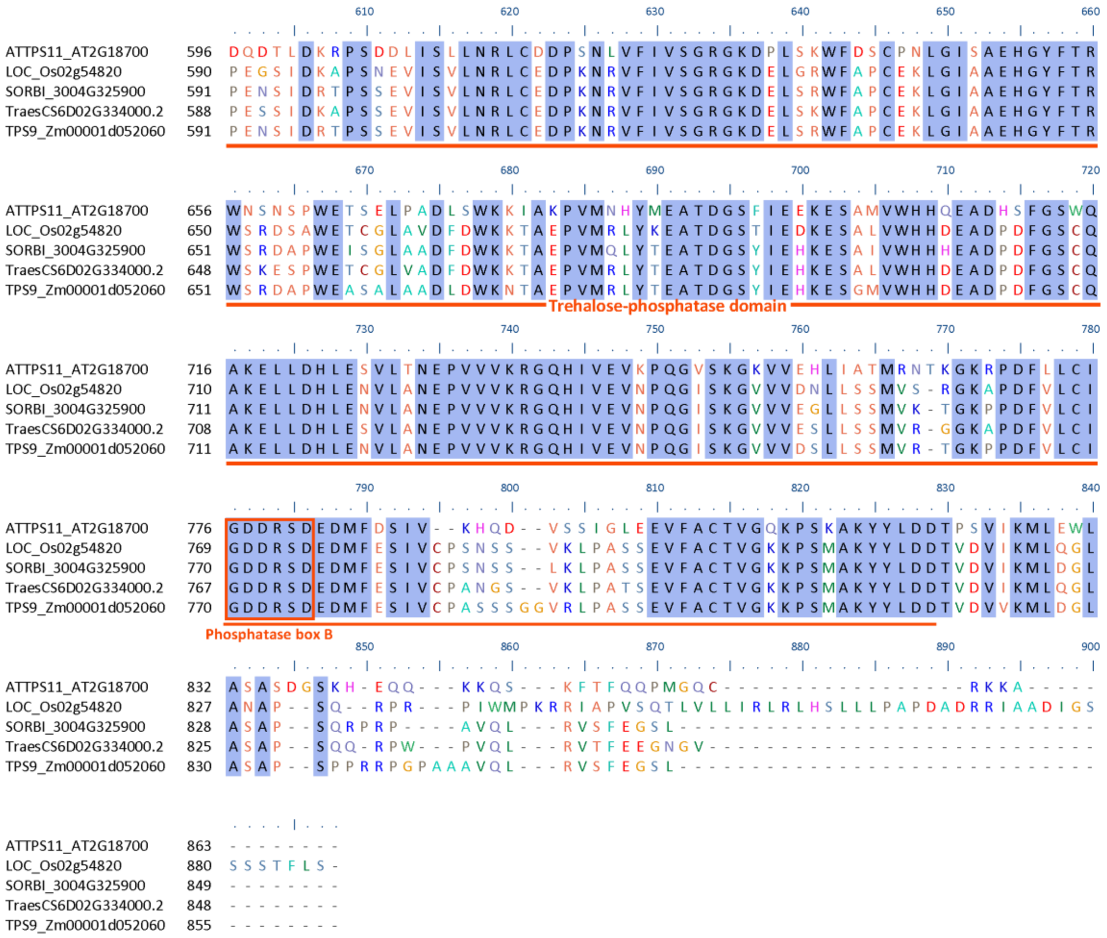


Figure S5. The amino acid sequence alignment of TPS proteins from *Arabidopsis thaliana*, *Oryza sativa*, *Sorghum bicolor*, *Triticum aestivum*, and *Zea mays*. Identical residues are highlighted in blue. The orange lines indicate the glycosyltransferase domain and trehalose-6-phosphate phosphatase-like domain. The trehalose-6-phosphate phosphatase-like domain contains conserved phosphatase consensus sequence boxes A and B, depicted in orange boxes. The scale above indicates the sequence position of amino acids after alignment. The orange star indicates the mutation position of *tps9*.

```
           Promoter
K22  1     CGGTAGCGTAAGCCACCGATGACTGAAAGTAGGGGACGTTCTGCATCACTGCATGAACGAGTATAAACCTTACGCCCTC--TGCAAACCT
CI7  1     CGGTAGCGTAAGCCACCGATGACTGAAAGTAGGGGACGTT--------CTGCATGAACGAGTATAAACCTTATGCCCTCCGCGCAAACCT

K22  89    CCGGAGCCCCAAACACGTATACTAAATTTACTTGTCAGAGCGATGTATGAAACACCAACAATAAGAATAAAAAATTTACTATCCCCGTCT
CI7  83    CCGGAGCCCCAAACACGTATAATAAATTTACTTGTCGGAGCGATGTATGAAACACCAACAATAAGAATAAAAAAATTACCATCCCCGTCT

K22  179   CAATTTATAATGTGCTTGACTTTTTTACTAAATTTGATTGGTTCGTCTTAATAAAGAAGTCTATAAAAAAT---AAAAAATCCAATGGCG
CI7  173   CAATTTATAATTTGCTTGACTTTTTTACTAAGTTTAATTGGTTCGTCTTAATAAAAAAGTCTATAAAAAATAAAAAAAAATCCAATGTCA

K22  266   TACTTAAA-----------GTTAAATAGTATTGCAATAAAAATTAATAATAATTATAAA-TTTTTTAATAAGATGTACCGATCAAACATA
CI7  263   TACTTAAAGCATATTATATGCTAAATACTATCACAATAAAAATTAATAAAAATTATAAATTTTTTTAATAAGATGAACCGATCAAACATA

K22  344   TGATAAAAAATATCAAAGGGAACATAGATTGGTCCCGATATCGAGGGTAACAAGTGGTATTGTGGGTATTTTTATTAAACTTGAACTTGG
CI7  353   TGATAAAAAATATCAAAGGGAACATAGATTGGTCCCGATATCGAGGGTAACAAGTGGTATTGTGGGTATTTTTATTAAACTTGAACTTGG

K22  434   TAAAGTATACTTTGGAATGATC---TATTAATTTGTTGAATATAGAGTATAGATTTAGCGGATTAGTTAAAATCTCAATCTGATTCCGTG
CI7  443   TAAAGTATACTTTGGAATGATCTAGTATTAATTTGTTGAATATAGAGTATAGATTTAGCGGATTAGTTAAAATCTCAATCTGATTCCGTG

K22  521   TACCAAGATCTGGTCAAACGGGCCCATGAACCCACCGTTAGCCGACCGCGACACTGATGCACCCAACCATCCCACATTCCCACGTCGGGG
CI7  533   TACCAAGATCTGGTCAAACGGGCCCGTGAACCCACCGTTAGCCGACCGCGACACTGATGCACCCAACCATCCCACATTCCCACGTCGGGG

K22  611   AAGAGCGGGGCAATGCCGTCATTTCCGCGGGCGAACGTTGTGGATAAGGGCATGTACAGTGGGTGTCTTAAGTTGTGTCTTAGAGTGTGT
CI7  623   AAGAGCGGGGCAGTGCCGTCATTTCCGCGGGCGAACGATGTGGA----------------------------------------------

K22  701   CTAGGGGGGTGAATGTAAAAAATCTCAAGACATGTATCTTGACGAAGACACAGTGTCTTAGCTCTATATTCGAGACAGGAGACTATCTGA
CI7  666   ------------------------------------------------------------------------------------------
                                                       InDel283
K22  791   TTGGTCACTTTAATTTATTGAATGTTCTGATTGGTACAATGAATATGGTAAGACACATGTTTTAGACATGACCACTGTATTATGTTGTGT
CI7  666   ------------------------------------------------------------------------------------------

K22  881   TTTAGCTGTGTCTTATACTTGGAGTACCGTGCAGCTGTATCTAGGTTGTACATGCCCTAAGACTAGAGGCCCAAGCCCATGCGCCCATGC
CI7  666   ---------------------------------------------------------TAAGACTAGAGGCCCAAGCCCATGCGCCCATGC
                                          5'UTR
K22  971   CGTCACACCTCCTACTCTGACCTCCGGGTCGCCCCGTCCGTTCCTTCTTCCTCGCCGCTCAGACACGCACACGCATATCCAGCTTGCCAT
CI7  700   CGTCACACCTCCTACTCTGACCTCCGGGTCGCCCCGTCCGTTCCTTCTTCCTCGCCGCTCAGACACGCACACGCATATCCAGCTTGCCAT

K22  1061  ATTTGTACCACCTGTCCTCCTTCCGTGCCCGAGACCCACCTCGGCGTTCTTATCCCCAGAAACTTCGGCAAAAGTGATCGATCCGTCTCC
CI7  790   ATTTGTACCACCTGTCCTCCTTCCGTGCCCGAGACCCACCTCGGCGTTCTTATCCCCAGAAACTTCGGCAAAAGTGATCGATCCGTCTCC

K22  1151  CACTCCCACCCCGACCGACCGACCGCCGAACGAACCCGTCACACCTCGTCGCGTCGCGGCGCTTCTCTTCTCTTCATGCGCTATCGCCGC
CI7  880   CACTCCCACCCCGACCGACCGACCGCCGAACGAACCCGACACACCTCGTCGCGTCGCGGCGCTTCTCTTCTCTTCATGCGCTATCGCCGC
                                                                Exon1
K22  1241  CGCCTCCACCTCTGCGACGCCCACAGATAGGCAAGAGGAGGACGAGGAGGCGATGCCTTCGCTCTCCTGCCACAACCTCCTCGACCTCGC
CI7  970   CGCCTCCACCTCTGCGACGCCCACAGATAGGCAGGAGGAGGACGAGGAGGCGATGCCTTCGCTCTCCTGCCACAACCTCCTCGACCTCGC

K22  1331  CGCTGCCGACGAGGTGCCCCTGCCGTCGCCGACGCCGCTCCGCCTCCCGCGCGTCATGTCCGTCGCGTCCCCGGCCTCGCCCACCTCGCC
CI7  1060  CGCTGCCGACGAGGTGCCCCTGCCGTCGCCGACGCCGCTCCGCCTCCCGCGCGTCATGTCCGTCGCGTCCCCGGCCTCGCCCACCTCGCC

K22  1421  GTCCCCGCCCGCCCCGCCGCGCCGCGTCATCGTCTCCCACCGTCTGCCGCTCCGCGCGTCGCCGGACCCTTCCGCGCCCTTCGGATTCAG
CI7  1150  GTCCCCGCCCGCCCCGCCGCGCCGCGTCATCGTCTCCCACCGTCTGCCGCTCCGCGCGTCGCCGGACCCTTCCGCGCCCTTCGGATTCAG

K22  1511  GTTCTCCGTCGACGCCGGCACCGTCGCGTACCAGCTCCGCTCGGGGCTGCCCACCAACGCGCCCGTCCTCCACATCGGCACCCTCCCAGC
CI7  1240  GTTCTCCGTCGACGCCGGCACCGTCGCGTACCAGCTCCGCTCGGGGCTGCCCACCAACGCGCCCGTCCTCCACATCGGCACCCTCCCAGC

K22  1601  GGCCGCGGCGGAGGCCGCCTCAGATGAGTTGTCCAACTACCTGCTGGCCAATTTCTCGTGCCTGCCGGTGTACCTTCCCACCGACCTCCA
CI7  1330  GGCCGCGGCGGAGGCCGCCTCAGATGAGTTGTCCAACTACCTGCTGGCCAATTTCTCGTGCCTGCCGGTGTACCTTCCCACCGACCTCCA

K22  1691  CCACCGCTTCTACCACGGATTCTGCAAGCACTACCTCTGGCCGCTCCTCCACTACCTCCTCCCGCTGACGCCGTCCTCGCTCGGTGGCCT
CI7  1420  CCACCGCTTCTACCACGGATTCTGCAAGCACTACCTCTGGCCGCTCCTCCACTACCTCCTCCCGCTGACGCCGTCCTCGCTCGGTGGCCT

K22  1781  CCCTTTCCAGCGAACGCTCTACCACTCCTTCCTCTCGGCCAACAGGGCGTTCGCCGACCGCCTCACCGAGGTGCTGTCCCCCGACGAGGA
CI7  1510  CCCTTTCCAGCGAACGCTCTACCACTCCTTCCTCTCGGCCAACAGGGCGTTCGCCGACCGCCTCACCGAGGTGCTGTCCCCCGACGAGGA

K22  1871  CCTCGTCTGGATCCACGATTACCACCTCCTCGCGCTCCCGACCTTCCTCCGCAAGCGCTTCCCGCGCGCCAAGGTCGGATTCTTCCTCCA
CI7  1600  CCTCGTCTGGATCCACGATTACCACCTCCTCGCGCTCCCGACCTTTCTCCGCAAGCGCTTCCCGCGCGCCAAGGTCGGATTCTTCCTCCA

K22  1961  CTCACCTTTCCCCTCGTCGGAGATCTTCCGTACGATCCCCGTGCGGGATGACCTCGTCCGCGCACTCCTCAACGCCGACCTCGTCGGCTT
CI7  1690  CTCACCTTTCCCCTCGTCGGAGATCTTCCGTACGATCCCCGTGCGGGATGACCTCGTCCGCGCACTCCTCAACGCCGACCTCGTCGGCTT

K22  2051  CCACACCTTCGACTACGCGCGCCACTTCCTGTCCGCGTGCTCGCGGCTGCTCGGCCTCGACTACCAGTCCAAGCGCGGGTACATCGGCAT
CI7  1780  CCACACCTTCGACTACGCGCGCCACTTCCTGTCCGCGTGCTCGCGGCTGCTCGGCCTTGACTACCAGTCCAAGCGCGGGTACATCGGCAT

K22  2141  CGAGTATTACGGCCGCACCGTGACTGTCAAGATACTCCCCGTTGGGATCGACATGGGGCAGCTCAGATCGGTGGTCTCGGCGCCGGAGAC
CI7  1870  CGAGTATTACGGCCGCACCGTGACTGTCAAGATACTGCCCGTCGGGATCGACATGGGGCAGCTCAGATCGGTGGTCTCGGCGCCGGAGAC

K22  2231  GGAGGACGCGGTGCGGCGGGTGACAGAGGCGTACAAGGGGAGGCGCCTCATGGTAGGCGTCGACGACGTCGATCTGTTCAAGGGGATCGG
CI7  1960  GGAGGACGCGGTGCGGCGGGTGACAGAGGCGTACAAGGGGAGGCGCCTCATGGTAGGCGTCGACGACGTCGATCTGTTCAAGGGGATCGG

K22  2321  GCTCAAGTTCCTGGCGATGGAGCAGCTGCTCGTGGAGCACCGGGAGCTCCGCGGCCGCGCCGTGCTGGTGCAGATCGCCAACCCTGCCCG
CI7  2050  GCTCAAGTTCCTGGCGATGGAGCAACTGCTCGTGGAGCACCGGGAGCTCCGCGGCCGCGCCGTGCTGGTGCAGATCGCCAACCCTGCCCG

K22  2411  CAGCGAAGGGCGCGACGTGCAAGGCGTGCAGGACGAGGCCAGGGCCATCAGCGCCCGGGTCAATGCGCGCTTCGGCACCCCCGGGTACAC
CI7  2140  CAGCGAAGGGCGCGACGTGCAAGGCGTGCAGGACGAGGCCAGGGCCATCAGCGCTCGGGTCAATGCGCGCTTCGGCACCCCCGGGTACAC
                               GAC (Asp)
K22  2501  GCCGATCGTGCTCATCGACGACCCGGTGACGCCGCAGGAGAAGGCCGCCTACTACGCGGCCGCCGAGTGCTGCGTGCTGAGCGCCGTGCG
CI7  2230  GCCGATCGTGCTCATCGACGGCCCGGTGACGCCGCAGGAGAAGGCCGCCTACTACGCGGCCGCCGAGTGCTGCGTGCTGAGCGCCGTGCG
                               GGC (Gly)
K22  2591  CGACGGGCTCAACCGGATCCCGTACATCTATACGGTGTGCCGGCAGGAGAGCACCGCGCTCGGAGACGACGCGCCCAAGCGGAGCGCCAT
CI7  2320  CGACGGGCTCAACCGGATCCCGTACATCTATACGGTGTGCCGGCAGGAGAGCACCGCGCTCGGAGACGACGCGCCCAAGCGGAGCGCCAT

K22  2681  CGTGCTCTCGGAGTTCGTCGGTTGCTCCCCGTCCCTCAGCGGCGCGATCCGCGTGAACCCGTGGAGCGTGGAGTCCGTGGCGGAGGCCAT
CI7  2410  CGTGCTCTCGGAGTTCGTCGGTTGTTCCCCGTCCCTCAGCGGCGCGATCCGCGTGAACCCGTGGAGCGTGGAGTCCGTGGCGGAGGCCAT

K22  2771  GAACGCCGCGCTGCGGATGCCGGAGGCCGAGCAGCGGCTGCGGCACGAGAAGCACTACAAGTACGTGAGCACCCACGACGTCGCCTACTG
CI7  2500  GAACGCCGCGCTGCGGATGCCGGAGGCCGAGCAGCGGCTGCGGCACGAGAAGCACTACAAGTACGTGAGCACCCACGACGTCGCCTACTG
```

Figure S6. Sequence comparisons of *ZmTPS9* between CI7 and K22. The numbers on the left indicate the nucleotide positions in the full-length sequences. Polymorphic sites between K22 and CI7 are shown in different colors. The promoter, three exons, and UTRs of *ZmTPS9* are shaded in green, blue, and orange, respectively. The ATG start codon and TGA stop codon are shown with yellow shading. The red underlines highlighted in exon1 and exon3 indicate nonsynonymous SNPs.

Table S1 Statistical summary, broad-sense heritability, and variances associated with starch content in six RIL populations.

Founders		RIL populations								
Inbred line	Starch content (mean ± SD)	Population	Population size	Starch content (mean ± SD)	Range	Variance[a]			h^2 (%)[b]	CI (%)[c]
						σ_g^2	σ_e^2	σ_ε^2		
DE3	69.73 ± 0.79	DE3/BY815	204	62.04 ± 1.96	55.86–66.05	3.85**	1.58**	1.82	86.4	83.3–89.0
K22	67.90 ± 1.02	K22/BY815	207	61.70 ± 2.19	55.00–66.98	4.59**	3.11**	2.01	87.2	84.3–89.7
DAN340	63.77 ± 1.04	BY815/KUI3	187	60.54 ± 1.43	57.45–64.71	2.00**	2.14**	2.03	74.7	68.0–80.0
KUI3	62.85 ± 0.41	K22/CI7	178	65.01 ± 1.55	61.45–69.12	2.76**	0.44**	1.9	81.3	76.8–85.1
B77	62.72 ± 0.34	DAN340/K22	189	66.11 ± 1.02	63.17–68.60	1.29**	0.34**	1.57	71.2	64.4–76.9
CI7	61.96 ± 0.04	KUI3/B77	176	65.25 ± 1.36	61.92–68.40	1.85**	0.82**	1.55	78.1	72.4–82.7
BY815	53.99 ± 0.01									

[a] σ_g^2 is genetic variance, σ_e^2 is the environmental variance, and σ_ε^2 is the residual variance. $^{**}P < 0.01$.

[b] Broad-sense heritability of starch content.

[c] 95% confidence intervals for broad-sense heritability.

Table S2 Summary of polymorphisms of five candidate genes in seven parents via resequencing

Gene ID (Gene Name)	Promoter		5’UTR		Exon			Intron	
	SNPs	InDels	SNPs	InDels	Synonymous SNPs	Non-synonymous SNPs	InDels	SNPs	InDels
Zm00001d029039 (*ZmABCI6*)	NA	NA	NA	NA	16	0	2	111	8
Zm00001d001877 (*ZmPSKR1*)	44	5	0	0	36	22	3	0	0
Zm00001d012818 (*ZmACP3*)	26	0	0	1	3	1	0	5	2
Zm00001d010607 (*ZmCPR*)	54	2	0	0	38	12	0	0	0
Zm00001d025917 (*ZmGTL3*)	34	6	0	0	14	1	1	0	0

NA: None available

Table S3 Associations between the polymorphic sites at the *ZmTPS9* locus and starch content in maize kernels.

Location	Marker[a]	Allele[b]	MAF[c]	*P*-value	Type
Promoter	chr4.S_178024173	C/**T**	0.06	1.2×10^{-1}	Resequencing
Promoter	chr4.S_178024172	C/**T**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178024170	A/**G**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178024134	C/**T**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178024116	**C**/T	0.22	3.2×10^{-1}	Resequencing
Promoter	chr4.S_178024115	**A**/G	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178024035	**C**/T	0.23	3.9×10^{-1}	Resequencing
Promoter	chr4.S_178024034	**A**/G	0.23	3.9×10^{-1}	Resequencing
Promoter	chr4.S_178023991	**1**/0	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023990	**C**/T	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023956	**A**/G	0.14	3.7×10^{-1}	Resequencing
Promoter	chr4.S_178023953	C/**T**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023950	**A**/**G**/-	0.04	2.2×10^{-1}	Resequencing
Promoter	chr4.S_178023910	A/**G**	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023882	**2**/0	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023881	**C**/T	0.22	8.9×10^{-1}	Resequencing
Promoter	chr4.S_178023866	C/**T**	0.2	9.2×10^{-2}	Resequencing
Promoter	chr4.S_178023863	**C**/T	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023851	A/**C**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023836	**A**/G	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023798	**A**/G/T	0.04	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023771	**G**/T	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023763	**C**/**T**/-	0.07	4.6×10^{-1}	Resequencing
Promoter	chr4.S_178023762	1/**0**	0.14	3.7×10^{-1}	Resequencing
Promoter	chr4.S_178023751	**A**/G	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023738	A/**G**	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023727	A/**G**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023724	**0**/1	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023712	**0**/**1**/2	0.003	9.0×10^{-1}	Resequencing
Promoter	chr4.S_178023698	**G**/T	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023696	A/**G**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023687	**11**/0	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023663	C/**T**	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023645	**0**/1	0.14	7.2×10^{-1}	Resequencing
Promoter	chr4.S_178023631	A/**T**	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023620	**C**/T	0.13	4.4×10^{-1}	Resequencing
Promoter	chr4.S_178023598	**A**/G	0.41	9.8×10^{-1}	Resequencing

Location	Marker[a]	Allele[b]	MAF[c]	*P*-value	Type
Promoter	chr4.S_178023597	A/**G**	0.49	8.1×10^{-1}	Resequencing
Promoter	chr4.S_178023596	A/**C**	0.41	9.8×10^{-1}	Resequencing
Promoter	chr4.S_178023545	A/**T**	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023521	C/**T**	0.09	6.1×10^{-1}	Resequencing
Promoter	chr4.S_178023298	0/**283**	0.002	1.6×10^{-1}	Agarose
5'UTR	chr4.S_178023233	**C**/G	0.37	8.9×10^{-1}	0.56 million
5'UTR	chr4.S_178023232	**A**/G	0.37	8.9×10^{-1}	0.56 million
5'UTR	chr4.S_178023154	A/**G**	0.12	6.0×10^{-1}	0.56 million
5'UTR	chr4.S_178023082	C/**G**	0.12	8.1×10^{-1}	0.56 million
5'UTR	chr4.S_178022994	A/**G**	0.45	7.4×10^{-2}	0.56 million
5'UTR	chr4.S_178022993	A/**G**	0.32	9.1×10^{-1}	0.56 million
Exon1	chr4.S_178022711	A/**G**	0.46	7.8×10^{-1}	0.56 million
Exon1	chr4.S_178022635	**A**/C	0.13	6.9×10^{-1}	0.56 million
Exon1	chr4.S_178022588	**A**/G	0.17	3.7×10^{-1}	0.56 million
Exon1	chr4.S_178022417	**G**/T	0.08	4.6×10^{-1}	0.56 million
Exon1	chr4.S_178022321	**A**/G	0.07	7.6×10^{-1}	0.56 million
Exon1	chr4.S_178022270	**A**/G	0.08	4.6×10^{-1}	0.56 million
Exon1	chr4.S_178022252	**G**/T	0.05	6.2×10^{-1}	0.56 million
Exon1	chr4.S_178022129	**A**/G	0.07	7.6×10^{-1}	0.56 million
Exon1	chr4.S_178022108	**C**/G	0.07	8.4×10^{-1}	0.56 million
Exon1	chr4.S_178022060	C/**G**	0.07	8.2×10^{-1}	0.56 million
Exon1	chr4.S_178022054	**A**/G	0.07	8.2×10^{-1}	0.56 million
Exon1	chr4.S_178021967	**C**/T	0.08	4.6×10^{-1}	0.56 million
Exon1	chr4.S_178021892	C/**T**	0.07	7.6×10^{-1}	0.56 million
Exon1	chr4.S_178021772	**A**/G	0.07	7.6×10^{-1}	0.56 million
Exon1	chr4.S_178021716	**C**/T	0.31	2.1×10^{-1}	0.56 million
Exon1	chr4.S_178021676	**A**/G	0.23	9.7×10^{-1}	0.56 million
Exon1	chr4.S_178021616	**A**/G	0.12	9.3×10^{-3}	0.56 million
Exon1	chr4.S_178021547	**A**/C	0.22	4.7×10^{-1}	0.56 million
Exon1	chr4.S_178021532	**A**/G	0.46	4.2×10^{-1}	0.56 million
Exon1	chr4.S_178021511	A/**C**	0.08	1.9×10^{-1}	0.56 million
Exon1	chr4.S_178021478	**C**/G	0.08	1.9×10^{-1}	0.56 million
Exon1	chr4.S_178021211	A/**G**	0.40	2.6×10^{-1}	0.56 million
Exon1	chr4.S_178021172	**C**/T	0.46	5.6×10^{-1}	0.56 million
Exon1	chr4.S_178021073	**A**/C	0.42	9.6×10^{-1}	0.56 million
Exon1	chr4.S_178021064	**A**/G	0.22	9.9×10^{-1}	0.56 million
Exon1	chr4.S_178021010	**A**/G	0.10	7.4×10^{-1}	0.56 million
Intron1	chr4.S_178020972	**G**/T	0.23	2.5×10^{-1}	0.56 million
Intron1	chr4.S_178020924	A/**G**	0.11	4.6×10^{-1}	0.56 million
Intron1	chr4.S_178020909	A/**G**	0.05	9.6×10^{-3}	0.56 million

Location	Marker[a]	Allele[b]	MAF[c]	*P*-value	Type
Intron1	chr4.S_178020888	**C**/T	0.12	2.5×10^{-1}	0.56 million
Intron1	chr4.S_178020886	**A**/T	0.13	1.2×10^{-1}	0.56 million
Intron1	chr4.S_178020880	**C**/G	0.06	1.6×10^{-1}	0.56 million
Intron1	chr4.S_178020872	**A**/T	0.13	3.2×10^{-1}	0.56 million
Intron1	chr4.S_178020842	**C**/G	0.12	4.6×10^{-1}	0.56 million
Intron1	chr4.S_178020478	**A**/C	0.21	8.5×10^{-1}	0.56 million
Intron1	chr4.S_178020477	C/**G**	0.21	8.4×10^{-1}	0.56 million
Exon2	chr4.S_178020361	**C**/T	0.26	1.6×10^{-1}	0.56 million
Intron2	chr4.S_178020086	**C**/T	0.08	5.7×10^{-2}	0.56 million
Intron2	chr4.S_178020047	**C**/G	0.06	8.7×10^{-2}	0.56 million
Intron2	chr4.S_178020017	**A**/C	0.16	4.8×10^{-1}	0.56 million
Intron2	chr4.S_178019995	**C**/G	0.15	9.6×10^{-1}	0.56 million
Intron2	chr4.S_178019877	**A**/C	0.10	7.6×10^{-1}	0.56 million
Intron2	chr4.S_178019876	A/**C**	0.36	4.3×10^{-1}	0.56 million
Exon3	chr4.S_178019687	A/**C**	0.44	4.7×10^{-1}	0.56 million
Exon3	chr4.S_178019586	C/**T**	0.47	1.2×10^{-1}	0.56 million
Exon3	chr4.S_178019585	**A**/G	0.47	1.2×10^{-1}	0.56 million
Exon3	chr4.S_178019462	**A**/C	0.19	2.1×10^{-1}	0.56 million
Exon3	chr4.S_178019441	C/**T**	0.17	8.6×10^{-1}	0.56 million
3'UTR	chr4.S_178019332	C/**T**	0.06	7.8×10^{-1}	0.56 million
3'UTR	chr4.S_178019319	C/**T**	0.30	4.8×10^{-1}	0.56 million
3'UTR	chr4.S_178019237	**A**/C	0.21	6.3×10^{-2}	0.56 million
3'UTR	chr4.S_178019236	**A**/C	0.20	1.0×10^{-2}	0.56 million
3'UTR	chr4.S_178019234	A/**C**	0.04	2.1×10^{-1}	0.56 million
3'UTR	chr4.S_178019233	A/**C**	0.04	2.1×10^{-1}	0.56 million
3'UTR	chr4.S_178019232	**A**/G	0.10	5.9×10^{-1}	0.56 million
3'UTR	chr4.S_178019220	A/**C**	0.09	3.8×10^{-1}	0.56 million
3'UTR	chr4.S_178019219	C/**G**	0.09	3.5×10^{-1}	0.56 million
3'UTR	chr4.S_178019218	**A**/G	0.11	6.3×10^{-1}	0.56 million
3'UTR	chr4.S_178019216	**G**/T	0.14	4.7×10^{-1}	0.56 million
3'UTR	chr4.S_178019210	G/**T**	0.15	2.9×10^{-2}	0.56 million
3'UTR	chr4.S_178019121	C/**G**	0.14	5.2×10^{-1}	0.56 million
3'UTR	chr4.S_178019103	**C**/T	0.26	4.2×10^{-1}	0.56 million

[a] The sites were named based on the physical position according to the B73 reference genome Version 4; blue shading indicates markers that show polymorphism between K22 and CI7.

[b] The underlined nucleotide in a bold font is the favorable allele for each SNP.

[c] Minor allele frequency

Table S4 Primers used in this study.

Type	Primer name	Forward Sequence (5' to 3')	Reverse Sequence (5' to 3')
EMS mutant identification	TPS9-EMS	GTCCAACTACCTGCTGGCTAA	CCGCTCACGATGAACACTCT
AMP508-promoter	TPS9-AMseq	GCCTCATGGCAAGGGAAGAA	GGGCTTGGGCCTCTAGTCTT
AMP508-283InDel	TPS9-AMp283	ACTGATGCACCCAACCATCC	ATATGCGTGTGCGTGTCTGA
Promoter	TPS9-CI7p	TTCCAAACACCGACACCTCG	GCAAGCTGGATATGCGTGTG
	TPS9-K22p	TTTTACACATGCCTTGCGGC	GCAAGCTGGATATGCGTGTG
Gene body	TPS9-G1	CCATGAACCCACCGTTAGCC	GCGTAGTCGAAGGTGTGGAA
	TPS9-EMS	GTCCAACTACCTGCTGGCTAA	CCGCTCACGATGAACACTCT
	TPS9-G2	GCCATCGTGCTCTCTGAGTT	GCTCATGACTCACCTGGGG
	TPS9-G3	GTGACAACGTCTCACCGACT	GCAGCTAATCCTACGCCACA
qRT-PCR	TPS9-qRT	CTGAATCGTCTGTGCGAGGA	GCGGTGTTCTTCCAGTCCAG
qRT-PCR	Actin	AATGCCCAGCAATGTATGT	TTAGGTGGTCGGTGAGGT
Resequencing	ABCI-G1	CACCTTTGCCTTGCAGGTTC	ACCTGCTGCTTTCACGCTAT
	ABCI-G2	CAAACGTCATCCGAATCCGC	GAACCTGCAAGGCAAAGGTG
	ABCI-G3	ATTGGGACTGCAAGGGATCG	CTGTCCTCTGGCTCCATGTC

Type	Primer name	Forward Sequence (5' to 3')	Reverse Sequence (5' to 3')
Resequencing	ABCI-G4	AGCGAGCGTTAGAATCTGGG	TGCGGACCTACGTAGACAGA
	PSKR1-P1	GCCACAAATGTCGAGCTGTC	CCTGTTGGAGAGATCCAGCG
	PSKR1-G1	TCCTTGGGAGTTTCTGCGAC	AGGGTAAATGGGCCATGCAA
	ACP-P1	CAAGGTCCAGCAGGGACAAT	GACGACCGTCGATGGAATCA
	ACP-P2	GCGAGTGACCACAACAGAGA	CGGGCGCTTTGTTATTACGG
	ACP-P3	GGCTTCCACTTGTGCTTTGG	GGAAAGCTTGCTCTTTGCCC
	ACP-G1	CCTTTGCCATTTTTGCGGGT	ACCAACGGCAGGTACAAGAG
	CPR-G1	AGGCGTCCCAAAACAAATGC	ATCCTGGCAATCTCCTGTGC
	CPR-P1	TCATCCCCGCTGGAATCTCA	GATGTCCTGCTGCTCCCATC
	GTL-P1	TCGGATTCAAGCCAGCAACT	TCCCGGAGAATCTGATGGGT
	GTL-G1	ACCCAACTAAAACCACAGGGA	GTGTCCAGGTGGGACTAATGT
Transient expression assays	TPS9-LUC (K22 and CI7)	ACTCACTATAGGGCGAATTGGGTACCGTAGCGTAAGCCACCGATGA	GAACTAGTGGATCCCCCGGGCTGCAGCGCCTCCTCGTCCTCCTC
	TPS9-LUC-1 (K22^{-283bp})	ACTCACTATAGGGCGAATTGGGTACCGTAGCGTAAGCCACCGATGA	GCTTGGGCCTCTAGTCTTATCCACAACGTTCGCCCGCG

Type	Primer name	Forward Sequence (5' to 3')	Reverse Sequence (5' to 3')
	TPS9-LUC-2 (K22^{-283bp})	CGCGGGCGAACGTTGTGGATAAGACTAGAGGCCCAAGC	GAACTAGTGGATCCCCCGGGCTGCAGCGCCTCCTCGTCCTCCTC
	TPS9-LUC-1 (CI7^{+283bp})	ACTCACTATAGGGCGAATTGGGTACCGTAGCGTAAGCCACCGATGA	CCCACTGTACATGCCCTTATCCACATCGTTCGCCCGCGG
	TPS9-LUC-2-indel (CI7^{+283bp})	CCGCGGGCGAACGATGTGGATAAGGGCATGTACAGTGGG	GCTTGGGCCTCTAGTCTTAGGGCATGTACAACCTAGATAC
	TPS9-LUC-3 (CI7^{+283bp})	GTATCTAGGTTGTACATGCCCTAAGACTAGAGGCCCAAGC	GAACTAGTGGATCCCCCGGGCTGCAGCGCCTCCTCGTCCTCCTC