

**Supplementary Table S1.** Steps and codes in TASSEL pipeline including example command lines and brief descriptions. Parameters are highlighted and described when first be used.

<b>FastqToTagCountPlugin</b>		
Step 1	Description	Derives a tagCount list for each FASTQ file. Keeps only good reads having a barcode and a cut site and no N's in the useful part of the sequence. Trims off the barcodes and truncates sequences that (1) have a second cut site, or (2) read into the common adapter.
	Code	<code>/home/tassel/run_pipeline.pl -fork1 -Xms8G -Xmx16G -FastqToTagCountPlugin -i fastq -k key.txt -e ApeKI -o tagcounts -endPlugin -runfork1</code>
	Parameters	-Xms8G, -Xmx16G Set minimum and maximum usage of RAM.
<b>MergeMultipleTagCountPlugin</b>		
Step 2	Description	Merges each tagCount file into a single “master” tagCount list.
	Code	<code>/home/tassel/run_pipeline.pl -Xms8G -Xmx16G -fork1 -MergeMultipleTagCountPlugin -i tagcounts -o mergedtagcounts/BCA965ANXX_4.cnt -c 5 -endPlugin -runfork1</code>
	Parameters	-c 5 Minimum number of times a tag must be present to be output, typically between 5 and 20. The lower value is, the more sequencing errors will be included analysis. Default: 1
	Note:	Sequencing errors can be tolerated and will be removed by following steps.
<b>TagCountToFastqPlugin</b>		
Step 3	Description:	Converts a master tagCount file containing all the tags of interest from binary (.cnt) format into a FASTQ format file (.fq) that can then be used as input to bowtie2.
	Code	<code>/home/tassel/run_pipeline.pl -Xms8G -Xmx16G -fork1 -TagCountToFastqPlugin -i mergedtagcounts/BCA965ANXX_4.cnt -o BCA965ANXX_4.fq -c 5 -endPlugin -runfork1</code>
<b>Bowtie2 Index</b>		
Step 4	Description	Creates a series of support files needed to operate bowtie2.
	Code	<code>bowtie2-build chr1.fa,chr2.fa,chr3.fa,chr4.fa,chr5.fa,chr6.fa,chr7.fa,chr8.fa,chr9.fa,chr10.fa /home/gbs/b73ref/b73_ref.fa</code>
	Notes	Chromosome 1 to 10 from B73_Ref_V4 genome are used to create index files.
<b>Alignment with bowtie2</b>		
Step 5	Description	Aligns the master set of GBS tags to the reference genome.
	Code	<code>bowtie2 -p 8 -N 0 -L 20 -i S,1,0.50 -D 20 -R 3 -x /home/b73ref/b73_ref.fa -U /home/gbs/analysis/BCA965ANXX_4.fq -S /home/gbs/analysis/BCA965ANXX_4.sam</code>
	Parameters	-p 8 The number of processors to be used.
		-N 0 The number of mismatches to allowed in a seed alignment during multiseed alignment. Can be set to 0 or 1. Higher value makes alignment slower but increases sensitivity. Default: 0.
		-L 20 The length of the seed substrings to align during multiseed alignment. Smaller values make alignment slower but more sensitive. Default: 20.
		-I S, 1, 0.50 Sets a function governing the interval between seed substrings to use during multiseed alignment. Decide the seed frame length and interval which can influence the accuracy of alignment.

		-D 20	The times that consecutive seed extension attempts can "fail" before Bowtie 2 moves on. Default: 15.	
		-R 3	the maximum number of times Bowtie 2 will "re-seed" reads with repetitive seeds. Default: 2.	
	Note	Arguments and values used here are identical to --very-sensitive-local.		
	<b>SAMConverterPlugin</b>			
Step 6	Description	Converts a SAM format alignment (.sam) file produced by bowtie2 into a binary TagsOnPhysicalMap (.topm) file that can be used by the TagsToSNPByAlignmentPlugin for calling SNPs.		
	Code	<code>/home/tassel/run_pipeline.pl -Xms8G -Xmx16G -fork1 -SAMConverterPlugin -i BCA965ANXX_4.sam -o topm/BCA965ANXX_4.topm -endPlugin -runfork1</code>		
Step 7	<b>FastqToTBTPlugin</b>			
	Description	Generates a TagsByTaxa file to parse FASTQ files containing raw GBS sequence data for good reads		
	Code	<code>/home/tassel/run_pipeline.pl -Xms8G -Xmx16G -fork1 -FastqToTBTPlugin -i fastq -k key.txt -e ApeKI -o tbt -y -t mergedtagcounts/BCA965ANXX_4.cnt -endPlugin -runfork1</code>		
Step 8	Parameters	-y	Output in TBTByte format (counts from 0-127) instead of TBTBit (0 or 1).	
	<b>MergeTagsByTaxaFilesPlugin</b>			
	Description	Merges all .tbt.bin and/or (preferably) .tbt.byte files present in the input directory.		
Step 9	Code	<code>/home/tassel/run_pipeline.pl -Xms8G -Xmx16G -fork1 -MergeTagsByTaxaFilesPlugin -i tbt -o mergedtbt/BCA965ANXX_4.tbt.byte -endPlugin -runfork1</code>		
	Parameters	-mnF “-0.1”	Minimum value of F. Samples that are not inbreed line should invoke this to be negative value.	
		-sC 1	Start chromosome.	
		-eC 10	End chromosome.	
Step 10	Note	This command is only available in TASSEL 3. The DiscoverySNPCaller in TASSEL 5 replaces this command and remains the same SNP calling functions but generate a db file rather than hapmap file. Except listed parameters, all others were used as default.		
	<b>MergeDuplicateSNPsPlugin</b>			
	Description	Finds duplicate SNPs in the input HapMap file, and merges them if they have the same pair of alleles		
Step 10	Code	<code>/home/tassel/run_pipeline.pl -Xms8G -Xmx16G -fork1 -MergeDuplicateSNPsPlugin -hmp hapmap/raw/BCA965ANXX_4_chr+.hmp.txt -o hapmap/mergedSNPs/BCA965ANXX_4_chr+.hmp.txt -misMat 0.1 -callHets -sC 1 -eC 10 -endPlugin -runfork1</code>		
	Parameters	-misMat 0.1	Threshold genotypic mismatch rate above which the duplicate SNPs won't be merged. Default: 0.05. For lines not fully inbred, value of 0.1 is recommended.	
		-callHets	When two genotypes at a replicate SNP disagree for a	

			taxon, call it a heterozygote. If the germplasm is not fully inbred and contains residual heterozygosity, then -callHets should be on.	
	Note	The -callHets parameter is to guarantee heterozygotes can be correctly output, rather than omitted as errors.		
	<b>GBSHapMapFiltersPlugin</b>			
	Description:	Filtering SNPs with specific requirements.		
Step 11	Code:	<pre>/home/tassel4/run_pipeline.pl -Xms10G -Xmx16G -fork1 -GBSHapMapFiltersPlugin -hmp hapmap/mergedSNPs/BCA965ANXX_4_chr+.hmp.txt -o hapmap/filt/BCA965ANXX_4_chr+.hmp.txt -mnTCov 0.01 -mnSCov 0.2 -mnMAF 0.01 -hLD -mnR2 0.2 -mnBonP 0.005 -sC 1 -eC 10 -endPlugin -runfork1</pre>		
	Parameters:	-mnTCov 0.01	Minimum taxon coverage. The minimum SNP call rate for a taxon to be included in the output	
		-mnSCov 0.2	Minimum site coverage. The minimum taxon call rate for a SNP to be included in the output	
		-mnMAF 0.01	Minimum minor allele frequency	
		-hLD True	filtered for those in statistically significant LD with at least one neighboring SNP	

**Supplementary Table S2.** R codes used for candidate locus/QTL analyses in this study

```
library(qtl)

all <- read.cross("csv", file="SNP.csv", genotypes = c("AA", "AB", "BB"),
na.strings = "NA", alleles = c("A", "B"))

all <- calc.genoprob(all, step=1.0, off.end = 0.0, error.prob = 1.0e-
4, map.function = "haldane", stepwidth = "fixed")

all <- sim.geno(all, n.draws=32, step=1.0, off.end = 0.0, error.prob = 1.0e-
4, map.function = "haldane", stepwidth = "fixed")

all.scan1 <- scanone(all, pheno.col=2, model="binary", method = "em")

all.scan1.perm <- scanone(all, pheno.col = 2, model = "binary", method="em",
n.perm = 1000)

plot(all.scan1, main="LOD plot of regrowth", ylim = c(0,6))

threshold <- summary(all.scan1.perm, alpha=c(0.1, 0.05, 0.01))

abline(h=threshold[1], lty="dashed", lwd=1, col="blue")
abline(h=threshold[2], lty="dashed", lwd=1, col="yellow")
abline(h=threshold[3], lty="dashed", lwd=1, col="red")

summary(all.scan1, perm=all.scan1.perm, lodcolumn=1, alpha=0.1)

mkname1 <- find.marker(all, chr=2, pos=24.244290)
mkname2 <- find.marker(all, chr=7, pos=5.060739)

effectplot(all, pheno.col=2, mname1= mkname1), ylim=c(0,1))
effectplot(all, pheno.col=2, mname1= mkname2), ylim=c(0,1))

write.csv(all.scan1, "all.scan1.csv", row.names = TRUE)
```

**Supplementary Table S3.** Oneway ANOVA Analysis of tiller number at tasseling by regrowth

Source	Df	Sum of Squares	Mean Square	F-Ratio	Prob>F
Regrowth	1	3.115385	3.11538	0.8967	0.3531
Error	24	83.38462	3.47436		
C. Total	25	86.50000			

**Supplementary Table S4.** Segregation of regrowthability among the B73-Zd F3s\*

Plant	PT	Plant	PT	Plant	PT	Plant	PT
BZ3-010-1-1	R	BZ3-010-1-20	NR	BZ3-010-1-46	NR	BZ3-010-1-69	R
BZ3-010-1-2	R	BZ3-010-1-22	R	BZ3-010-1-47	R	BZ3-010-1-70	R
BZ3-010-1-3	R	BZ3-010-1-23	R	BZ3-010-1-48	R	BZ3-010-1-71	R
BZ3-010-1-4	R	BZ3-010-1-24	R	BZ3-010-1-50	R	BZ3-010-1-72	R
BZ3-010-1-5	R	BZ3-010-1-25	R	BZ3-010-1-51	NR	BZ3-010-1-73	R
BZ3-010-1-6	R	BZ3-010-1-26	R	BZ3-010-1-52	R	BZ3-010-1-74	R
BZ3-010-1-7	R	BZ3-010-1-27	R	BZ3-010-1-53	R	BZ3-010-1-75	R
BZ3-010-1-8	NR	BZ3-010-1-30	R	BZ3-010-1-54	R	BZ3-010-1-76	NR
BZ3-010-1-9	NR	BZ3-010-1-31	R	BZ3-010-1-56	R	BZ3-010-1-77	NR
BZ3-010-1-11	NR	BZ3-010-1-32	R	BZ3-010-1-57	R	BZ3-010-1-78	R
BZ3-010-1-12	R	BZ3-010-1-33	NR	BZ3-010-1-68	NR	BZ3-010-1-79	R
BZ3-010-1-13	NR	BZ3-010-1-34	R	BZ3-010-1-59	R	BZ3-010-1-80	R
BZ3-010-1-14	NR	BZ3-010-1-35	R	BZ3-010-1-61	NR	BZ3-010-1-81	NR
BZ3-010-1-15	R	BZ3-010-1-38	NR	BZ3-010-1-62	R	BZ3-010-1-82	NR
BZ3-010-1-16	R	BZ3-010-1-39	R	BZ3-010-1-63	R	BZ3-010-1-83	NR
BZ3-010-1-17	R	BZ3-010-1-40	R	BZ3-010-1-64	NR	BZ3-010-1-84	R
BZ3-010-1-18	R	BZ3-010-1-41	NR	BZ3-010-1-67	R	BZ3-010-1-85	R
BZ3-010-1-19	R	BZ3-010-1-42	NR	BZ3-010-1-68	R	BZ3-010-1-87	R

\*R: regrowth; NR: non-regrowth

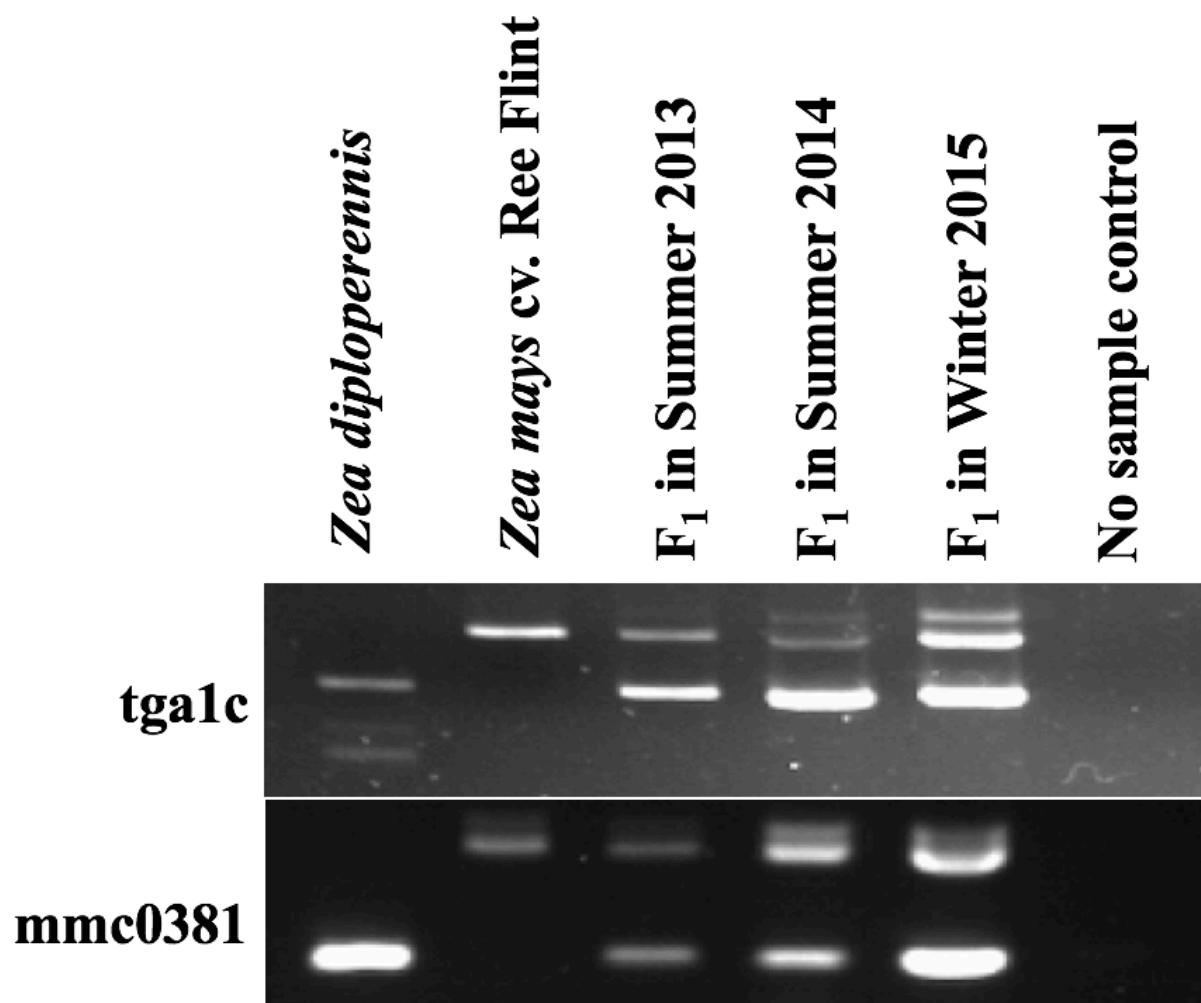
**Supplementary Table S5.** Phenotypes and the *gt1*, *id1* and *tb1* haplotypes of 26 F2 plants and three F3 populations of the *Zea mays* cv Rhee Flint x *Z. diploperennis* cross\*.

Line	PT	<i>tb1</i>	<i>id1</i>	<i>gt1</i>	Line	PT	<i>tb1</i>	<i>id1</i>	<i>gt1</i>	Line	PT	<i>tb1</i>	<i>id1</i>	<i>gt1</i>
F <sub>2</sub> -1	R	3	1	3	F <sub>3</sub> -3-2	R	1	2	1	F <sub>3</sub> -5-13	R	1	2	1
F <sub>2</sub> -2	NR	2	2	2	F <sub>3</sub> -3-3	R	1	2	1	F <sub>3</sub> -5-14	NR	1	2	1
F <sub>2</sub> -3	R	1	2	3	F <sub>3</sub> -3-4	NR	1	2	3	F <sub>3</sub> -5-15	NR	1	2	1
F <sub>2</sub> -4	NR	2	2	2	F <sub>3</sub> -3-6	R	1	2	3	F <sub>3</sub> -5-16	NR	1	2	1
F <sub>2</sub> -5	R	1	2	1	F <sub>3</sub> -3-7	R	1	2	1	F <sub>3</sub> -9-1	R	1	2	1
F <sub>2</sub> -6	R	3	1	1	F <sub>3</sub> -3-8	R	1	2	3	F <sub>3</sub> -9-2	R	1	2	2
F <sub>2</sub> -7	NR	2	2	2	F <sub>3</sub> -3-9	R	1	2	3	F <sub>3</sub> -9-3	R	1	2	2
F <sub>2</sub> -9	R	1	2	3	F <sub>3</sub> -3-10	R	1	2	3	F <sub>3</sub> -9-4	R	1	2	3
F <sub>2</sub> -10	NR	3	1	3	F <sub>3</sub> -3-11	NR	1	2	3	F <sub>3</sub> -9-5	R	1	2	1
F <sub>2</sub> -11	R	2	2	3	F <sub>3</sub> -3-12	R	1	2	1	F <sub>3</sub> -9-6	NR	1	2	2
F <sub>2</sub> -12	R	3	1	1	F <sub>3</sub> -3-13	R	1	2	3	F <sub>3</sub> -9-7	R	1	2	3
F <sub>2</sub> -13	NR	3	1	3	F <sub>3</sub> -3-14	R	1	2	1	F <sub>3</sub> -9-8	R	1	2	1
F <sub>2</sub> -14	NR	2	2	3	F <sub>3</sub> -3-15	R	1	2	1	F <sub>3</sub> -9-9	R	1	2	3
F <sub>2</sub> -15	R	3	1	1	F <sub>3</sub> -3-16	NR	1	2	2	F <sub>3</sub> -9-10	R	1	2	3
F <sub>2</sub> -16	NR	2	2	1	F <sub>3</sub> -5-1	R	1	2	1	F <sub>3</sub> -9-11	R	1	2	1
F <sub>2</sub> -17	R	3	1	3	F <sub>3</sub> -5-2	R	1	2	1	F <sub>3</sub> -9-12	R	1	2	3
F <sub>2</sub> -18	NR	3	1	2	F <sub>3</sub> -5-3	NR	1	2	1	F <sub>3</sub> -9-13	R	1	2	1
F <sub>2</sub> -19	NR	3	1	2	F <sub>3</sub> -5-4	NR	1	2	1	F <sub>3</sub> -9-14	NR	1	2	3
F <sub>2</sub> -20	R	3	1	3	F <sub>3</sub> -5-5	R	1	2	1	F <sub>3</sub> -9-15	NR	1	2	3
F <sub>2</sub> -21	NR	2	2	3	F <sub>3</sub> -5-6	R	1	2	1	F <sub>3</sub> -9-16	R	1	2	1
F <sub>2</sub> -22	NR	2	2	3	F <sub>3</sub> -5-7	R	1	2	1	F <sub>3</sub> -12-1	R	1	2	1
F <sub>2</sub> -23	R	2	2	3	F <sub>3</sub> -5-8	R	1	2	1	F <sub>3</sub> -12-2	R	1	2	1
F <sub>2</sub> -24	R	1	2	3	F <sub>3</sub> -5-9	R	1	2	1	F <sub>3</sub> -12-3	R	1	2	1
F <sub>2</sub> -25	R	2	1	1	F <sub>3</sub> -5-10	R	1	2	1	F <sub>3</sub> -12-4	R	3	1	1
F <sub>2</sub> -26	R	3	2	1	F <sub>3</sub> -5-11	NR	1	2	1	F <sub>3</sub> -12-5	R	3	1	1
F <sub>3</sub> -3-1	R	1	2	3	F <sub>3</sub> -5-12	NR	1	2	1					

\* For phenotype (PT), "R" is for regrowth and "NR" is for non-regrowth; for genotypes, "1" is homozygous for the *Zea diploperennis* allele, "2" is homozygous for the *Z. mays*'s allele and "3" is for heterozygous.



**Supplementary Figure S1.** A photo showing the growth of *Zea diploperennis* and its F<sub>1</sub> with *Z. mays* B73 or Mo17 in the field in the Summer 2017. These plants started the growth in 2013 and have since lived through life cycles alternatively in greenhouse in winters and in the field in summers at SDSU campus at Brookings, SD.



**Supplementary Figure S2.** An agarose gel image showing that two molecular markers confirmed the heterozygosity of a *Z. diploperennis*-*Z. mays* cv. Rhee Flint  $F_1$  plant over three life-cycles.



**Supplementary Figure S3.** A photo showing growth of the B73 - *Z. diploperennis* F<sub>4</sub>s in the field in the summer of 2017.