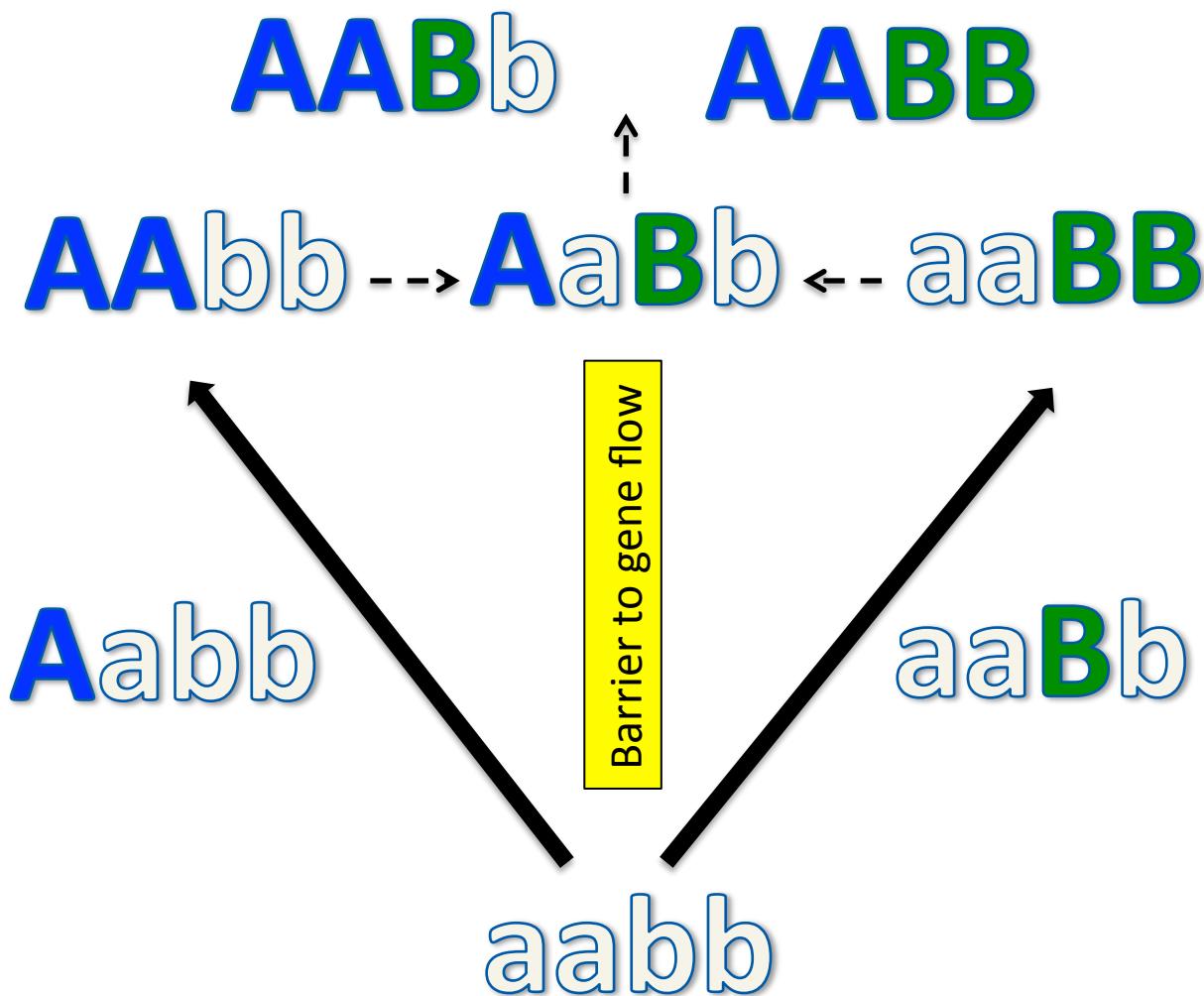


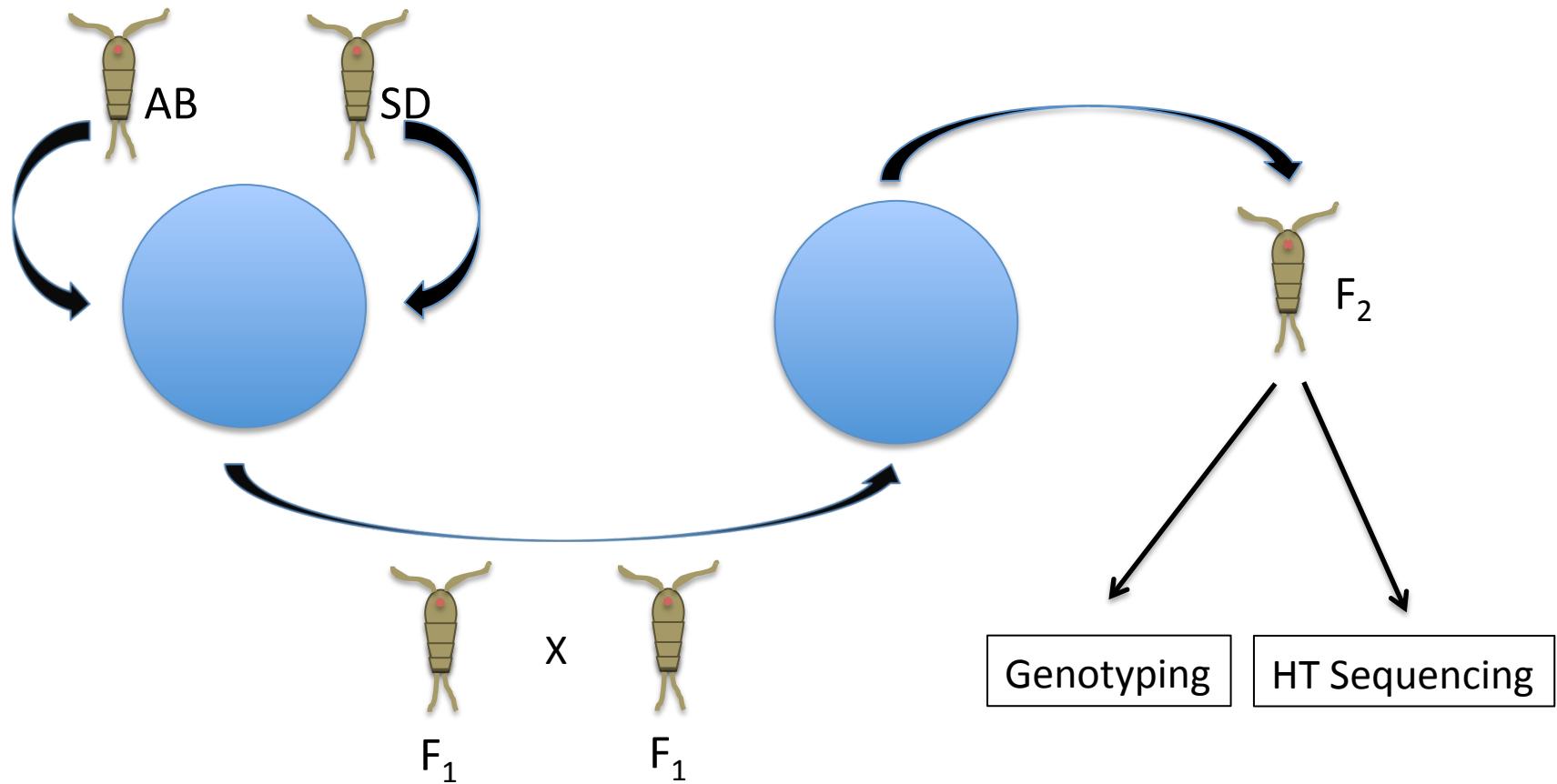
Searching for hybrid inviability across a hybrid genome using Pool-Seq data

Thiago G. Lima
Burton Lab

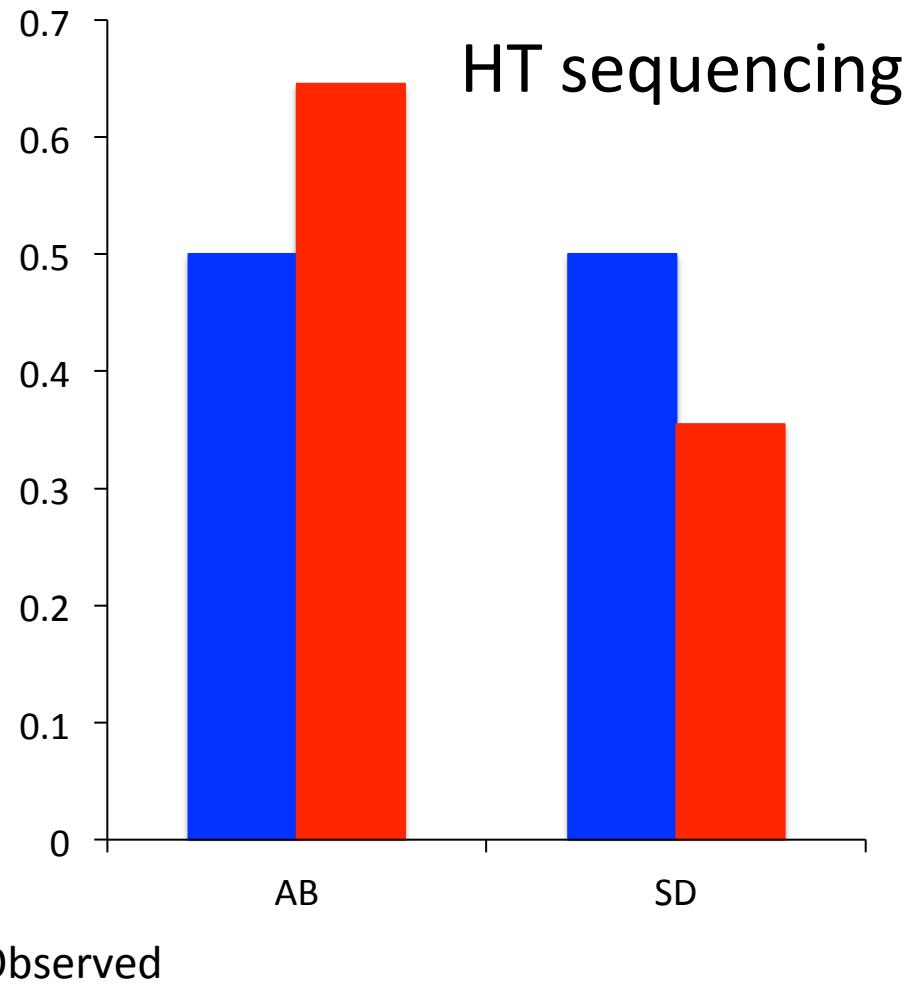
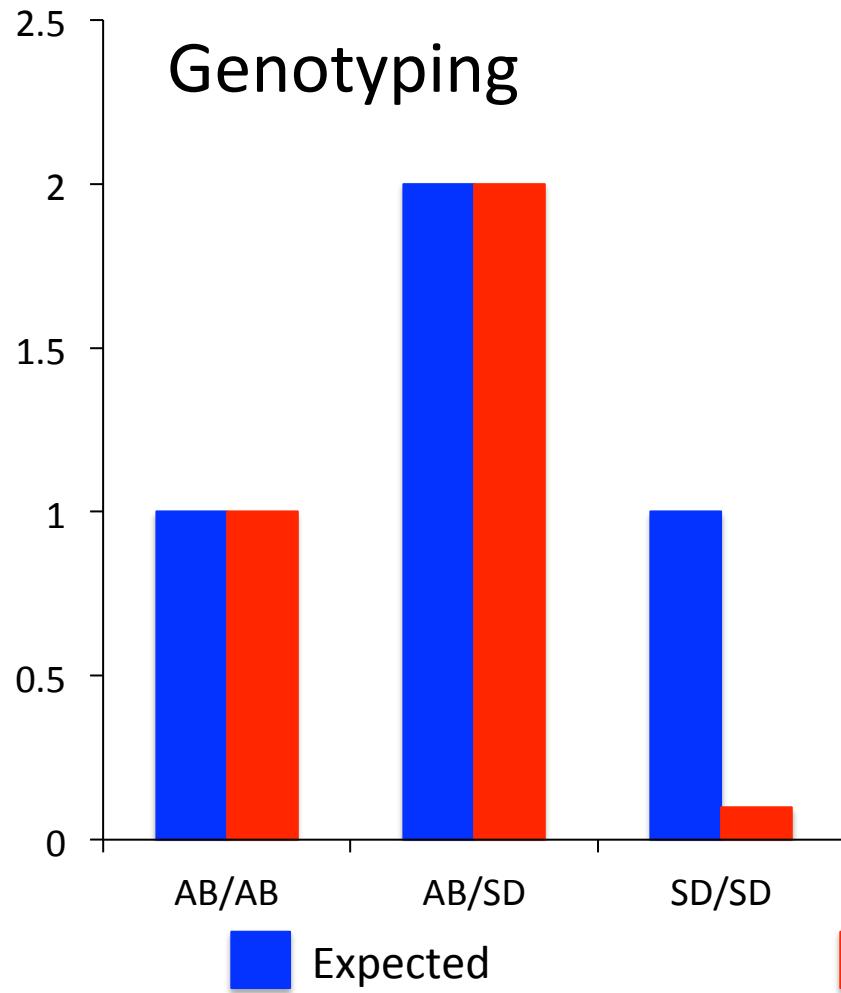
Dobzhansky-Muller Incompatibilities (DMI)



Determining the genetics of F_2 hybrid inviability



Determining the genetics of F_2 hybrid inviability



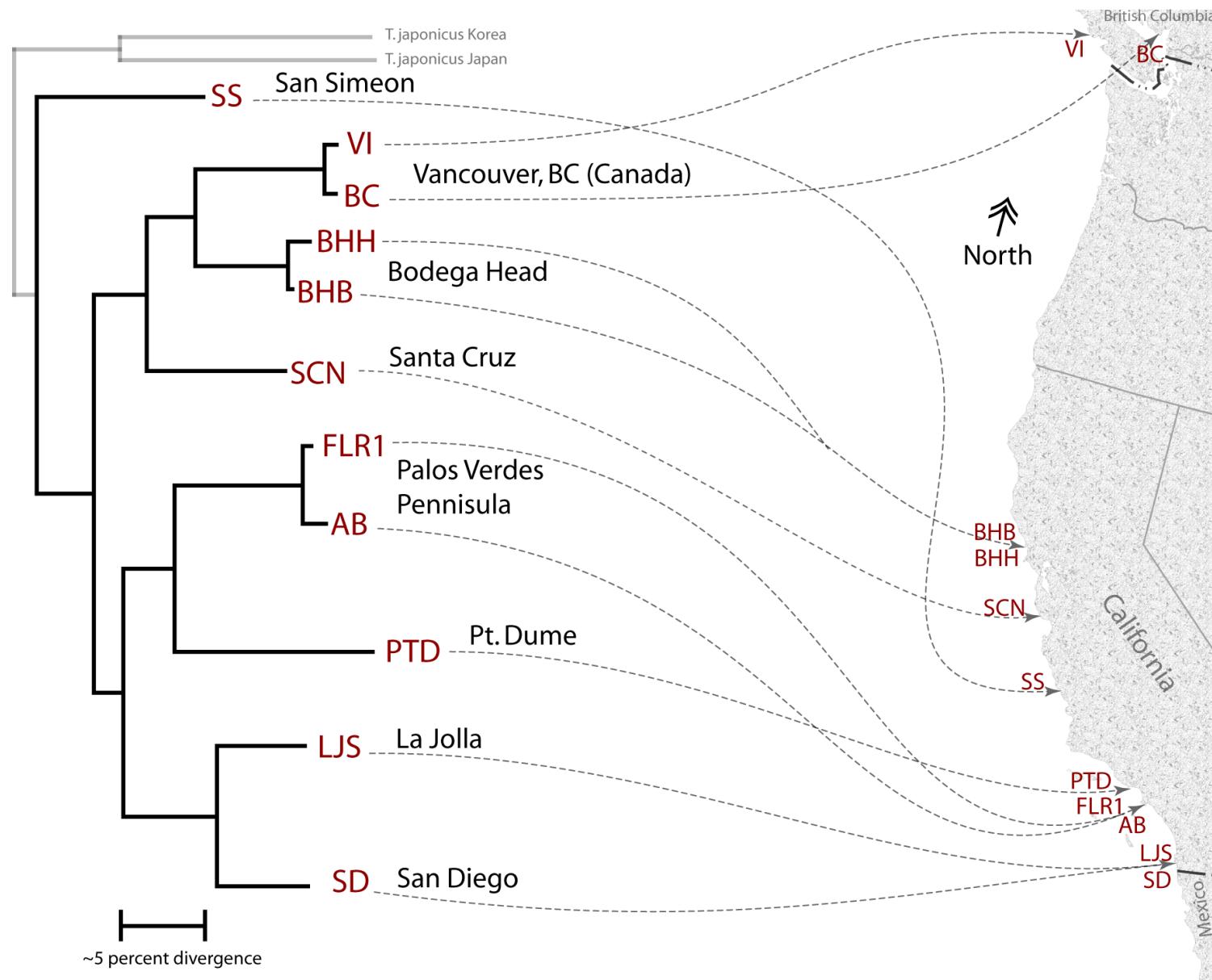
Tigriopus californicus

- Harpacticoid copepod
- Inhabits splash pools in rocky intertidal Pacific coast of North America

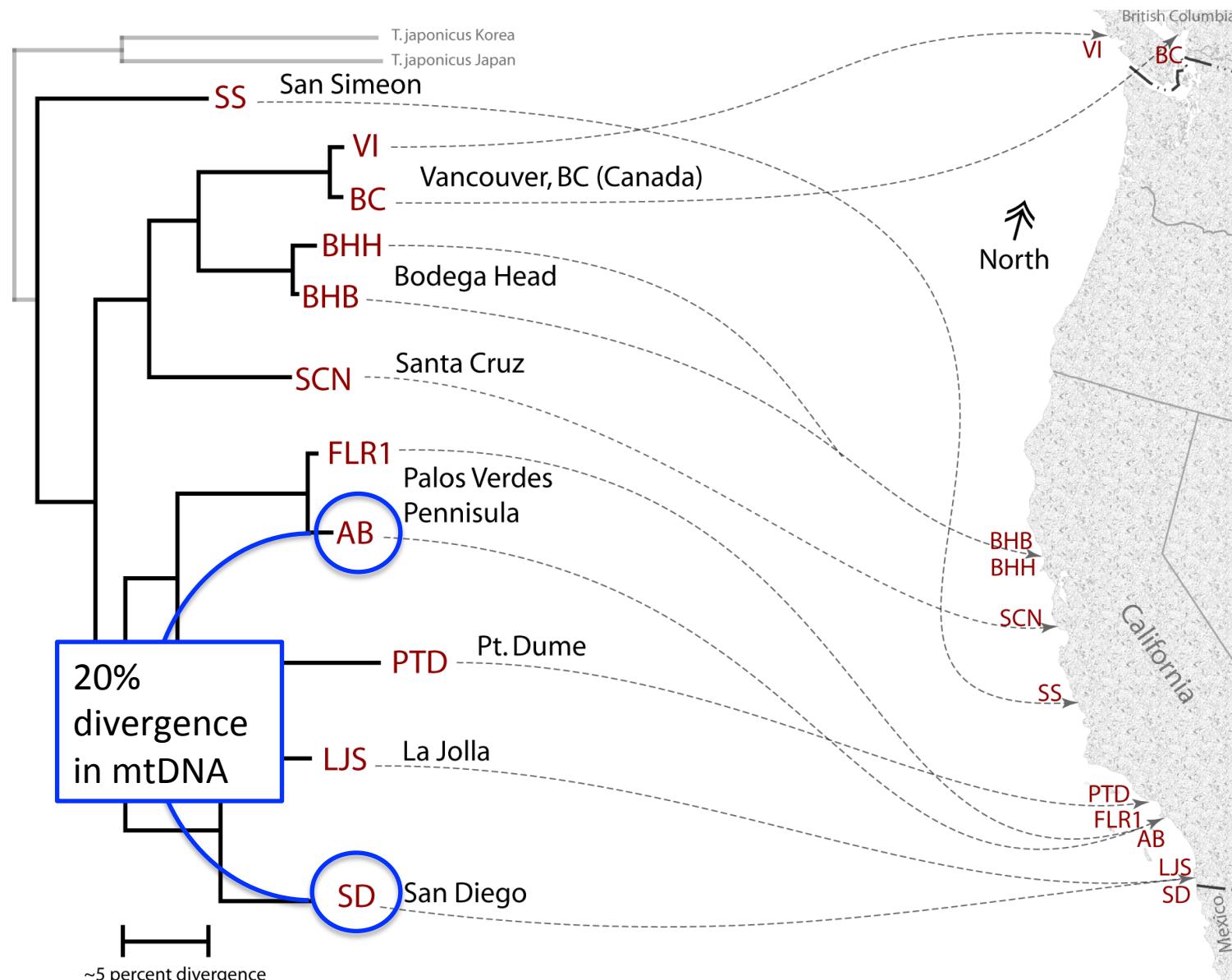


Photo R. Burton

High levels of divergence between populations



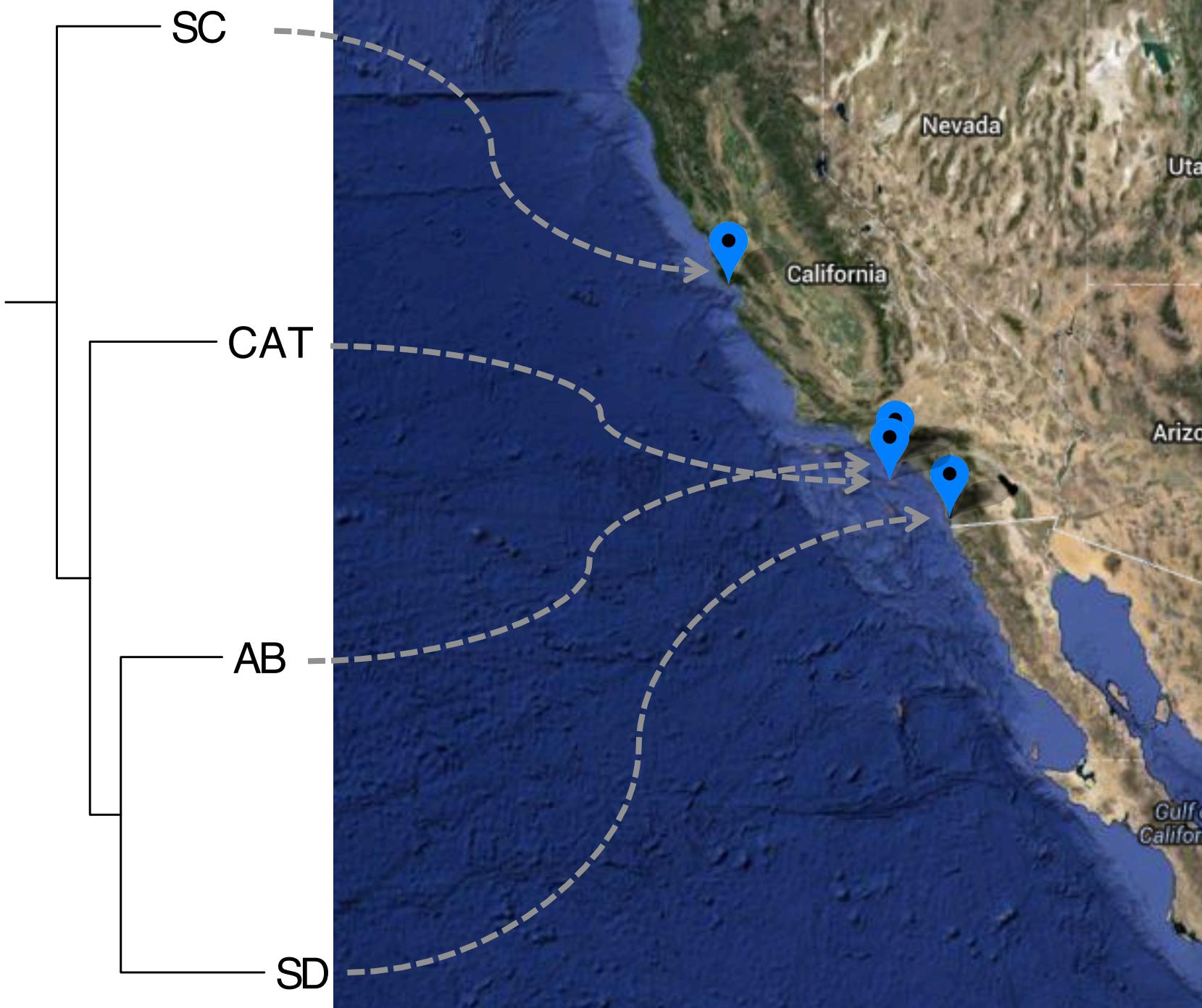
High levels of divergence between populations



Tigriopus californicus

- Does well in the lab
- Small genome size (~240Mb)
- Short generation time
- No sex chromosomes
 - polygenic sex determination





Genome-wide scan for hybrid incompatibilities

- Sequenced pools of 300 F₂ hybrid individuals:
 - SDf x ABm nauplii (from egg sacs)
 - SDf x ABm adults
 - CATf x ABm adults
 - SCf x ABm adults

Creating alternative population references

SD genome assembly



Map other populations' reads

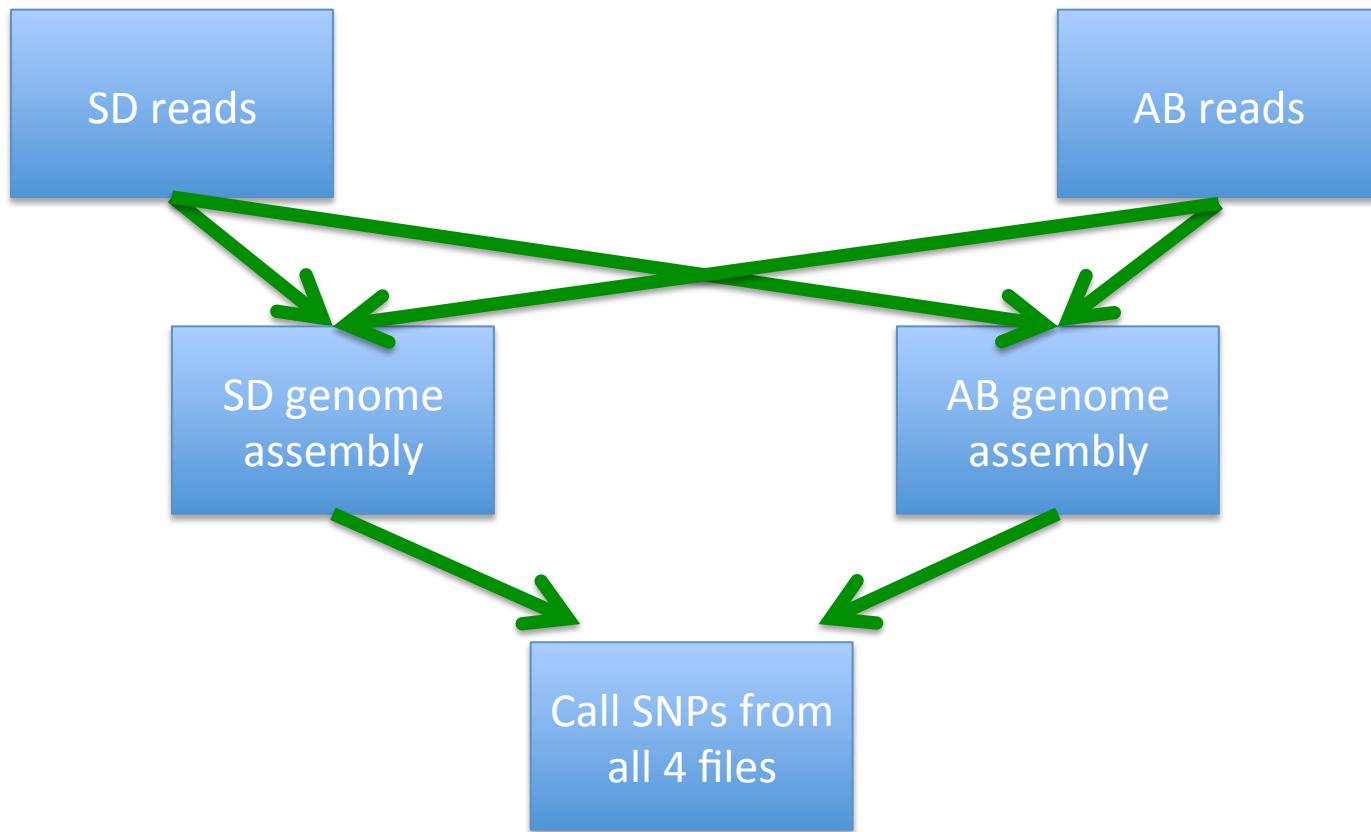


Create other references

```
bwa mem ref.fa trimmed_reads_1.fq trimmed_reads_2.fq > out.sam
```

```
samtools mpileup -uf ref.fa aln.bam | bcftools view -cg - | vcfutils.pl vcf2fq > cns.fq
```

Calling SNPs between populations

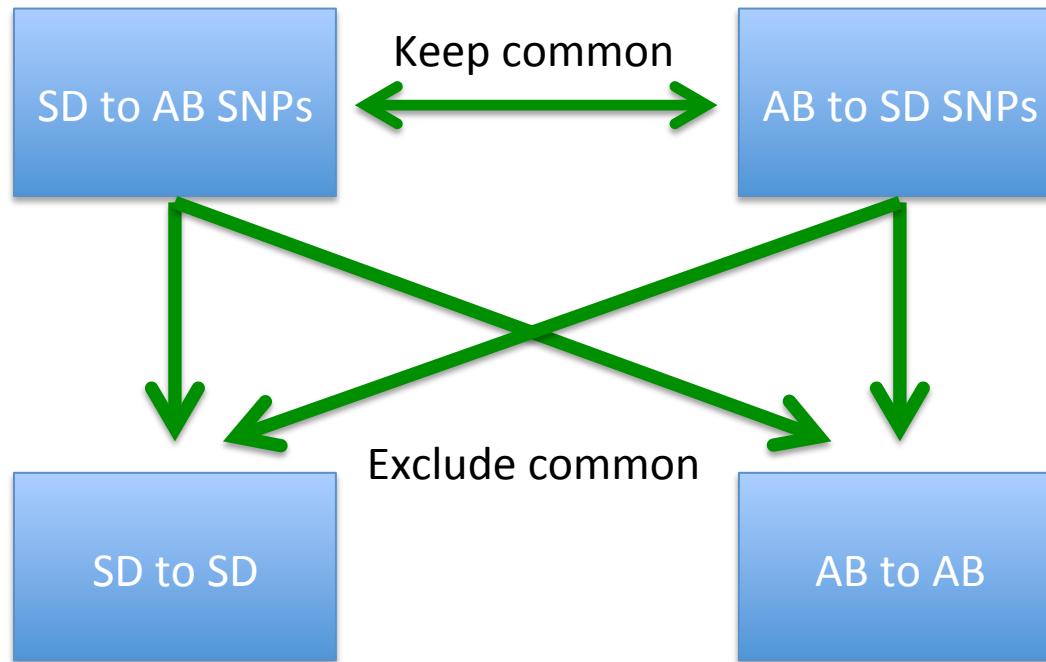


```
samtools mpileup -uf ref.fa aln1.bam | bcftools view -bvcg - > var.raw.bcf
```

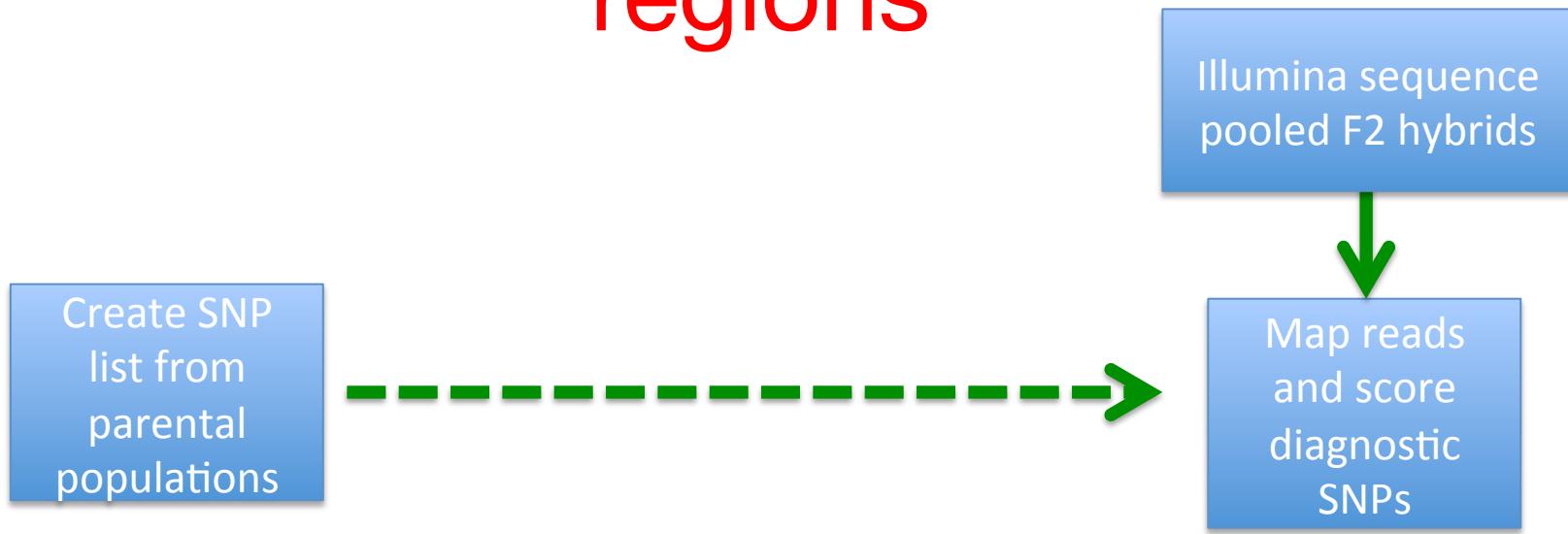
```
bcftools view var.raw.bcf | vcfutils.pl varFilter -D100 > var.flt.vcf
```

Use 'awk' to remove unwanted info

Calling SNPs between populations



Determination of hybrid inviability regions



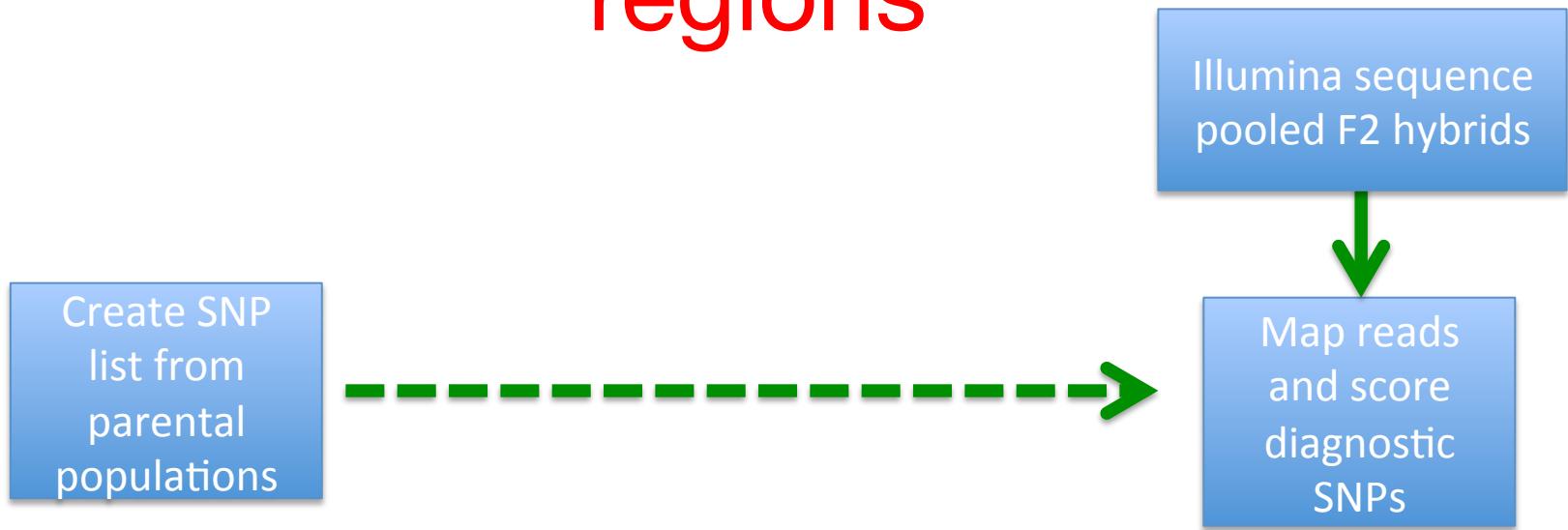
```
samtools mpileup -Bf ref.fa hybrid_aln.bam > out.pileup
```

```
perl popoolation2_1201/mpileup2sync.pl -input out.pileup2 --fastq-type sanger -output out.sync
```

Sync file from PoPoolation 2

scaffold_68	521786	T	0:41:46:0:0:0
scaffold_68	521787	G	0:0:0:89:0:0
scaffold_68	521788	T	0:87:0:0:0:0
scaffold_68	521789	G	0:0:0:86:0:0
scaffold_68	521790	T	0:90:0:0:0:0
scaffold_68	521791	G	0:0:0:89:0:0
scaffold_68	521792	C	0:0:88:0:0:0
scaffold_68	521793	C	0:0:89:0:0:0
scaffold_68	521794	G	0:46:0:41:0:0

Determination of hybrid inviability regions



```
samtools mpileup -Bf ref.fa hybrid_aln.bam > out.pileup
```

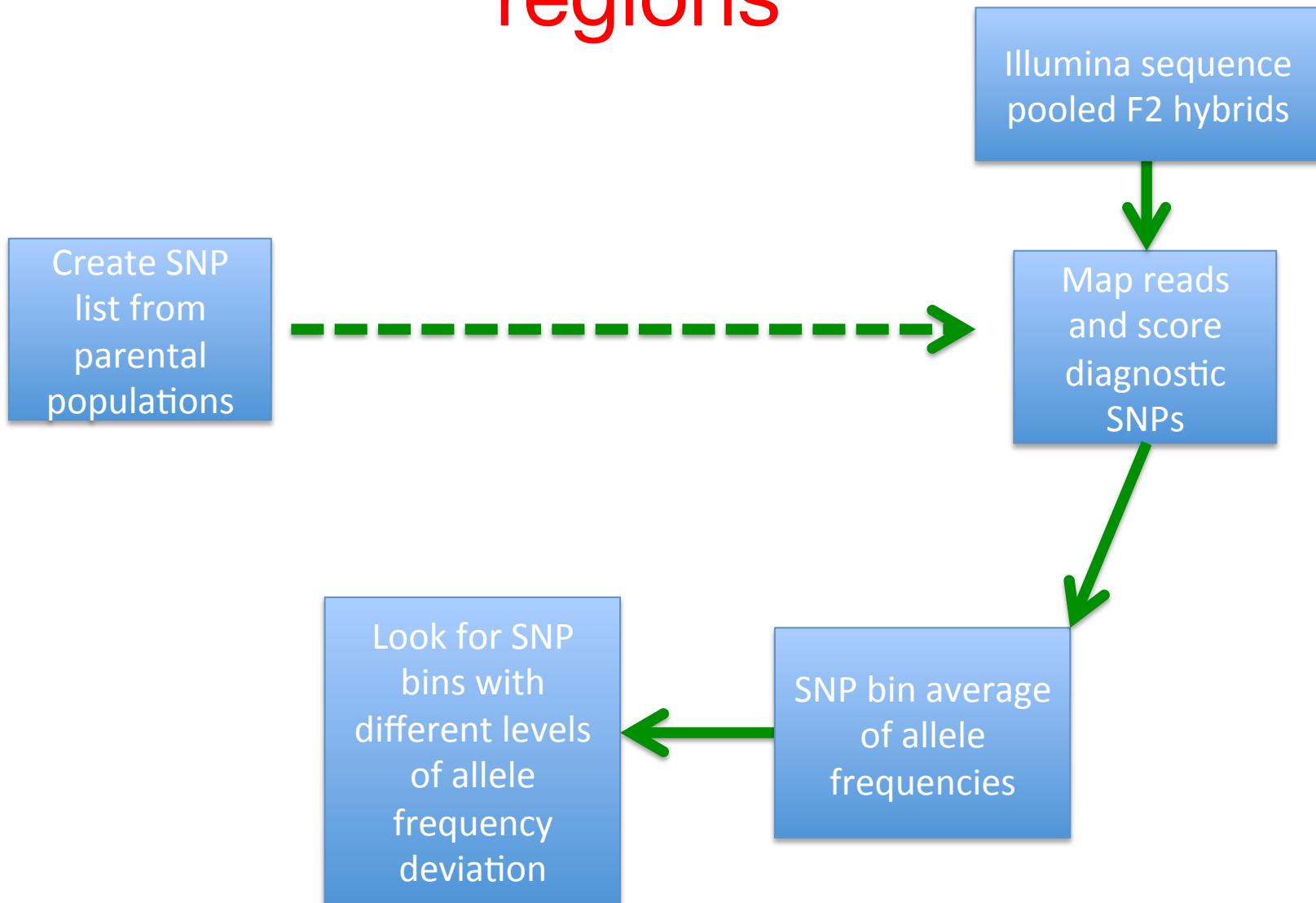
```
popoolation2_1201/mpileup2sync.pl –input out.pileup2 --fastq-type sanger –output out.sync
```

```
popoolation2_1201/snp-frequency-diff.pl --input out.sync --output-prefix out --min-count 4  
--min-coverage 10 --max-coverage 500
```

rc file from PoPoolation 2

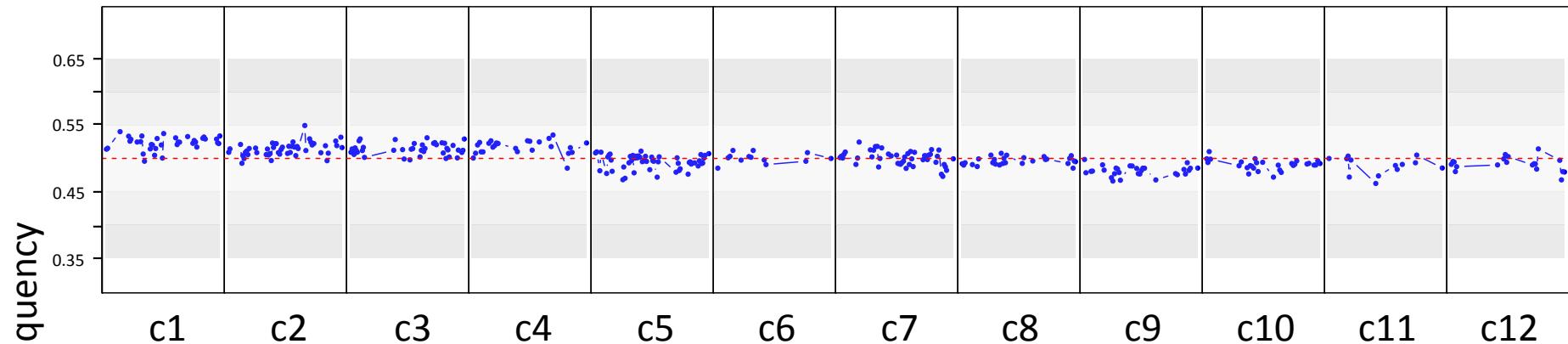
##chr	pos	rc	allele count	allele states	deletion sum	snp_type	major allele	minor allele	maa_1	mia_1
scaffold_15	576	C	2	C/T	0	pop	C	T	76/114	38/114
scaffold_15	601	A	2	A/G	0	pop	A	G	74/116	42/116
scaffold_15	617	C	2	C/T	0	pop	C	T	89/125	36/125
scaffold_15	624	T	2	T/G	0	pop	T	G	97/134	37/134
scaffold_15	636	T	2	T/A	0	pop	T	A	101/134	33/134
scaffold_15	648	A	2	A/G	0	pop	A	G	106/139	33/139
scaffold_15	659	T	2	T/A	0	pop	T	A	100/142	42/142
scaffold_15	677	G	2	G/A	0	pop	G	A	89/124	35/124
scaffold_15	704	A	2	A/G	0	pop	A	G	101/139	38/139
scaffold_15	707	G	2	G/T	0	pop	G	T	99/137	38/137

Determination of hybrid inviability regions

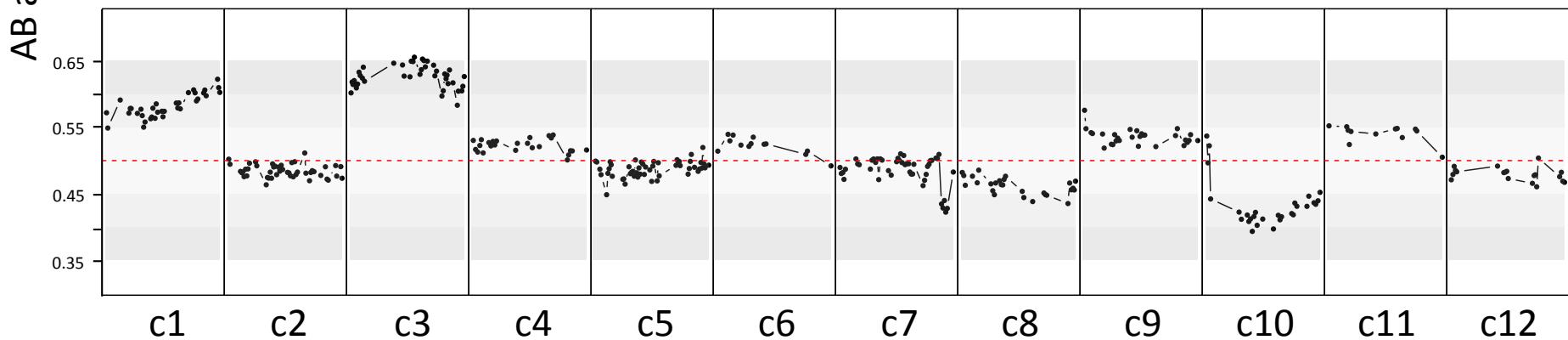


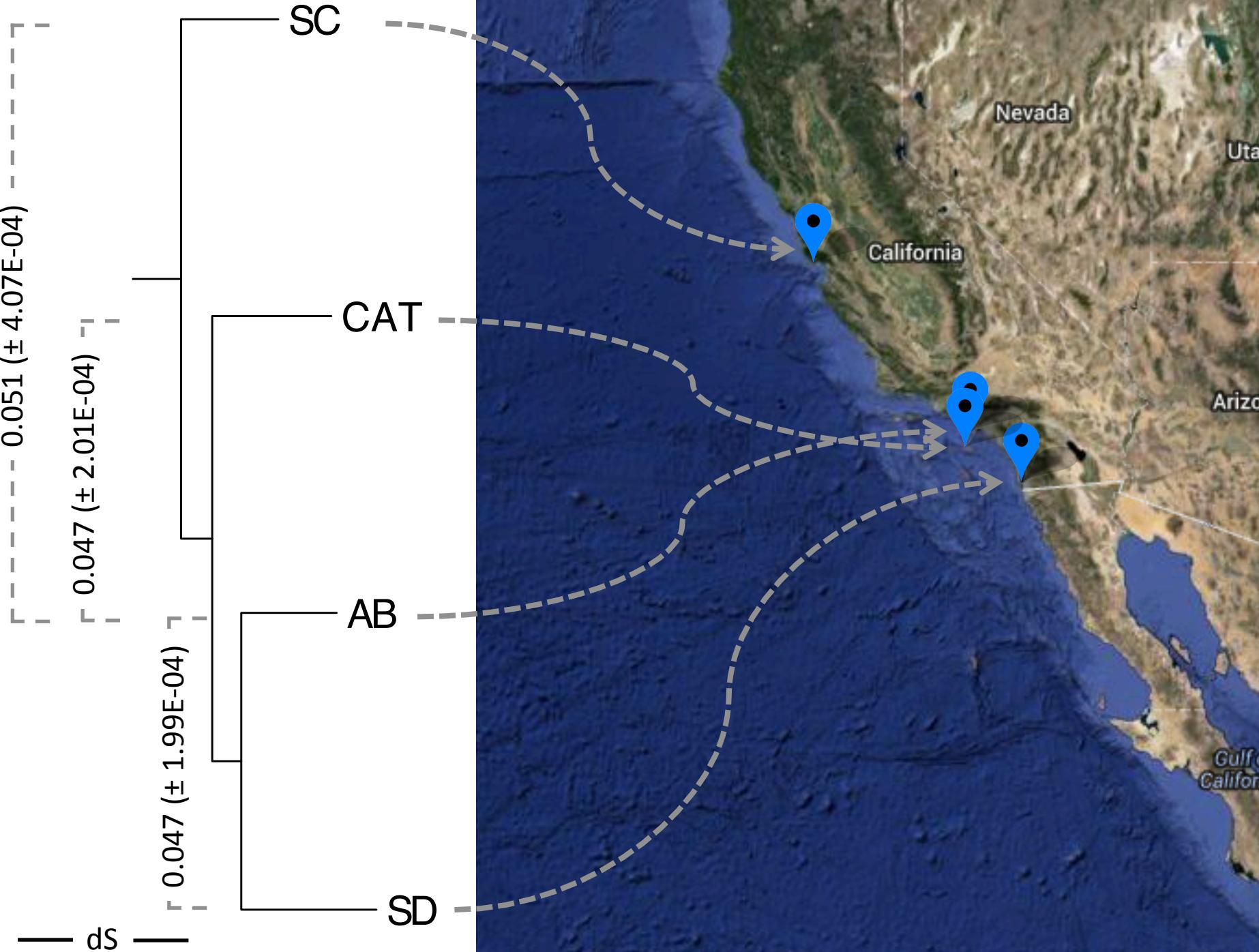
SD x AB F₂ hybrids

Nauplii

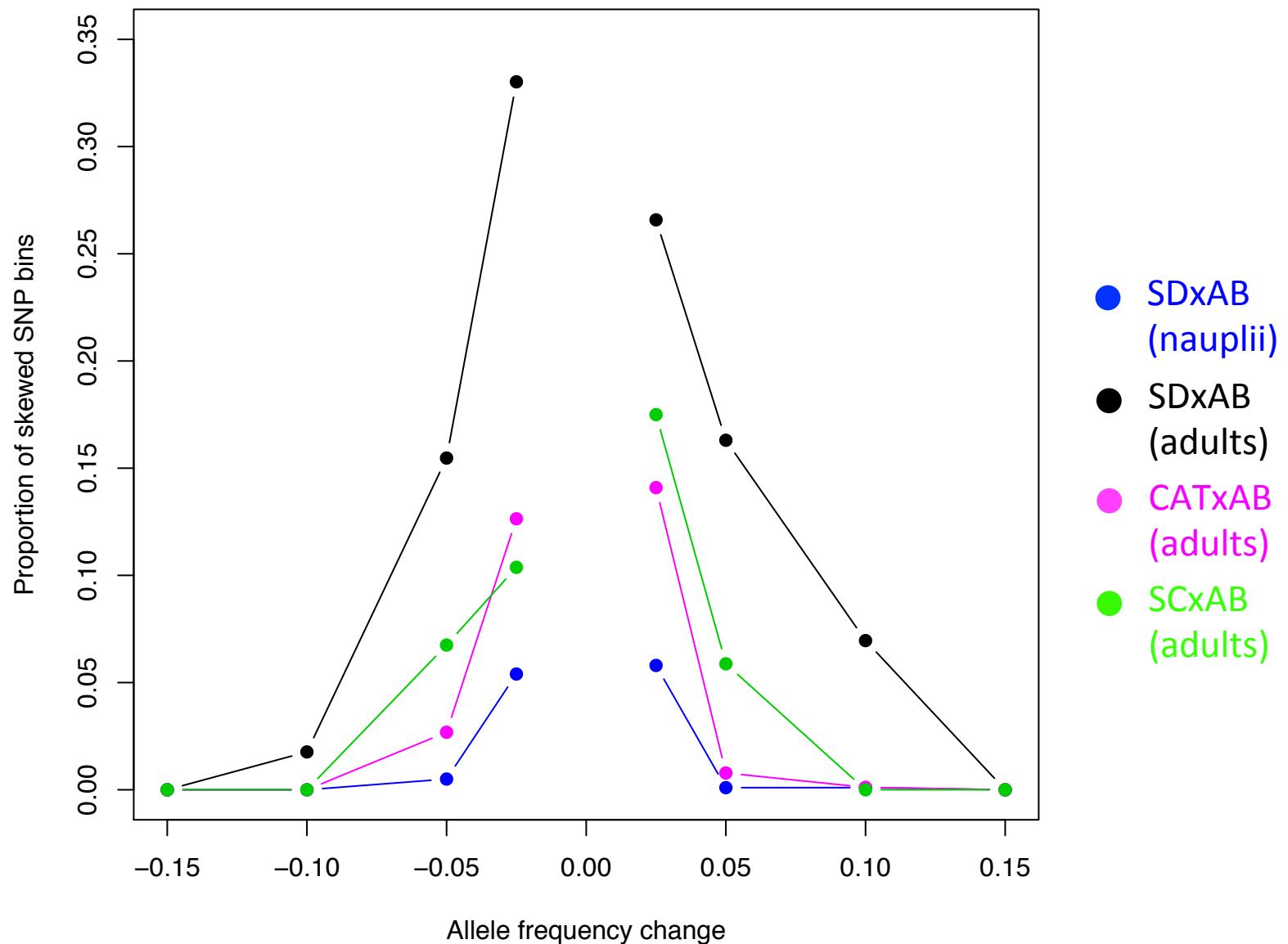


Adults

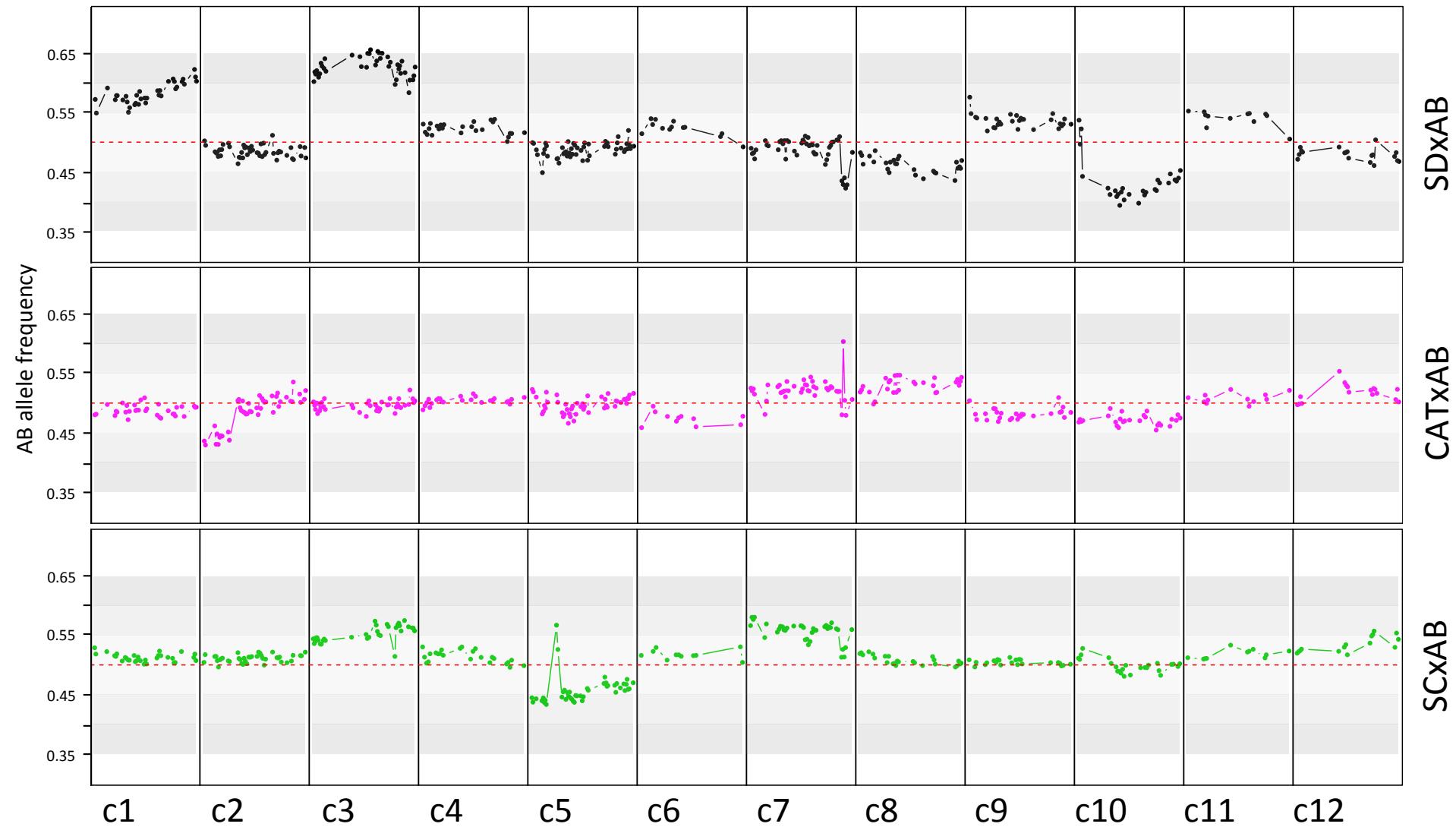




Proportion of skewed SNP bins



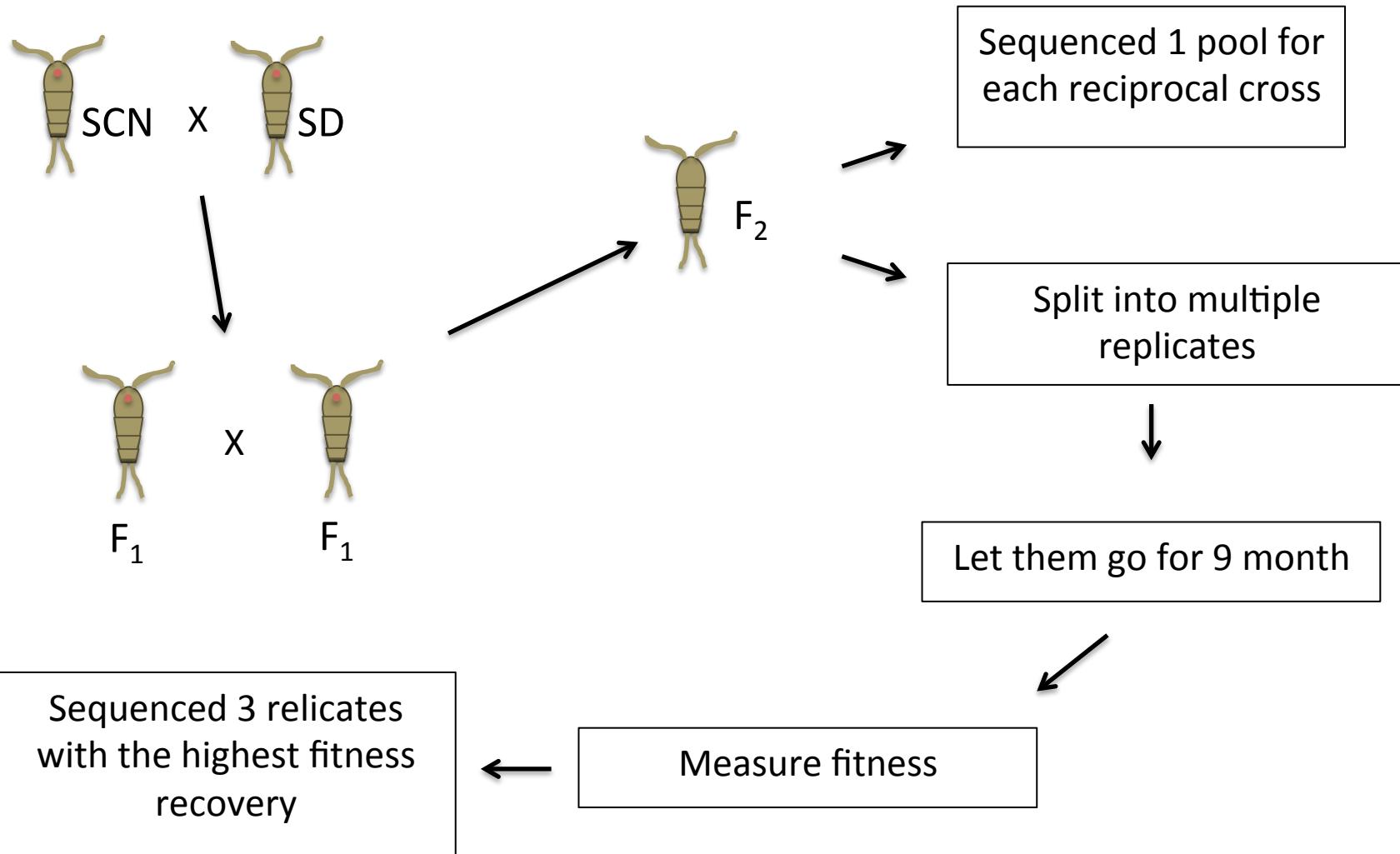
Genome-wide effects from hybrid inviability



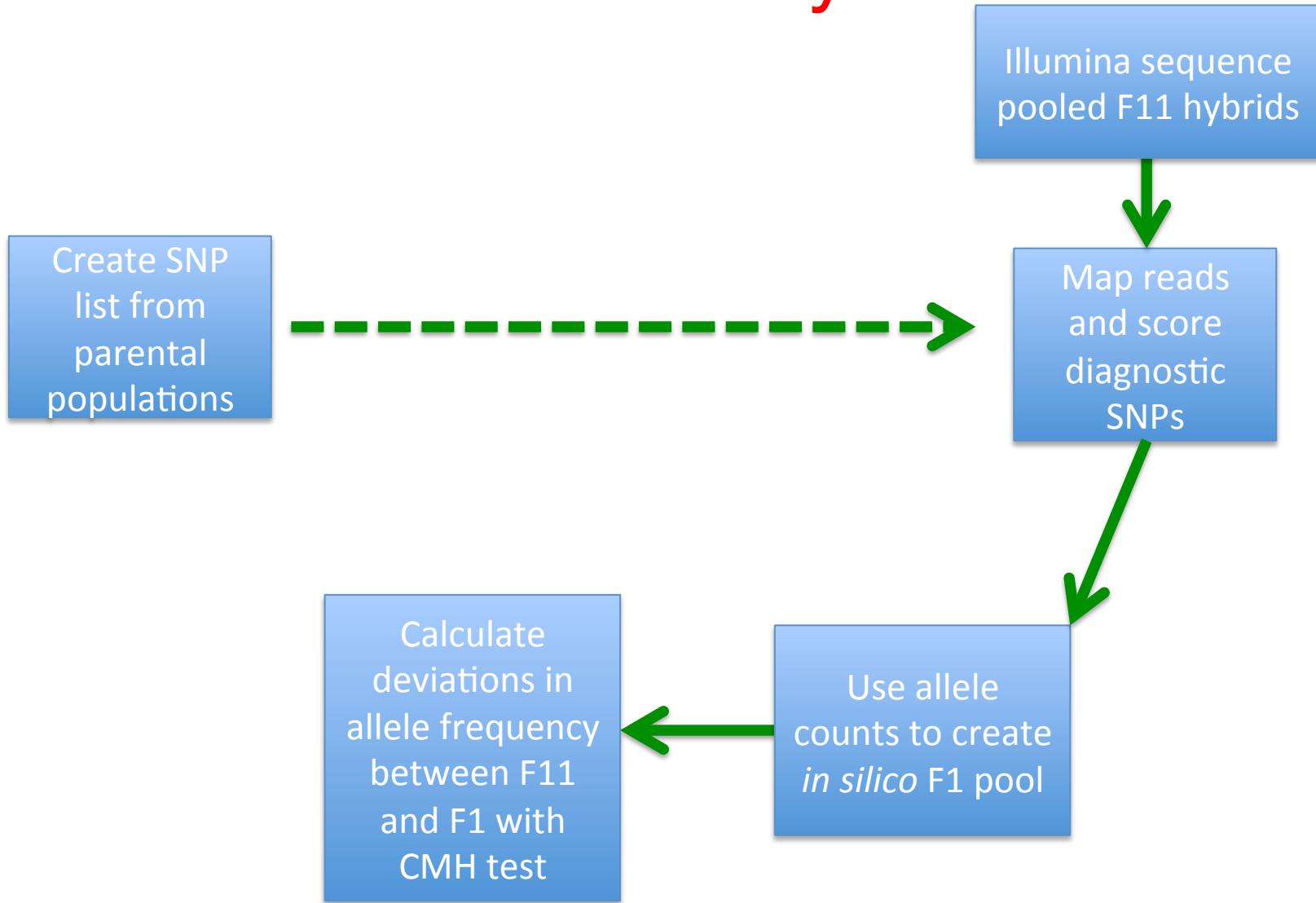
Conclusions

- SDxAB may have accumulated more incompatibilities than the other crosses
- Or in comparisons between crosses with similar levels of divergence the number of incompatibilities is less important
- Strength and positioning of incompatibility may determine strength of isolation

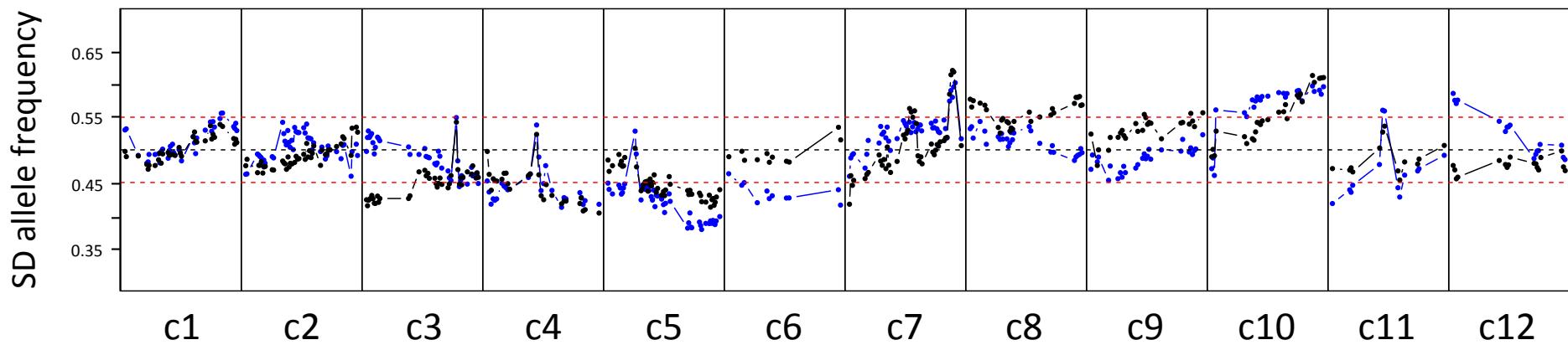
Evolve and resequence for hybrid inviability



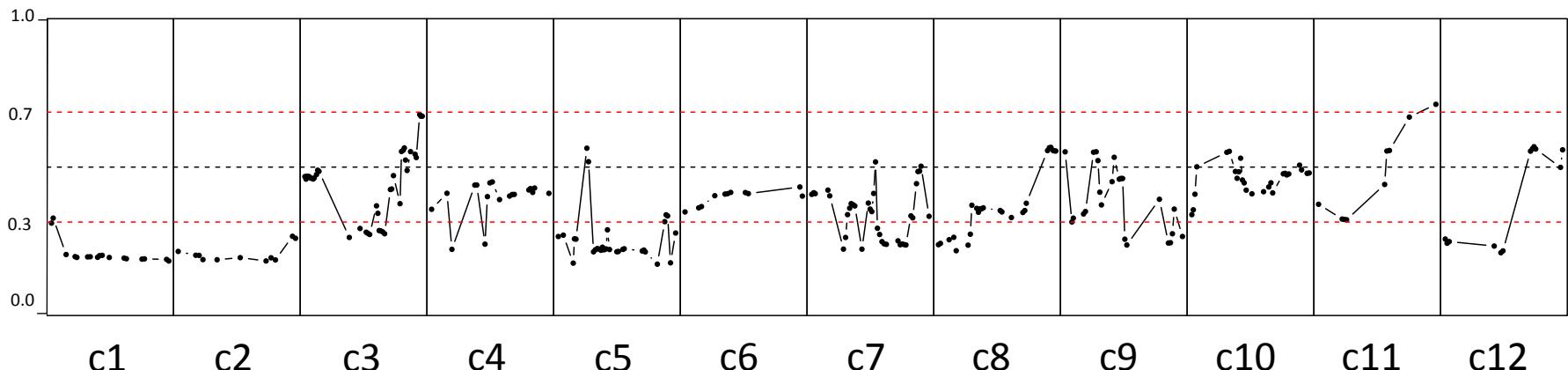
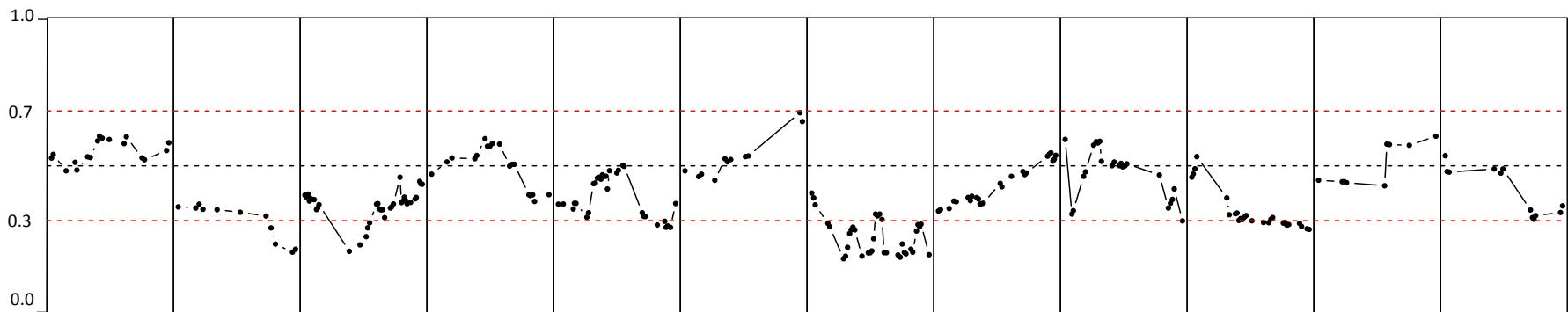
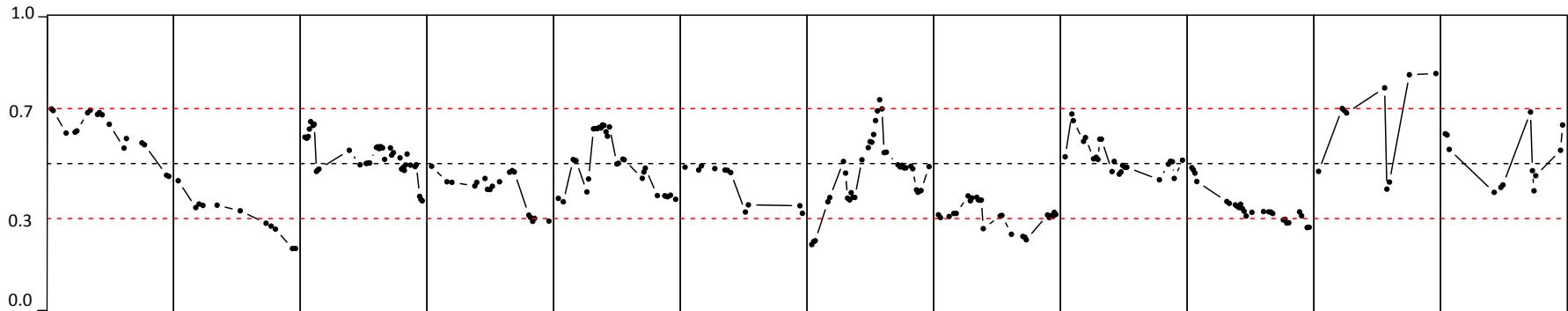
Evolve and resequence for hybrid inviability



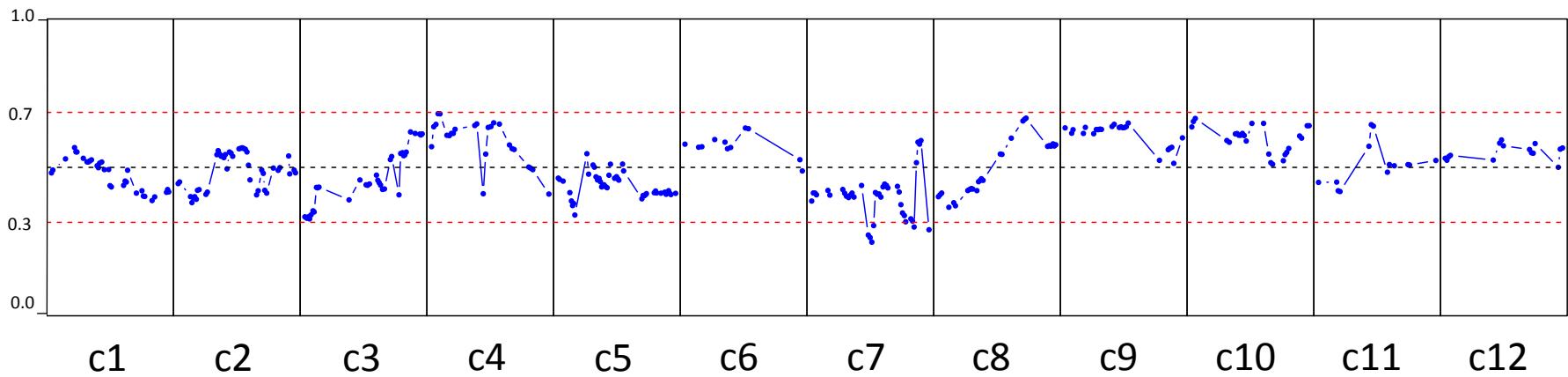
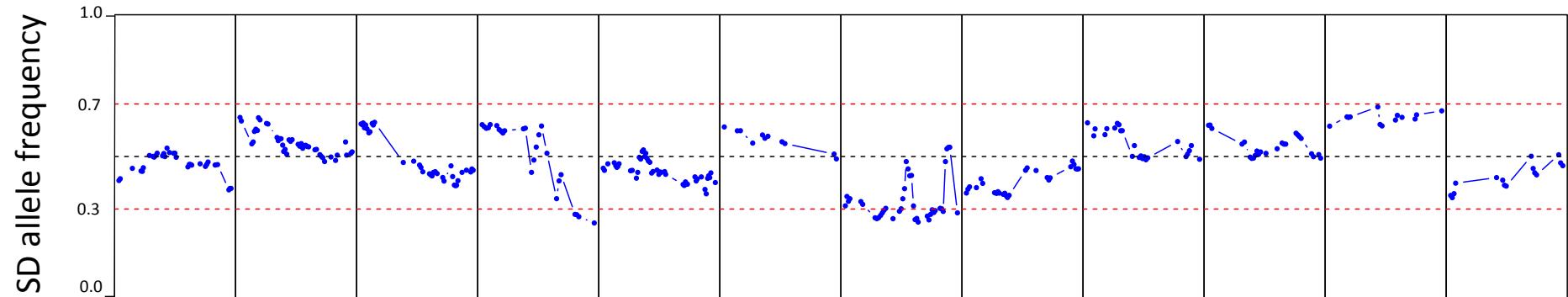
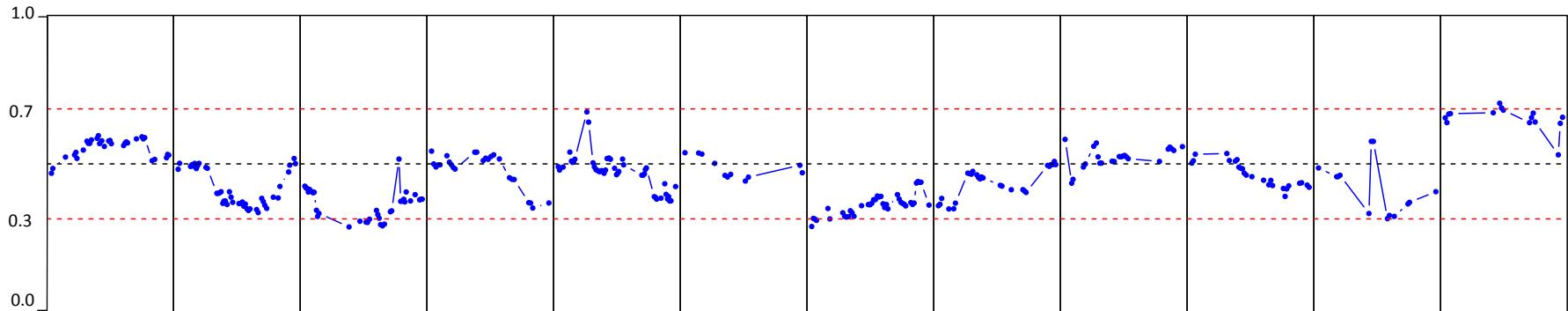
Reciprocal SD x SCN F2 hybrids



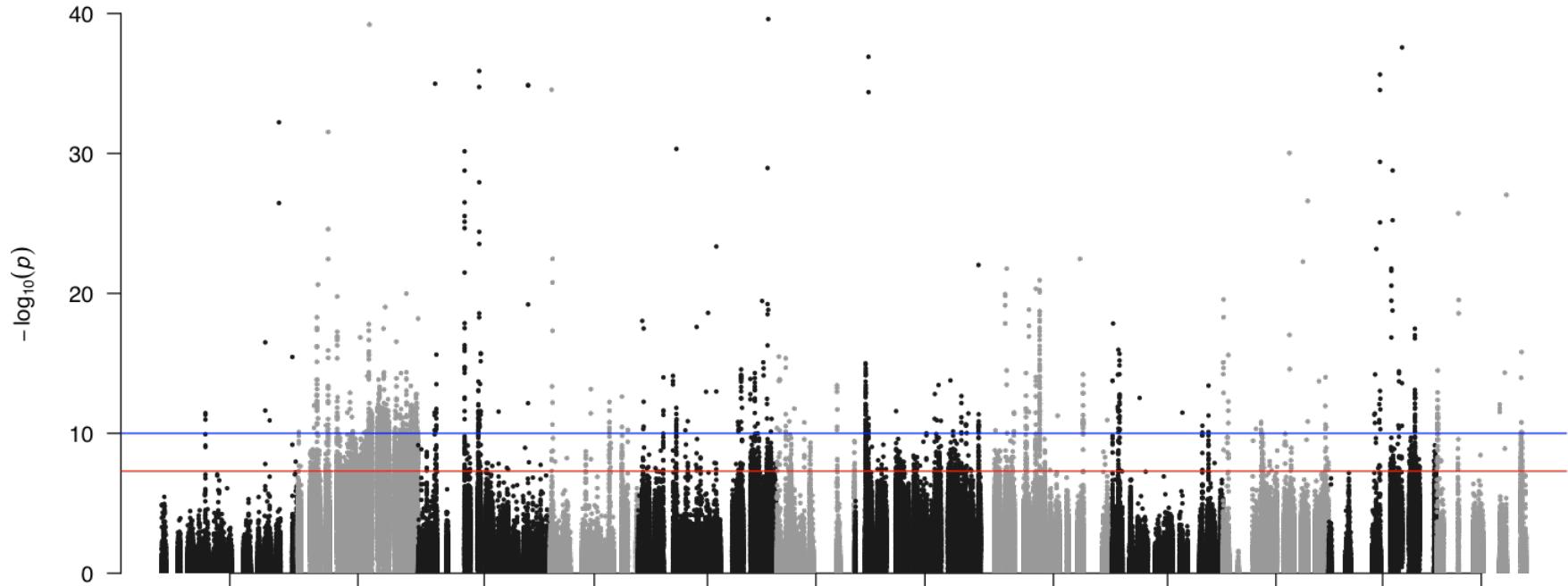
SCNf x SDm F11 hybrids



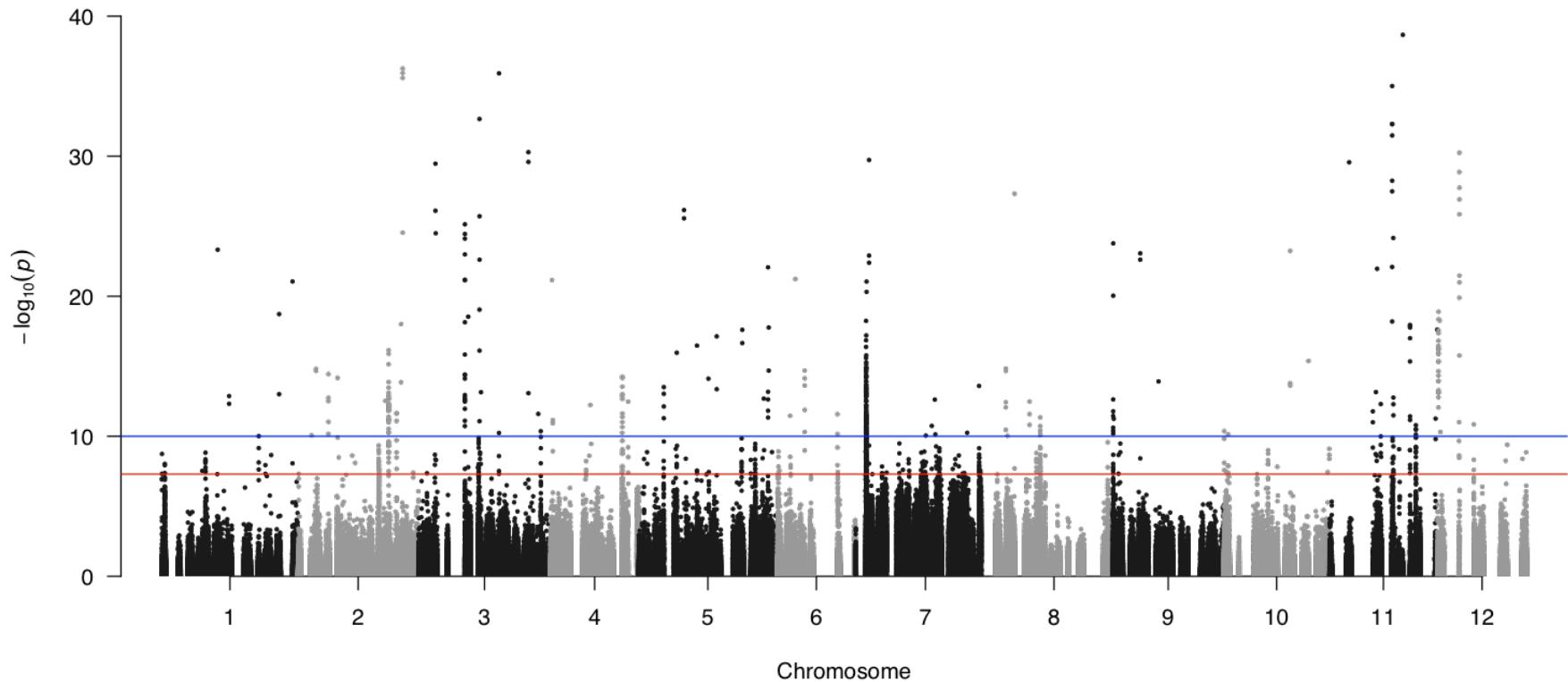
SDf x SCN_m F11 hybrids



SCNf x SDm



SDf x SCNm



Thank you!