

QTL mapping in MAGIC populations

Part 1

Karl Broman

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`kbroman.org`

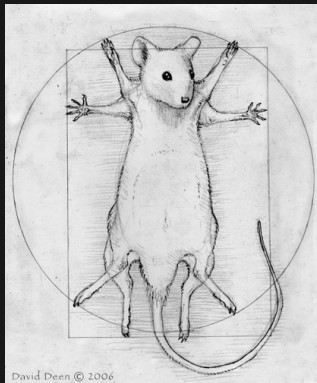
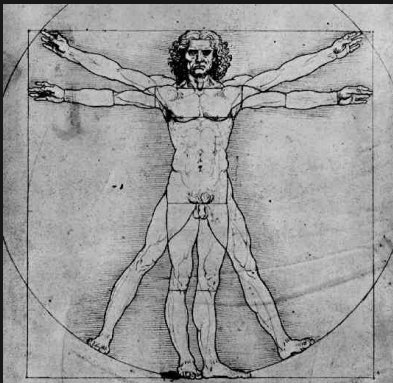
`github.com/kbroman`

`@kwbroman`

Slides: `kbroman.org/Talk_MAGIC2021`

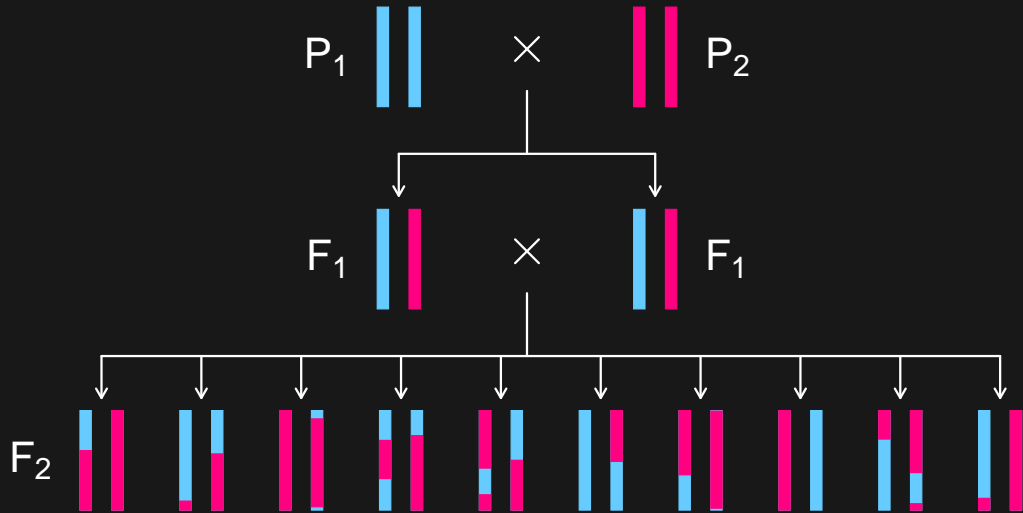




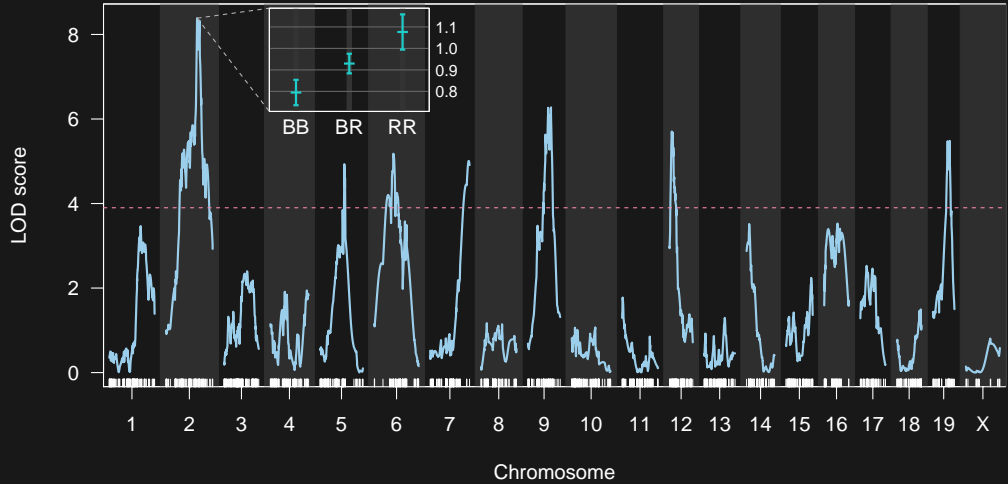


daviddeen.com

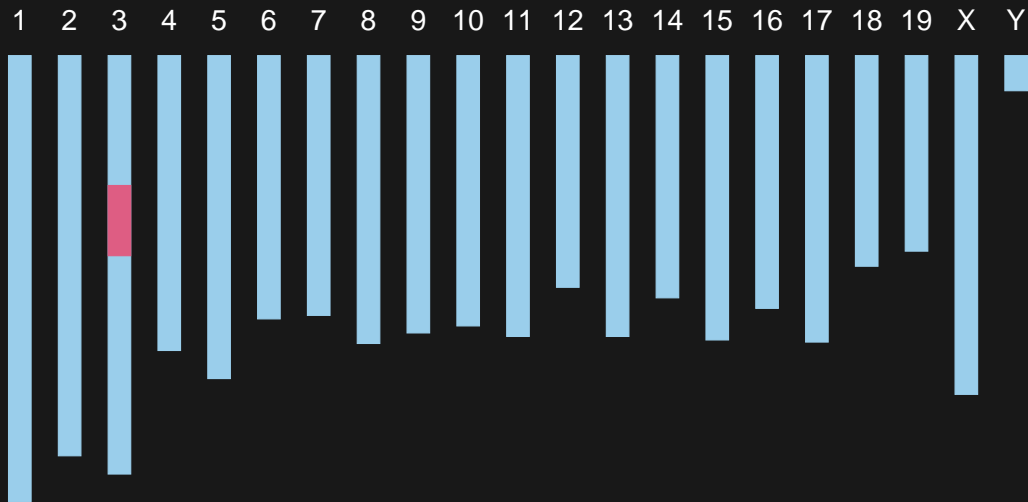
Intercross



QTL mapping



Congenic line/NIL



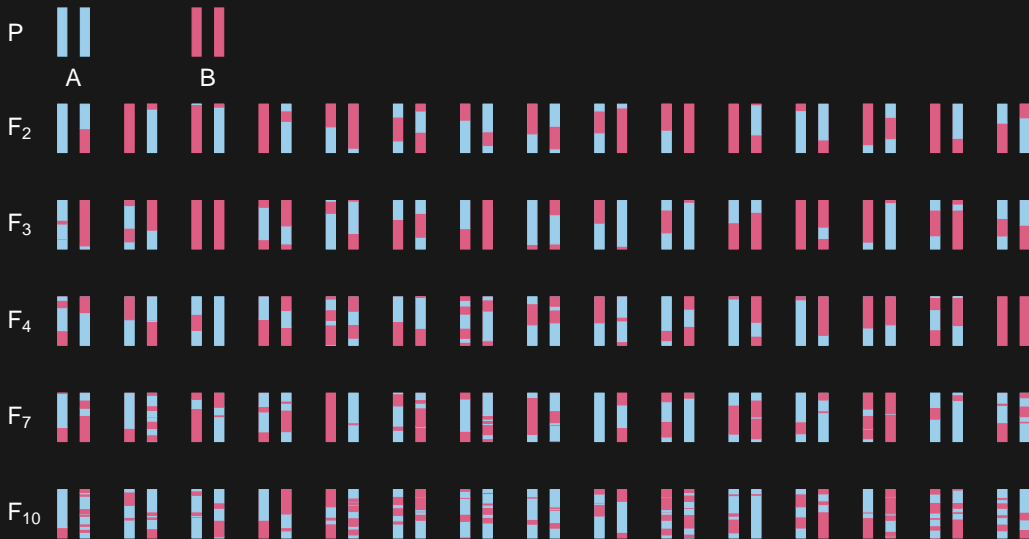
Improving precision

- ▶ more recombinations
- ▶ more individuals
- ▶ more precise phenotype
- ▶ lower-level phenotypes
 - transcripts, proteins, metabolites

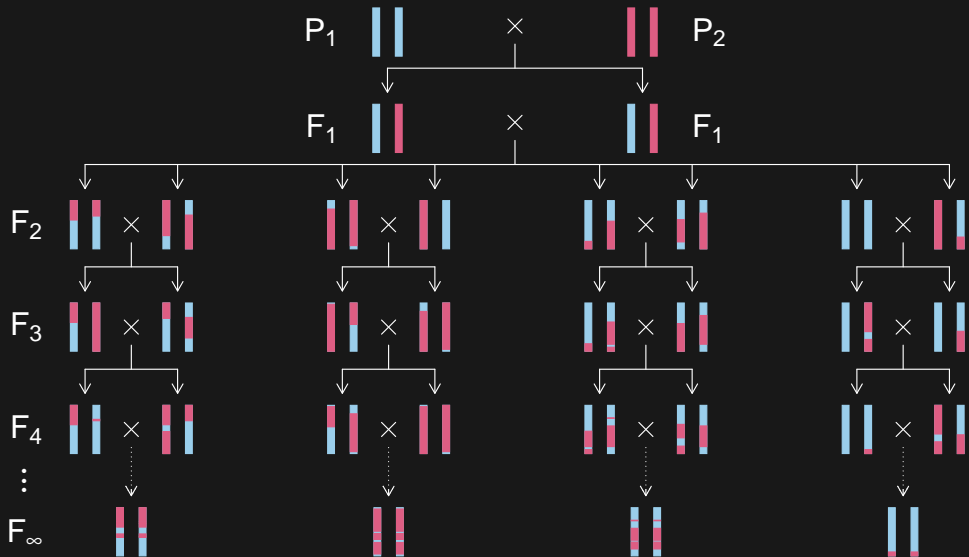
Genome-scale phenotypes



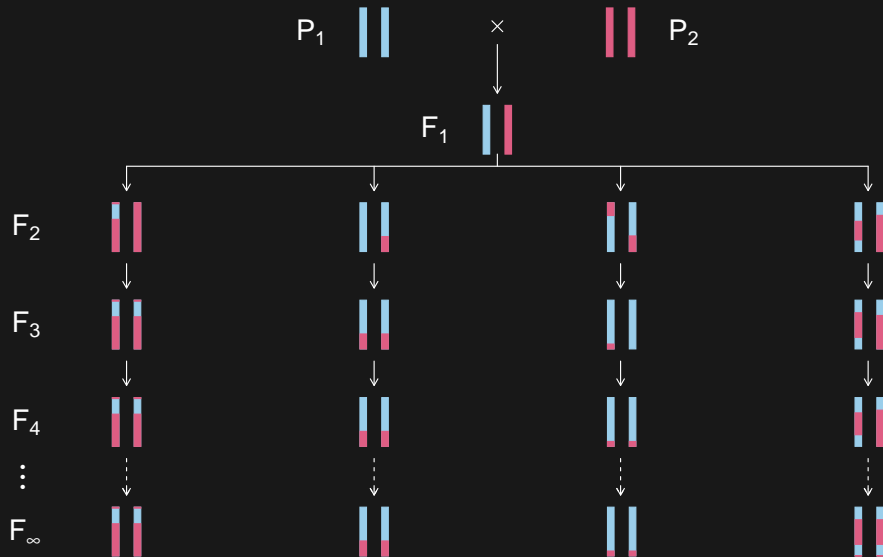
Advanced intercross lines



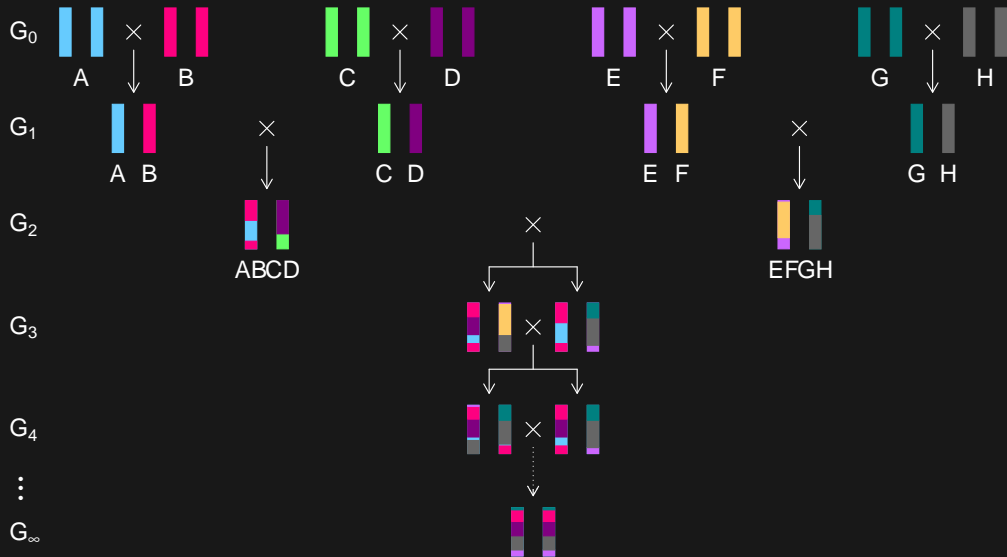
Recombinant inbred lines



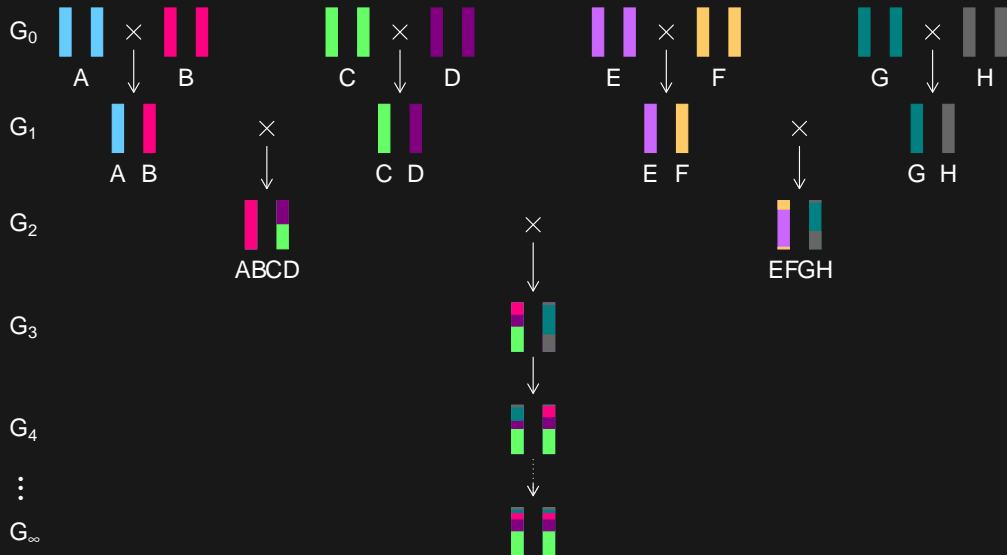
Recombinant inbred lines



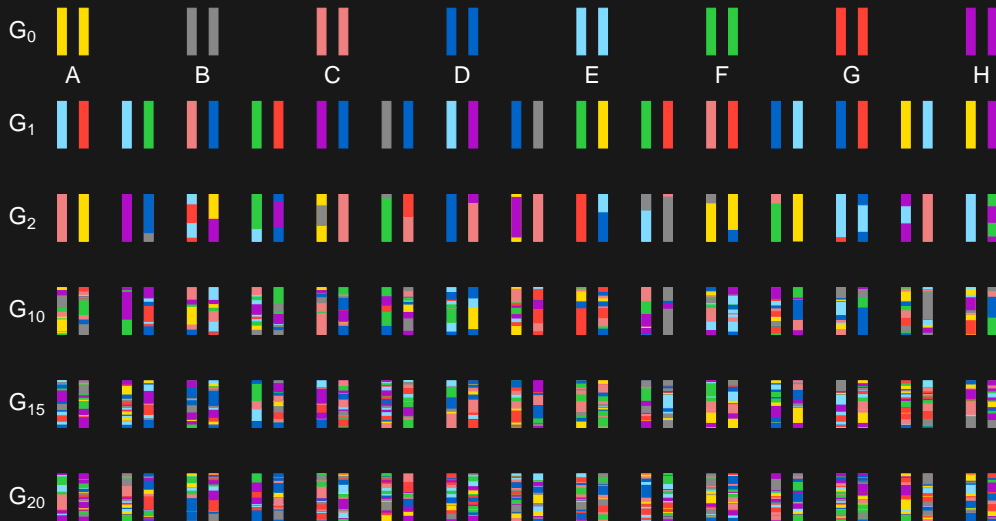
Collaborative Cross



MAGIC



Heterogeneous stock



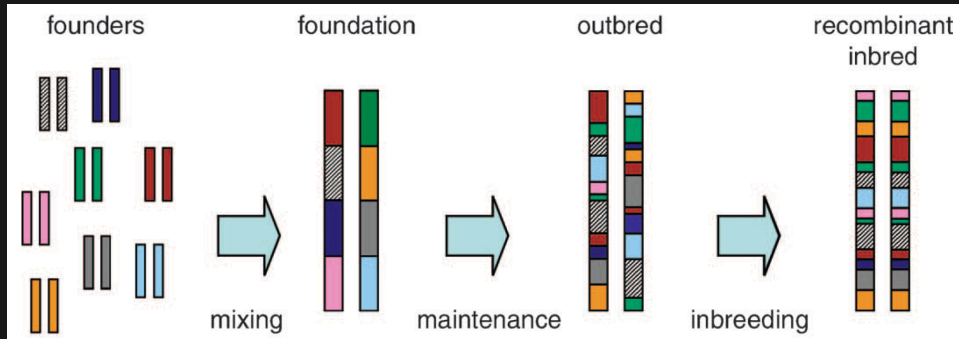
MAGIC is magic

- ▶ Genetic diversity
- ▶ High-precision mapping
- ▶ Predictable linkage disequilibrium
- ▶ No rare alleles
- ▶ Phenotype replicates to reduce individual variation
- ▶ Pool phenotypes from multiple labs, environments, treatments
- ▶ Genotype once

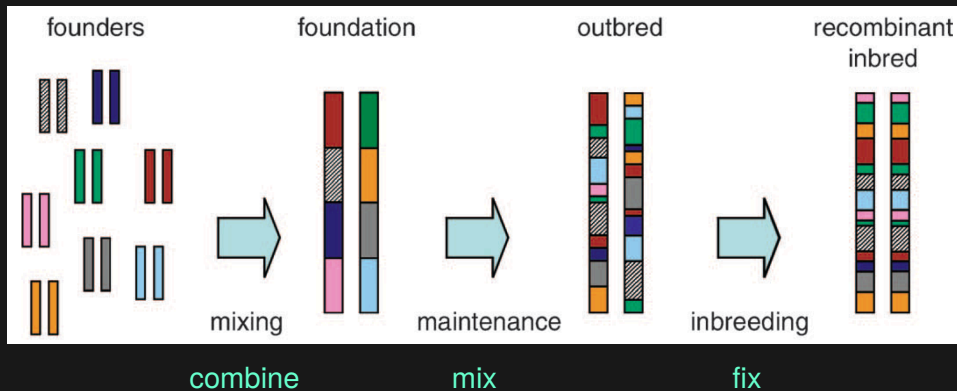
MAGIC is magic

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- ▶ No rare alleles
- ▶ Phenotype replicates to reduce individual variation
- ▶ Pool phenotypes from multiple labs, environments, treatments
- ▶ Genotype once
- ▶ Cool name

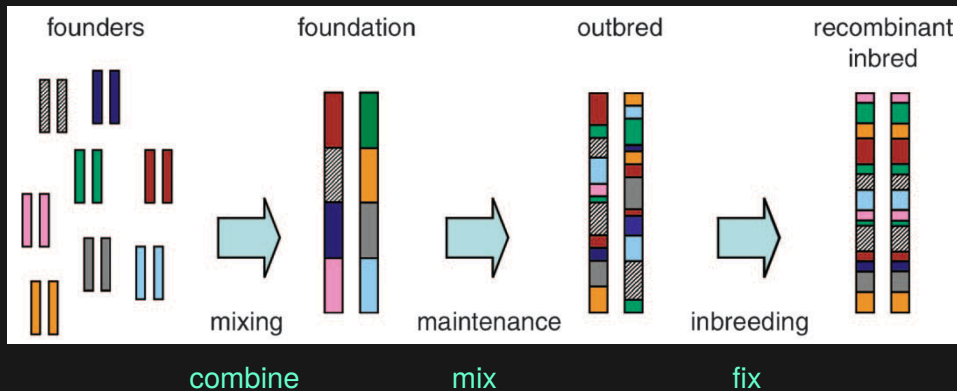
MAGIC lines



MAGIC lines

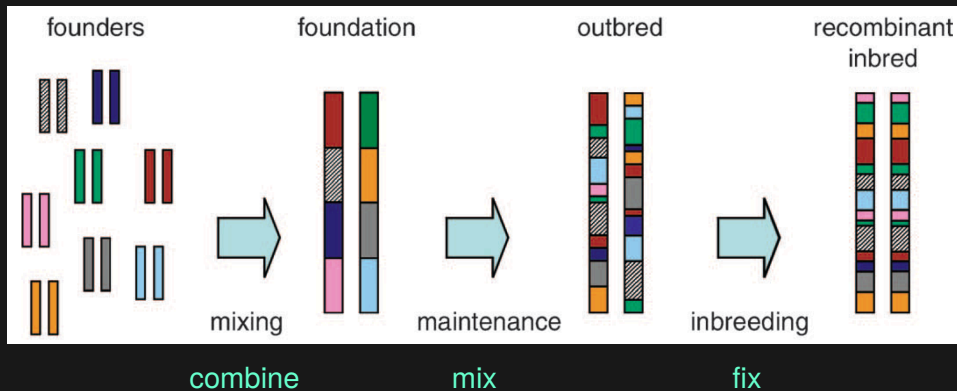


MAGIC lines



How many?

MAGIC lines

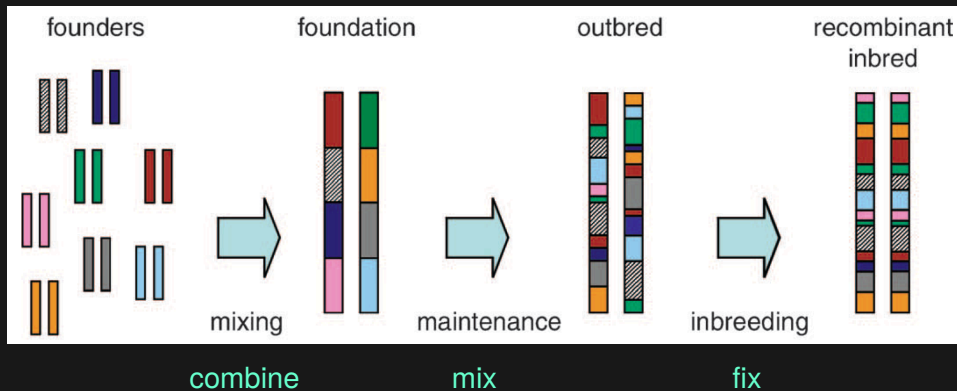


How many?

Which?

Valdar et al., Genetics 172:1783, 2006

MAGIC lines



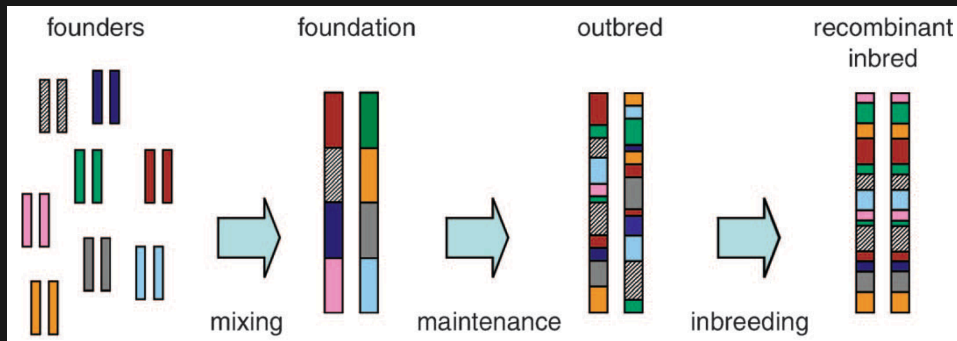
How many?

How long?

Which?

Valdar et al., Genetics 172:1783, 2006

MAGIC lines



combine

mix

fix

How many?

How long?

How?

Which?

Valdar et al., Genetics 172:1783, 2006

The goal

Identify QTL

- ▶ Power
- ▶ Mapping precision

The goal

Identify QTG

- ▶ Power
- ▶ Mapping precision

The goal

Identify QT**G**

- ▶ Power
- ▶ Mapping precision
- ▶ Estimate QTL allele frequencies

Principles

- ▶ Avoid population structure
- ▶ Tradeoff between *power for de novo discovery* and *mapping precision*
- ▶ More QTL to find \Rightarrow more QTL getting in the way?
- ▶ More QTL alleles \Rightarrow less information about each
- ▶ Are QTL alleles common or rare?

How many founders?

More

- ▶ More general use
- ▶ More QTL
- ▶ Greater precision
- ▶ Estimate allele frequencies
- ▶ Haplotype analysis in founders

Fewer

- ▶ Lower residual variance
- ▶ Greater power for a particular QTL?
- ▶ Better power for epistasis
- ▶ Rare alleles are less rare

Which founders?

- ▶ Diverse
- ▶ Interesting
- ▶ No breeding problems
- ▶ Balanced: star phylogeny

How much mixing?

- ▶ More mixing \Rightarrow Greater mapping precision
- ▶ ...but lower power for *de novo* mapping
- ▶ Potential for population structure, missing alleles
- ▶ Random mating or curated mating?
- ▶ Start with many random cross directions?

Selfing or DH?

- ▶ Inbreeding gives added recombination
- ▶ But not so much as at the mixing stage
- ▶ If doubled haploids are feasible, use them

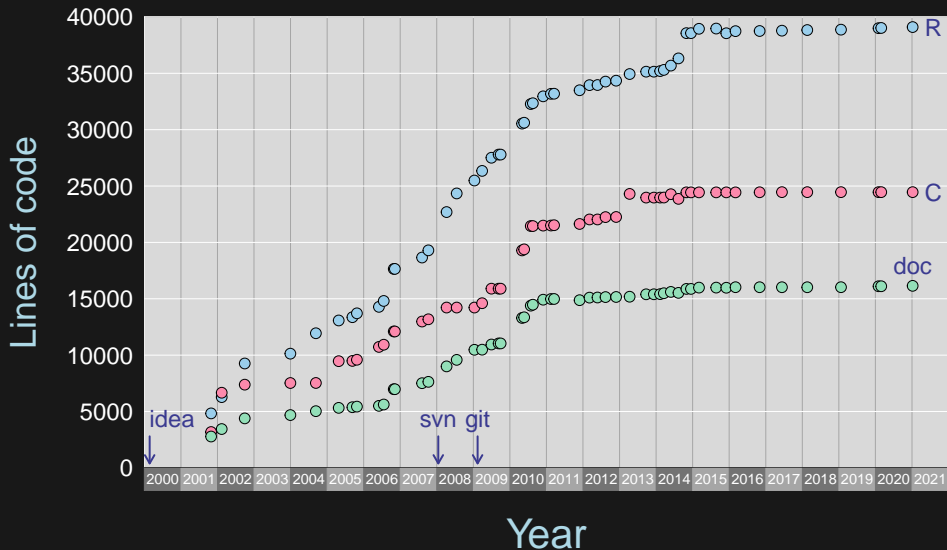
Sharing is also key

- ▶ The greatest power of MAGIC comes from sharing
Pooling data, exploring multiple environments/treatments
- ▶ Common software needs
Analysis software, database infrastructure
- ▶ Many students need to learn the same stuff
Joint training opportunities

Summary

- ▶ How many founders?
 - Tradeoff between **diversity** and information about **particular alleles**
- ▶ Which founders?
 - Diverse, interesting, no breeding problems, star phylogeny
- ▶ How long to mix?
 - Tradeoff between **power** and **precision**
- ▶ How to fix?
 - Doubled haploids are great if feasible
- ▶ Let's share!
 - Lines, data, software, training

21 years of R/qtI



R/qtl cross types

- ▶ backcross, doubled haploids, haploid
- ▶ intercross
- ▶ 2-way RIL by selfing or sibling mating
- ▶ phase-known 4-way cross



R/qtl2 cross types

- ▶ backcross, doubled haploids, haploid
- ▶ intercross
- ▶ 2-, 4-, 8-, 16-way RIL by selfing
- ▶ 2-, 4-, 8-way RIL by sibling mating
- ▶ 2-, 3-, 8-way advanced intercross
- ▶ 6- and 19-way MAGIC
- ▶ Diversity Outbred (DO) mice
- ▶ F_1 of DO \times inbred
- ▶ general RIL or AIL

Data files

	A	B	C	D	E	F	G
1	id	bolting_days	seed_weight	seed_area	ttl_seedspruit	branches	height
2	MAGIC.1	15.33	17.15	0.64	45.11	10.5	NA
3	MAGIC.2	22	22.71	0.75	49.11	4.33	42.33
4	MAGIC.3	23	21.03	0.68	57	4.67	50
5	MAGIC.4	18.67	22.45	0.74	54.33	6.33	NA
6	MAGIC.5	18.67	25.36	0.82	38.33	5.67	42.25
7	MAGIC.6	25	21.53	0.71	52	4.33	NA
8	MAGIC.7	15.33	20.92	0.71	39	4	37.35
9	MAGIC.8	14.33	24.2	0.79	50.56	7.33	43.23
10	MAGIC.9	16.33	18.86	0.63	75.78	5.33	42.4
11	MAGIC.10	30	30.46	0.91	56.33	3	48.5
12	MAGIC.11	14	20.58	0.66	41.56	5.67	36.25
13	MAGIC.12	21.33	19.05	0.67	52.33	4.67	49.35
14	MAGIC.14	18.67	21.14	0.72	49.78	6	48.5
15	MAGIC.15	15	21.71	0.73	49	6.33	41.9
16	MAGIC.16	18.33	21.37	0.71	66.67	3.33	18.17

Data files

	A	B	C	D	E	F	G	
1	id	bolting_days	seed_weight	seed_area	ttl_seedspruit	branches	height	
2		A	B	C	D	E	F	G
3	1	marker	MAGIC.1	MAGIC.10	MAGIC.100	MAGIC.101	MAGIC.102	MAGIC.103
4	2	MN1_29291	A	A	A	B	B	A
5	3	MN1_29716	A	A	A	B	B	A
6	4	MN1_112907	A	A	A	B	B	A
7	5	MASC03771	A	B	A	A	B	A
8	6	MN1_197787	A	B	A	B	B	A
9	7	MN1_340810	A	B	A	A	A	A
10	8	MN1_395107	A	A	A	A	B	A
11	9	MN1_444820	A	A	–	A	A	A
12	10	MN1_494205	A	A	A	A	A	A
13	11	MN1_592863	A	B	A	A	A	A
14	12	MN1_592760	A	A	A	A	A	A
15	13	BKN118	A	A	A	A	A	A
16	14	MN1_1042427	A	B	B	A	B	B
	15	CRY2_1021	A	A	A	A	A	A
	16	CRY2_429	A	A	A	A	B	A

Data files

	A	B	C	D	E	F	G															
1	id	bolting_days	seed_weight	seed_area	ttl_seedspruit	branches	height															
2		A	B	C	D	E	F	G														
3	1	marker	MAGIC.1	MAGIC.10	MAGIC.100	MAGIC.101	MAGIC.102	MAGIC.103														
4	2		A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T
5	3	1	marker	Bur	Can	Col	Ct	Edi	Hi	Kn	Ler	Mt	No	Oy	Po	Rsch	Sf	Tsu	Wil	Ws	Wu	Zu
6	4	2	MN1_29291	B	A	A	B	A	A	B	A	A	A	B	B	A	B	A	A	A	A	A
7	5	3	MN1_29716	B	A	A	B	A	A	B	A	A	A	B	B	A	B	A	A	A	A	A
8	6	4	MN1_112907	B	A	A	A	B	B	B	B	A	A	B	B	A	B	B	A	A	A	A
9	7	5	MASC03771	A	B	A	A	B	A	B	A	A	A	A	A	A	A	A	A	A	A	A
10	8	6	MN1_197787	B	B	A	A	A	B	B	A	A	A	A	A	A	B	A	A	A	B	B
11	9	7	MN1_340810	B	B	A	B	A	B	A	A	A	A	A	A	A	A	A	A	B	B	A
12	10	8	MN1_395107	A	A	A	A	A	A	B	A	A	A	A	A	A	A	A	A	A	A	A
13	11	9	MN1_444820	B	A	A	B	B	A	A	A	A	A	A	A	A	A	B	A	B	B	A
14	12	10	MN1_494205	B	A	A	A	B	A	A	A	A	A	A	A	A	A	A	A	B	A	A
15	13	11	MN1_592863	A	B	A	B	A	A	A	B	A	A	A	A	A	A	B	A	B	A	A
16	14	12	MN1_592760	B	A	A	A	B	A	A	A	A	A	A	A	A	A	A	A	A	A	A
	15	13	BKN118	B	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
	16	14	MN1_1042427	A	B	A	B	A	B	B	A	B	A	A	A	B	A	A	B	A	B	A
		15	CRY2_1021	A	A	A	A	B	A	A	A	A	A	A	A	A	A	A	A	A	A	A
		16	CRY2_429	A	A	A	A	A	B	B	B	A	B	A	A	A	A	A	A	B	A	A
		17	MASC07014	A	A	B	A	A	A	A	A	A	A	A	A	B	A	A	B	A	A	A
		18	MASC03609	A	A	A	A	A	A	B	B	A	B	B	B	A	A	A	A	B	A	A
		19	MN1_1296068	A	A	A	A	A	A	B	A	A	A	A	A	A	A	A	A	B	B	A
		20	MN1_1399466	A	A	A	A	A	A	B	A	A	A	A	A	A	A	A	-	B	A	A
		21	AXR1_381	A	B	A	A	A	B	A	A	A	B	A	A	A	A	A	A	B	A	B
		22	MASC07424	A	B	B	A	A	A	A	A	A	A	A	A	A	B	-	A	A	A	A

Data files

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2		A	B	C	D	E	F	G															
3	1	marker	MAGIC.1	MAGIC.10	MAGIC.100	MAGIC.101	MAGIC.102	MAGIC.103															
4	2		A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	
5	3	1	marker	Bur	Cap	Col	Ct	Edi	Hi	Kn	Ler	Mt	No	Ov	Po	Rsch	Sf	Tsu	Wil	Ws	Wu	Zu	
6	4	2	MN1_292	A			B			C													
7	5	3	MN1_297	1	marker			chr			pos												
8	6	4	MN1_112	2	MN1_29291			1			0.029291												
9	7	5	MASC037	3	MN1_29716			1			0.029757												
10	8	6	MN1_197	4	MN1_112907			1			0.112907												
11	9	7	MN1_340	5	MASC03771			1			0.174605												
12	10	8	MN1_395	6	MN1_197787			1			0.197787												
13	11	9	MN1_444	7	MN1_340810			1			0.34081												
14	12	10	MN1_494	8	MN1_395107			1			0.395107												
15	13	11	MN1_592	9	MN1_444820			1			0.444764												
16	14	12	MN1_1042	10	MN1_494205			1			0.494205												
	15	13	CRY2_10	11	MN1_592863			1			0.592867												
	16	14	CRY2_4	12	MN1_592760			1			0.592984												
		15	MASC070	13	BKN118			1			0.761584												
		16	MASC036	14	MN1_1042427			1			1.042428												
			MN1_1296	15	CRY2_1021			1			1.187841												
			MN1_1399	16	CRY2_429			1			1.188433												
			AXR1_3	17	MASC07014			1			1.189374												
			MASC074	18	MASC03609			1			1.22655												

Control file (json or yaml)

```
{
  "description": "Arabidopsis MAGIC data, Gnan et al (2014)",
  "crosstype": "magic19",
  "sep": ",",
  "na.strings": ["-", "NA"],
  "comment.char": "#",
  "geno": "arabmagic_geno.csv",
  "founder_geno": "arabmagic_foundergeno.csv",
  "gmap": "arabmagic_pmap_tair9.csv",
  "pmap": "arabmagic_pmap_tair9.csv",
  "pheno": "arabmagic_pheno.csv",
  "genotypes":
    "A": 1
    "H": 2
    "B": 3
},
  "geno_transposed": true,
  "founder_geno_transposed": true
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  "pheno": "arabmagic_pheno.csv",
  "genotypes":
    "A": 1
    "H": 2
    "B": 3
},
  "geno_transposed": true,
  "founder_geno_transposed": true
}
```

Reading data into R

```
library(qtl2)  
arab <- read_cross2("arab_magic.json")
```

Reading data into R

```
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arab <- read_cross2("arab_magic.json")
```

19-way Arabidopsis MAGIC

Kover et al. (2009) PLoS Genet

Gnan et al. (2014) Genetics

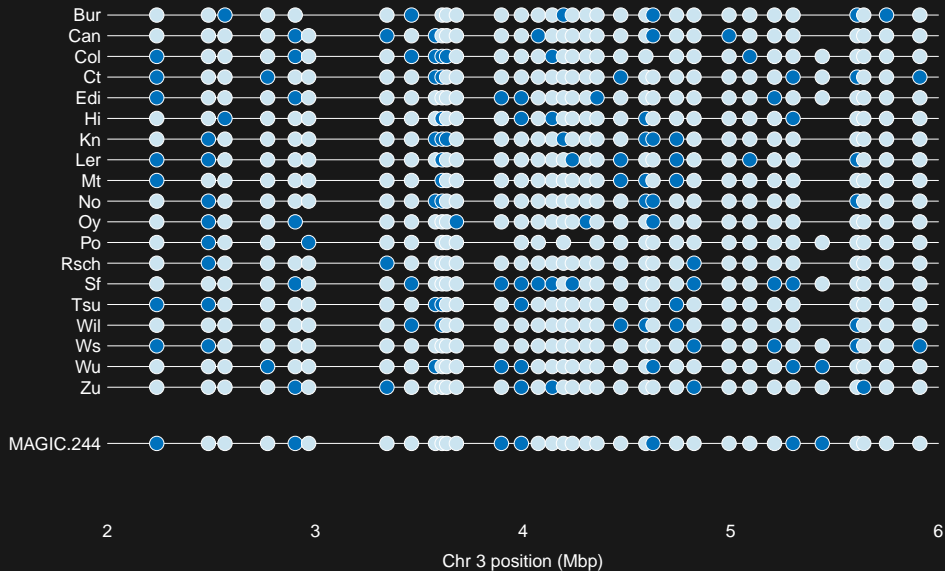
github.com/rqtl/qtl2data

Data diagnostics

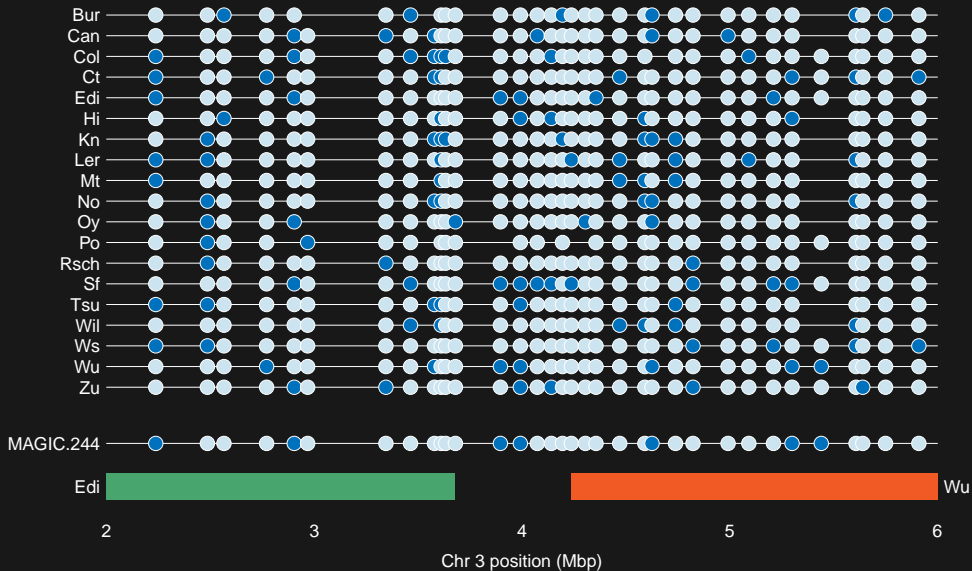
See Broman et al. (2019) Cleaning genotype data from
Diversity Outbred mice. G3 9:1571–1579

doi: [10.1534/g3.119.400165](https://doi.org/10.1534/g3.119.400165)

Genotype reconstruction



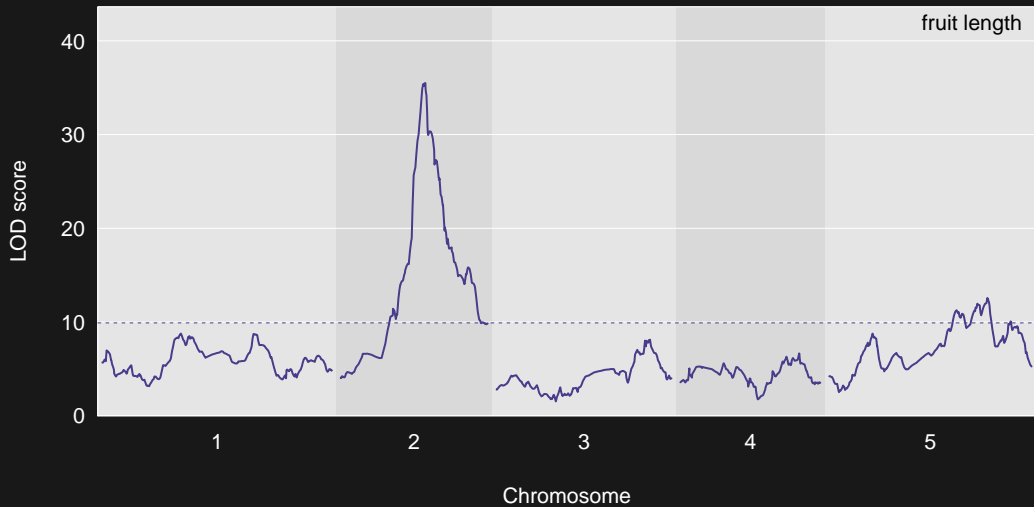
Genotype reconstruction



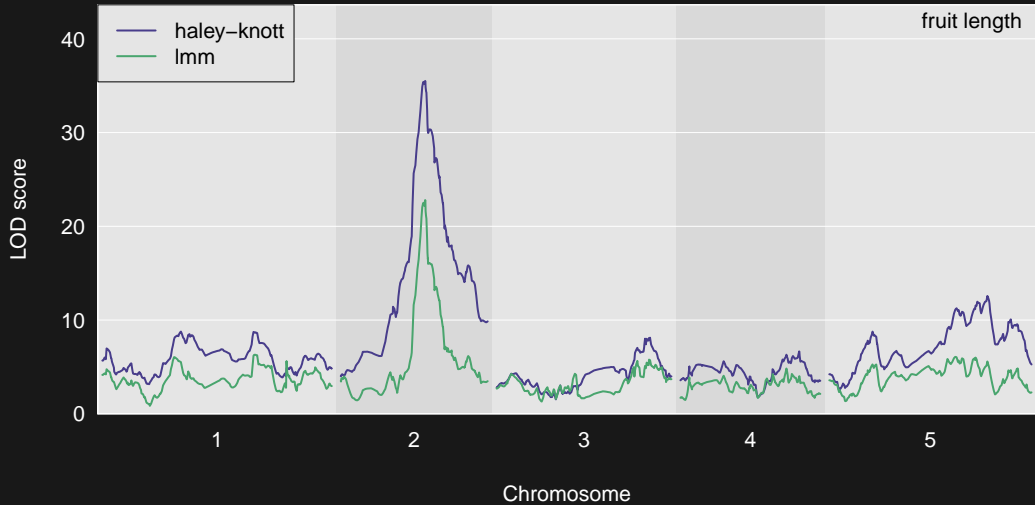
Genotype reconstruction

```
gmap <- insert_pseudomarkers(arab$gmap, step=0.2, stepwidth="max")  
pmap <- interp_map(gmap, arab$gmap, arab$pmap)  
  
pr <- calc_genoprob(arab, gmap, error_prob=0.002, cores=24)
```

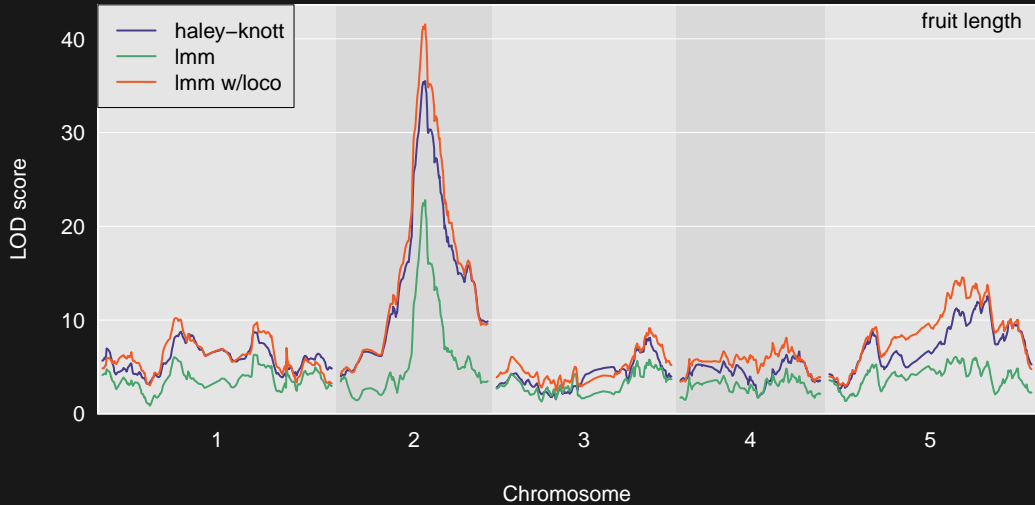
Genome scan



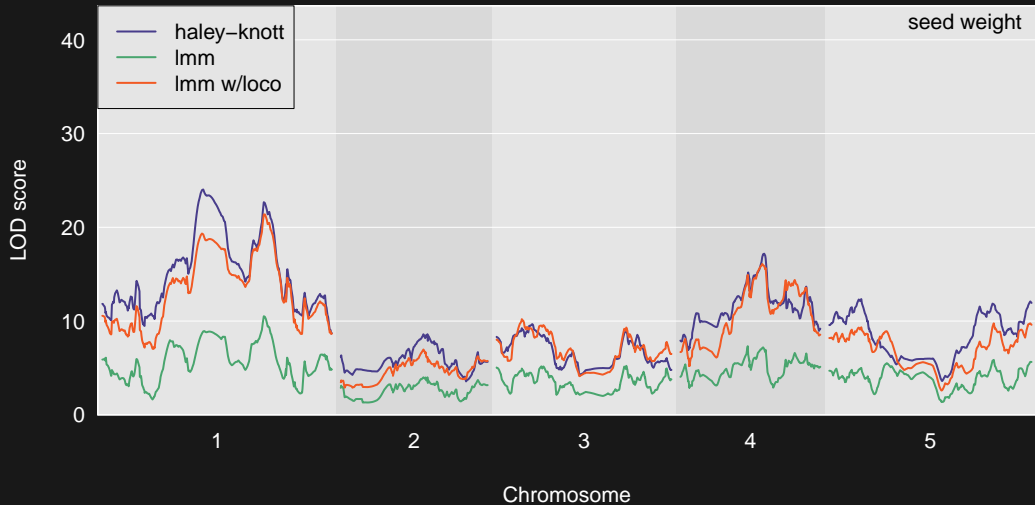
Genome scan



Genome scan



Genome scan



Genome scan

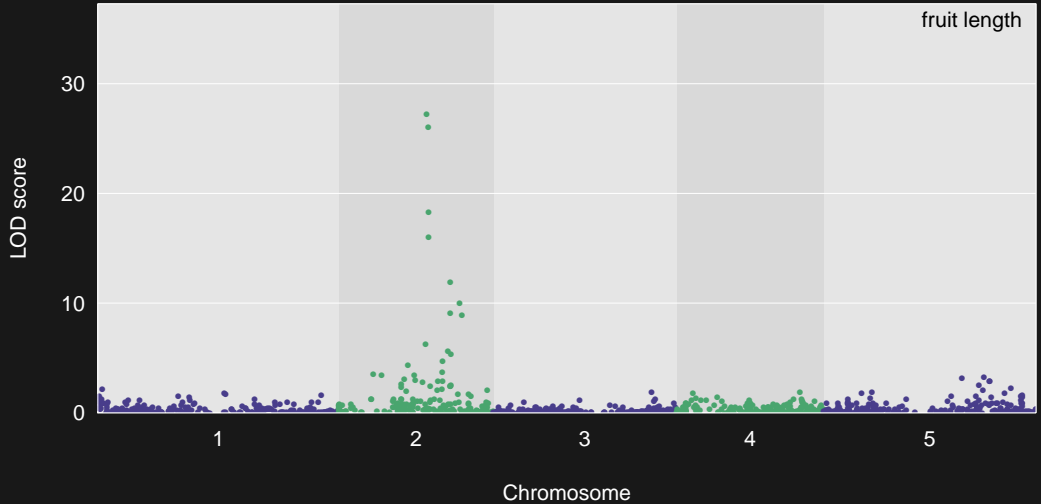
```
out_hk <- scan1(pr, arab$pheno, cores=24)

operm_hk <- scan1perm(pr, arab$pheno, n_perm=1000, cores=24)

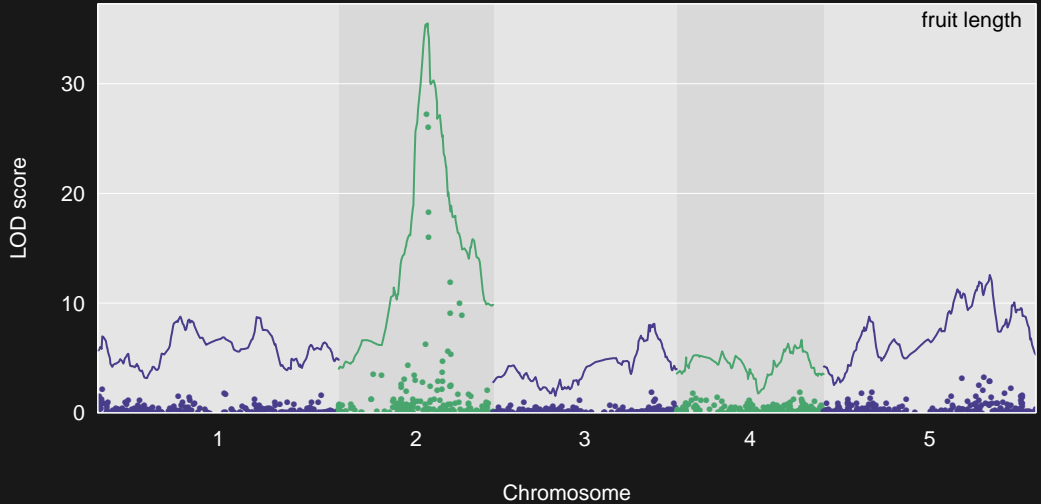
k <- calc_kinship(pr, cores=24)
out_lmm <- scan1(pr, arab$pheno, k, cores=24)

k_loco <- calc_kinship(pr, "loco", cores=24)
out_loco <- scan1(pr, arab$pheno, k_loco, cores=24)
```

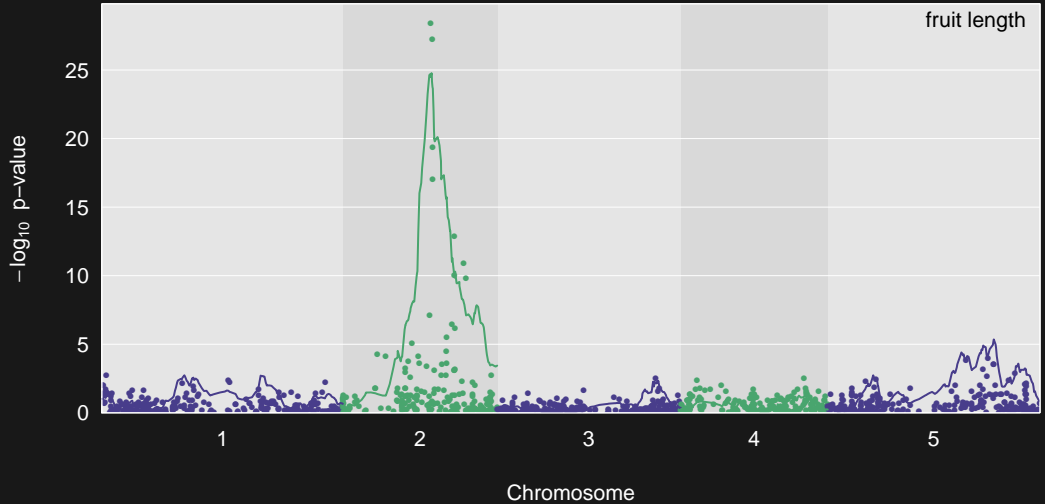

SNP association scan



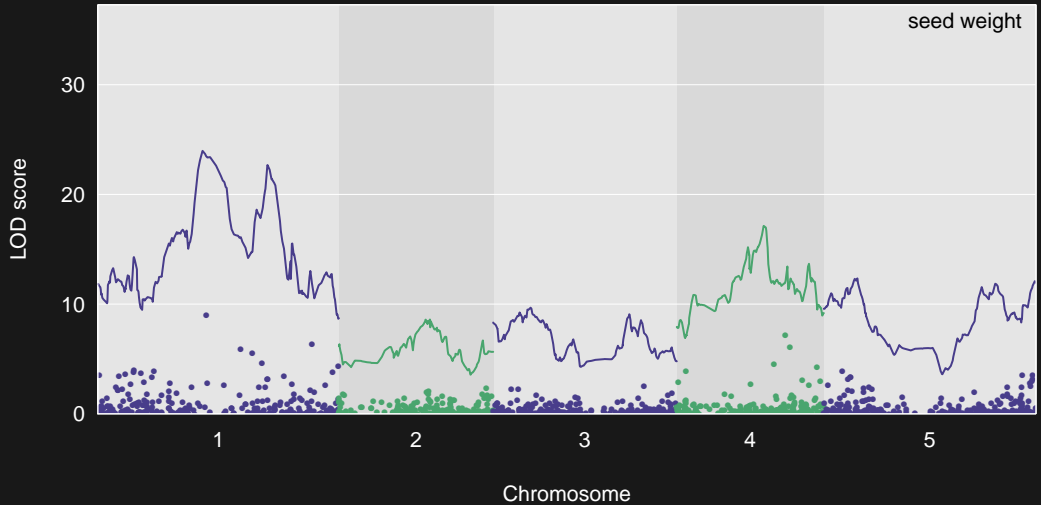
SNP association scan



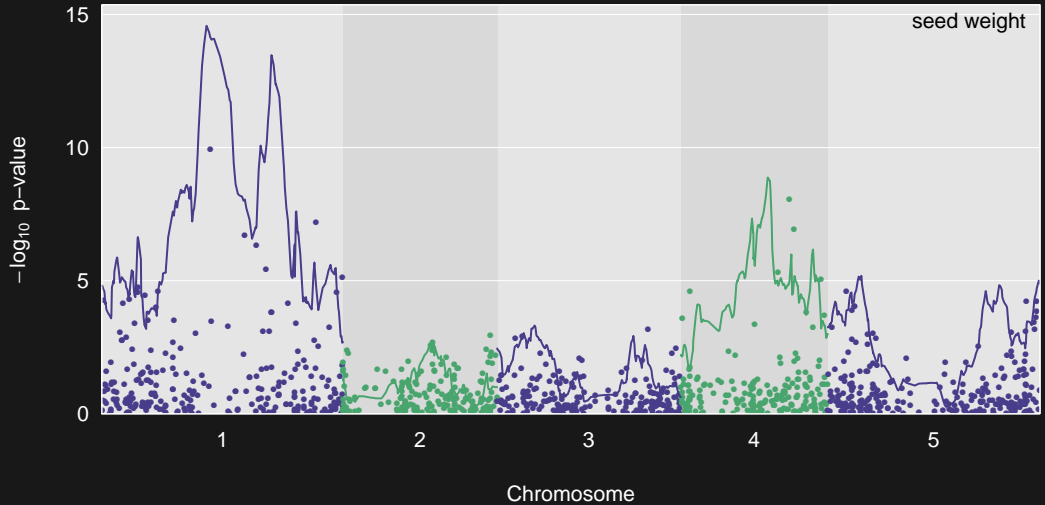
SNP association scan



SNP association scan



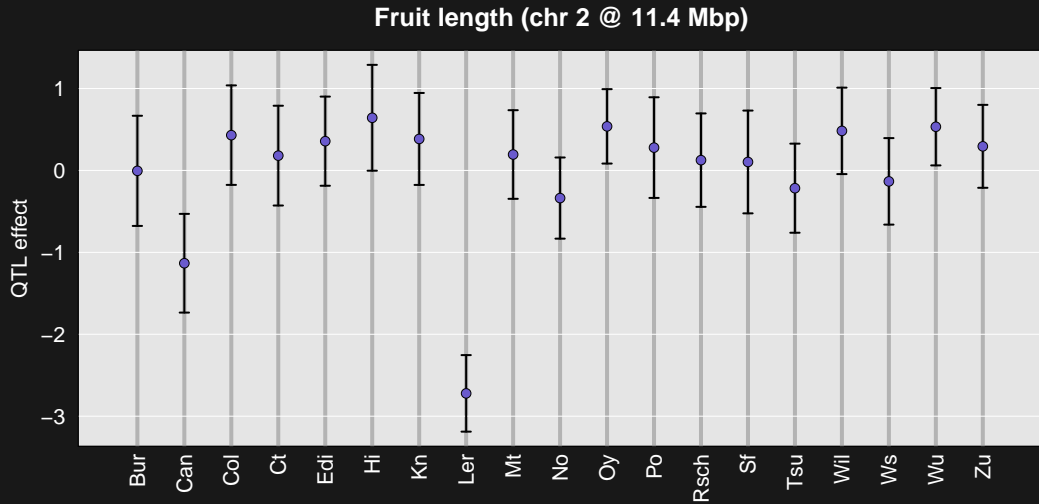
SNP association scan



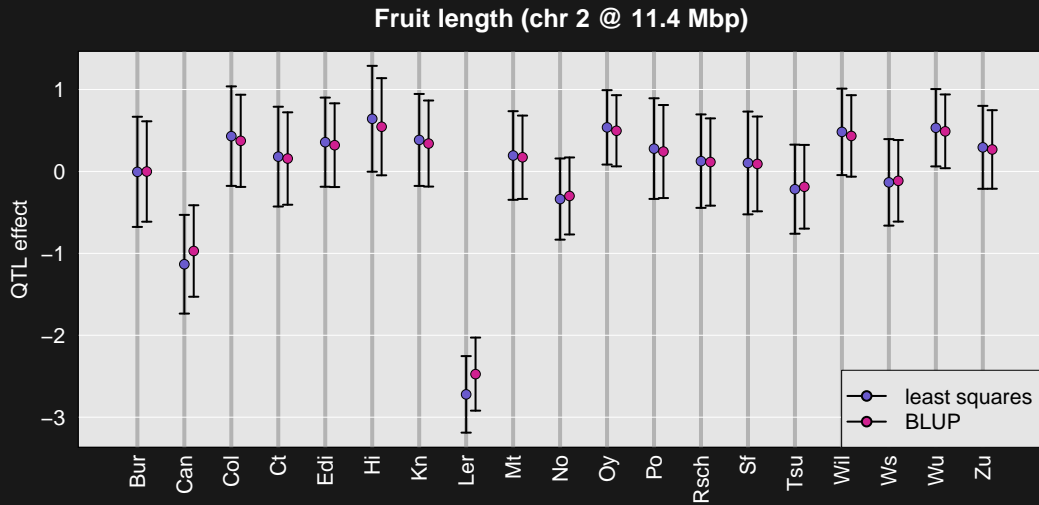
SNP association scan

```
snp_pr <- genoprob_to_snpprob(pr, arab)  
out_snps <- scan1(snp_pr, arab$pheno, cores=24)
```

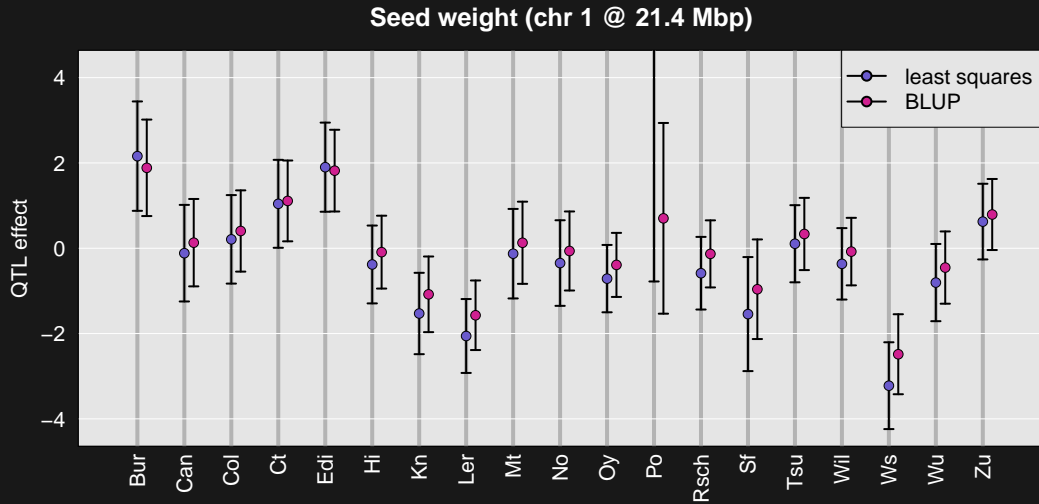
QTL effects



QTL effects



QTL effects



QTL effects

```
fl_peak <- max(out_hk, pmap, lodcolumn="fruit_length")
fl_pr <- pull_genoprobpos(pr, pmap, fl_peak$chr, fl_peak$pos)

fl_fit1 <- fit1(fl_pr, arab$pheno[, "fruit_length"])
fl_blup <- fit1(fl_pr, arab$pheno[, "fruit_length"], blup=TRUE)
```

Goals

- ▶ Genotype reconstructions from external software
- ▶ Sequencing-based genotype data
- ▶ Multiple-QTL models
- ▶ QTL \times environment interactions
- ▶ Interactive data visualization

Slides: kbroman.org/Talk_MAGIC2021



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