



**Context Dependent Effects of Alleles Affecting
Quantitative Traits: Insights From *Drosophila***

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The case of the missing heritability

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REVIEWS

Finding the missing heritability of complex diseases

Teri A. Manolio¹, Francis S. Collins², Nancy J. Cox³, David B. Goldstein⁴, Lucia A. Hindorf⁵, David J. Hunter⁶, Mark I. McCarthy⁷, Erin M. Ramos⁵, Lon R. Cardon⁸, Aravinda Chakravarti⁹, Judy H. Cho¹⁰, Alan E. Guttmacher¹, Augustine Kong¹¹, Leonid Kruglyak¹², Elaine Mardis¹³, Charles N. Rotimi¹⁴, Montgomery Slatkin¹⁵, David Valle⁹, Alice S. Whittemore¹⁶, Michael Boehnke¹⁷, Andrew G. Clark¹⁸, Evan E. Eichler¹⁹, Greg Gibson²⁰, Jonathan L. Haines²¹, Trudy F. C. Mackay²², Steven A. McCarroll²³ & Peter M. Visscher²⁴

Why are Effects of Variants Affecting Human Complex Traits so Small?

h^2 overestimated using twin studies

“SNP Chips” have limitations:

SNPs disproportionately in coding regions

Not all common variants represented

No rare variants represented

Cannot detect copy number variants, insertions and deletions

Human genome has limitations:

SNPs located in LD blocks

Associated SNPs may not be casual, but in LD with true causal variant, reducing estimated effect size

True causal variant may be common or rare

Analytical methods have limitations:

Low power to detect gene-gene interactions (epistasis)

Low power to detect gene-environment interactions

Need very large samples to detect truly additive variants with very small effects

Missing heritability may be due to:

SNPs in non-coding regions

Large effect rare variants in LD with Chip SNP

Large effect common variants in LD with Chip SNP

Large effect undetected structural variants

Common variants with truly small effects

Teri A. Manolio¹, Francis S. Collins², Nancy L. Cox³, David B. Goldstein⁴, Lorna A. Hindorf⁵, David J. Hunter⁶, Mark I. McCarthy⁷, Erin M. Ramos⁸, Lon R. Cardon⁹, Aravinda Chakravarti¹⁰, Judy H. Cho¹⁰, Alan E. Guttmacher¹, Augustine Kong¹¹, Leonid Kruglyak¹², Elaine Mardis¹³, James N. Rotimi¹⁴, Montgomery Slatkin¹⁵, David Valle⁹, Alice S. Whittemore¹⁰, Michael Boehnke¹⁶, Andrew G. Clark¹⁷, Evan E. Eichler¹⁸, Greg Gibson²⁰, Jonathan L. Haines²¹, Trudy F. C. Mackay²², Steven A. McCarroll²³ & Peter M. Visscher²¹

REVIEWS

Finding the missing heritability of complex diseases

Why are Effects of Variants Affecting Human Complex Traits so Small?



The case of the missing heritability

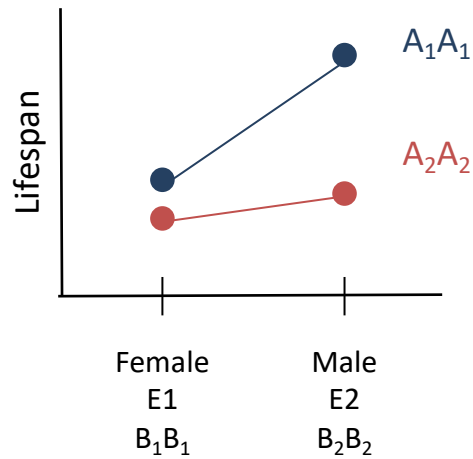
Vol 461 **Genotype by sex interaction (genetic variation in sexual dimorphism)** nature
Genotype by environment interaction (genetic variation in environmental plasticity)
Genotype by genetic background interaction (epistasis)

REVIEWS

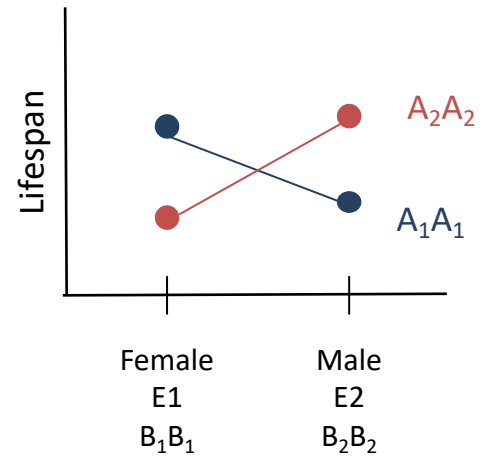
Finding the missing heritability of complex diseases

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Genetic Interactions



Change of variance
Effect may be large in one context only



Change of rank order
Opposite effects in different contexts
("Antagonistic pleiotropy")

Do these phenomena occur?



Needed:

Whole genome sequences

Minimal Local LD

Controlled environments

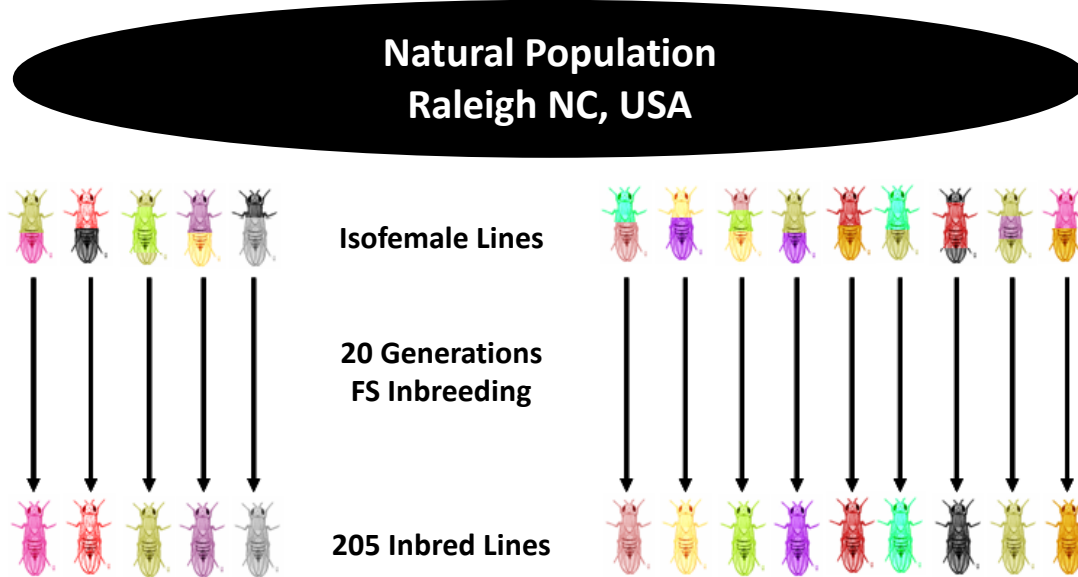
Power to detect individual variants associated with quantitative variation

Power to detect gene-environment interaction

Power to detect epistasis

***Drosophila* fulfills many of these needs**

Drosophila melanogaster Genetic Reference Panel (DGRP)



Illumina genome sequences (27X)

Highly polymorphic

Rapid decay LD with physical distance

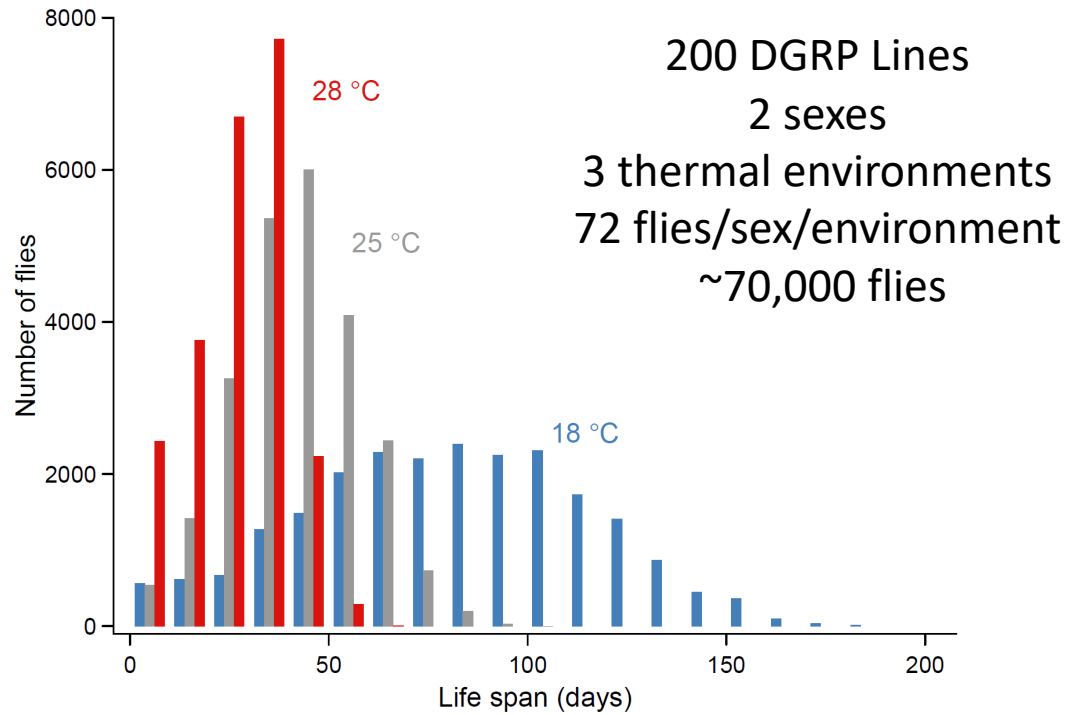
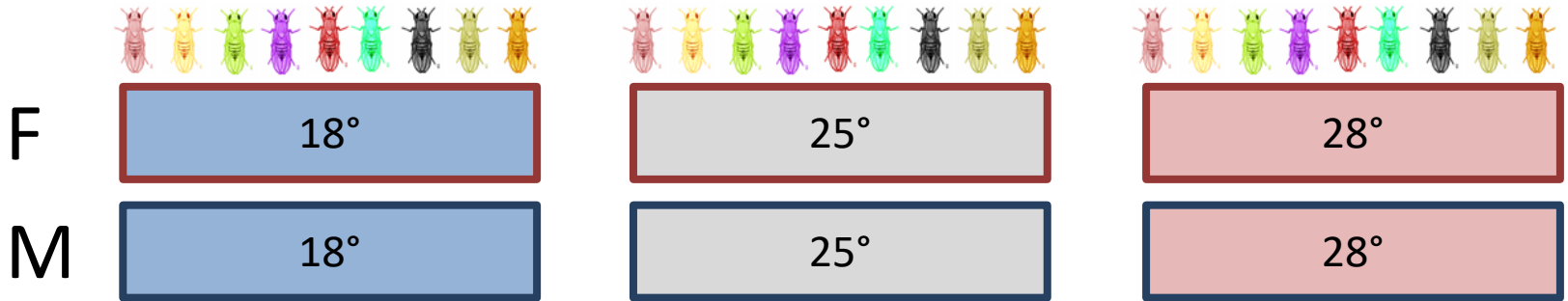
Increased genetic variance compared to outbred population ($cov_G = 2V_A + 4V_{AA} + 8V_{AAA} + \dots$)

Replicated genotypes:

High heritability ($H^2 = V_G / (V_G + V_E/n)$, for n individuals per genotype)

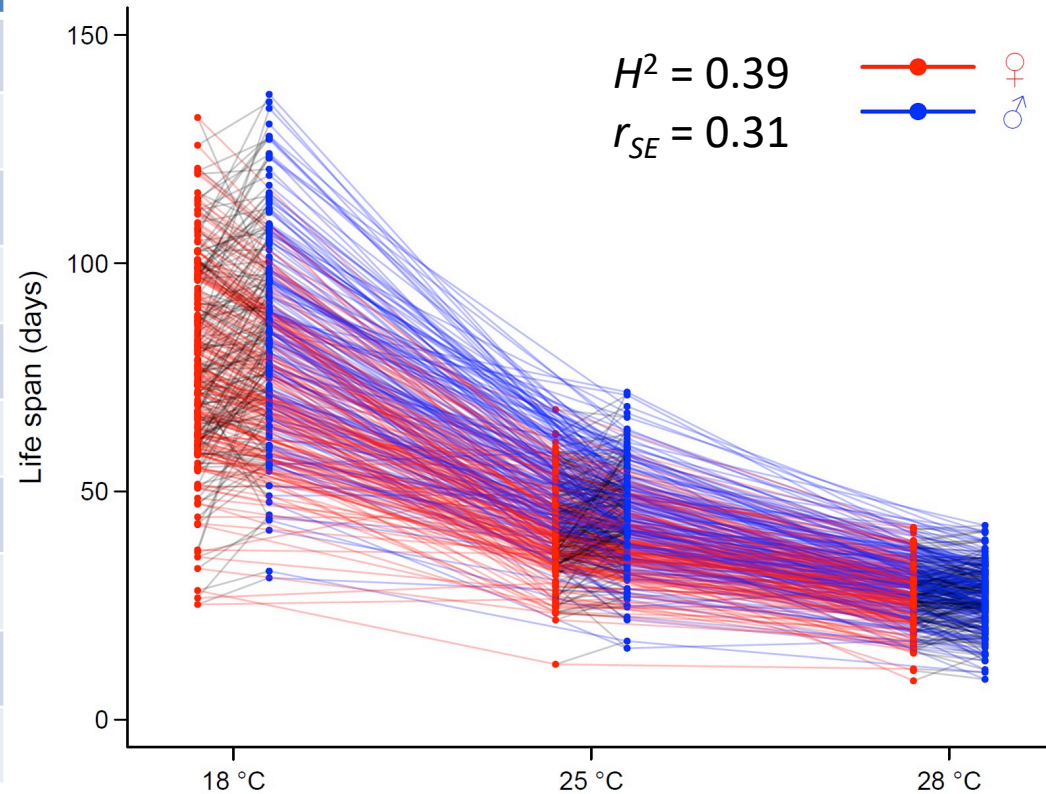
Multiple environments

Variation for Lifespan in the DGRP



Variation for Lifespan in the DGRP

Source of Variation	P-Value	σ^2
Sex (<i>S</i>)	7.67e-10	Fixed
Temperature (<i>T</i>)	5.80e-163	Fixed
DGRP Line (<i>L</i>)	1.94e-13	59.78
<i>S</i> × <i>T</i>	4.59e-14	Fixed
<i>S</i> × <i>L</i>	2.22e-5	8.72
<i>T</i> × <i>L</i>	6.72e-56	93.08
<i>S</i> × <i>T</i> × <i>L</i>	1.14e-241	32.59
Rep(<i>T</i> × <i>L</i>)	5.07e-29	16.34
<i>S</i> ×Rep(<i>T</i> × <i>L</i>)	7.32e-92	32.40
Residual		295.72



Sex dimorphism

Environmental plasticity

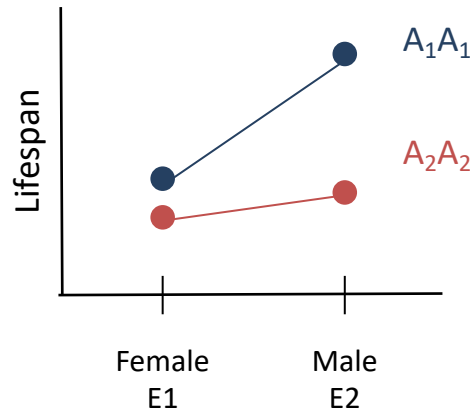
Genetic variation

Genetic variation in sex dimorphism (genotype by sex interaction, GSE)

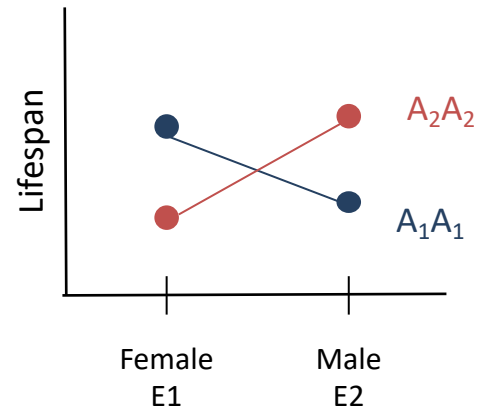
Genetic variation in environmental plasticity (genotype by environment interaction, GEI)

Genetic variation in sex dimorphism varies with thermal environment (GSEI)

Genetic Interactions



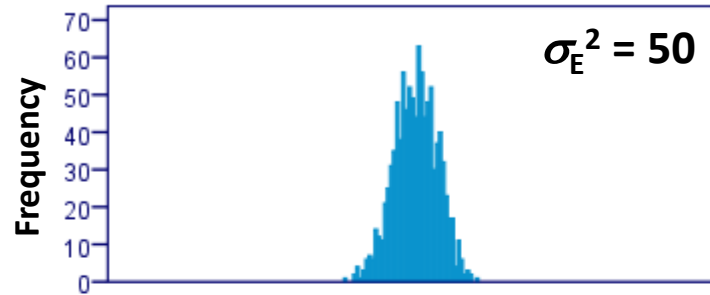
Change of variance
Sex or environment-specific



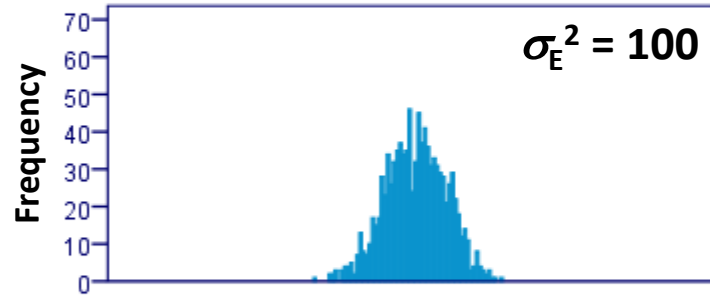
Change of rank order
Sex or environment antagonistic pleiotropy

“Micro-Environmental Plasticity” for Lifespan

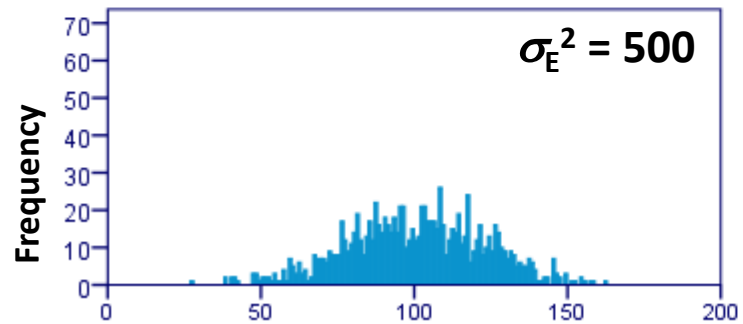
DRGP Line 1



DRGP Line 2



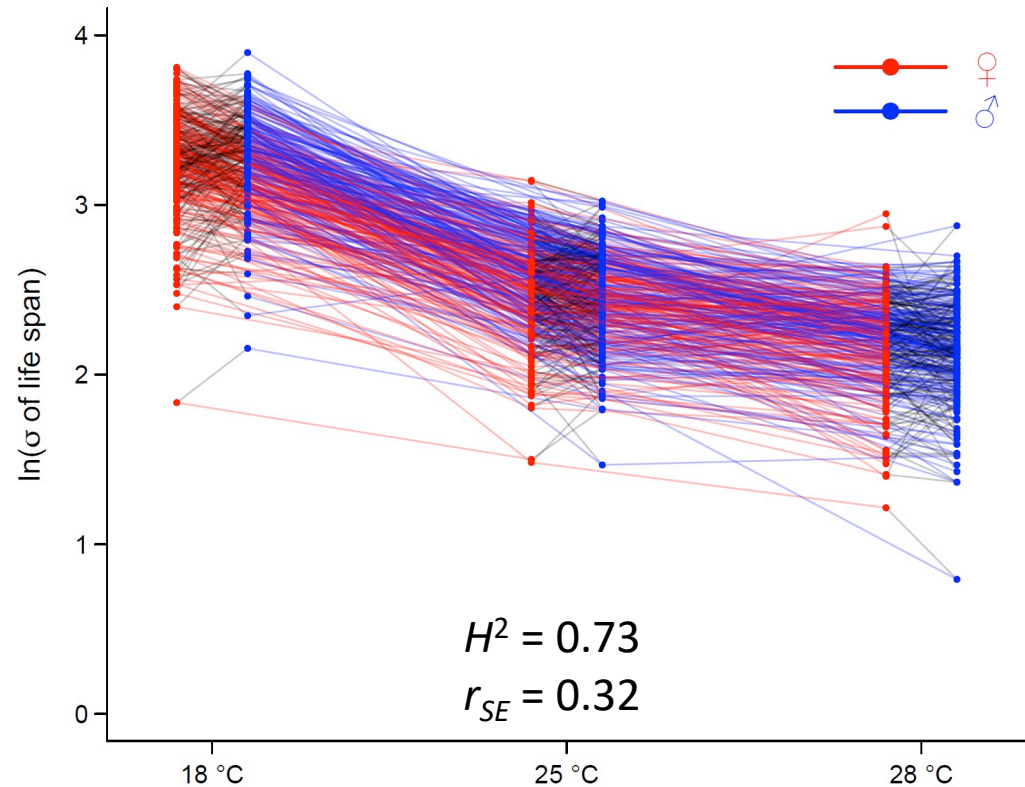
DRGP Line 3



Lifespan (days)

Variation for Micro-Environmental Plasticity of Lifespan in the DGRP

Source of Variation	P-Value	σ^2
Sex (<i>S</i>)	9.289E-01	Fixed
Temperature (<i>T</i>)	4.682E-164	Fixed
DGRP Line (<i>L</i>)	1.353E-11	0.0241
<i>S</i> × <i>T</i>	3.620E-03	Fixed
<i>S</i> × <i>L</i>	5.037E-03	0.0037
<i>T</i> × <i>L</i>	6.804E-27	0.0318
<i>S</i> × <i>T</i> × <i>L</i>	6.462E-19	0.0149
Error		0.0282



Sex dimorphism

Environmental plasticity

Genetic variation

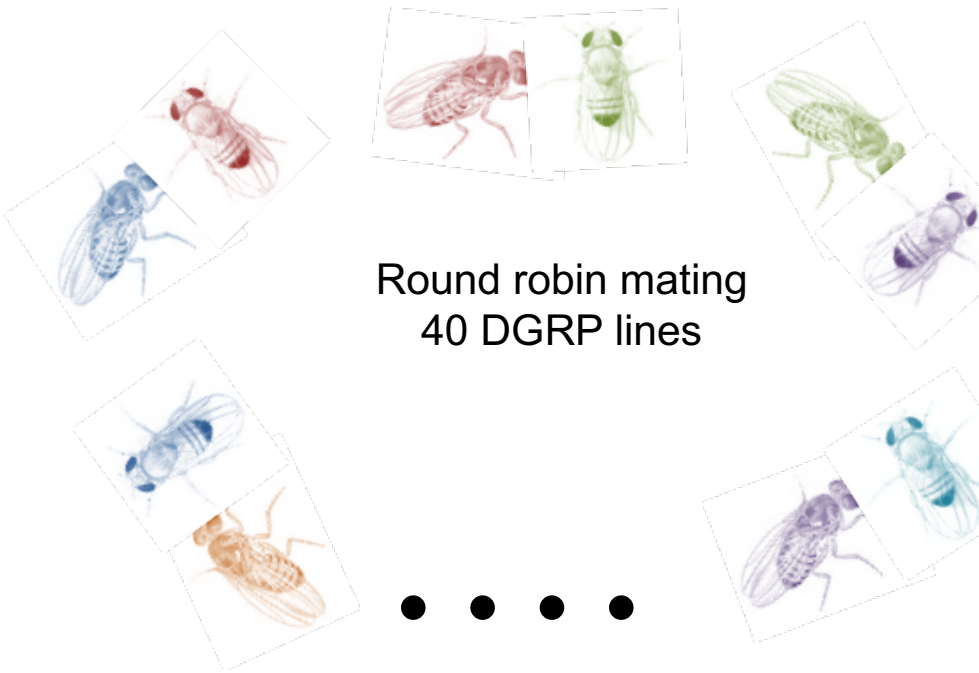
Genetic variation in sex dimorphism (genotype by sex interaction, GSE)

Genetic variation in environmental plasticity (genotype by environment interaction, GEI)

Genetic variation in sex dimorphism varies with thermal environment (GSEI)

Advanced Intercross Populations

AIP

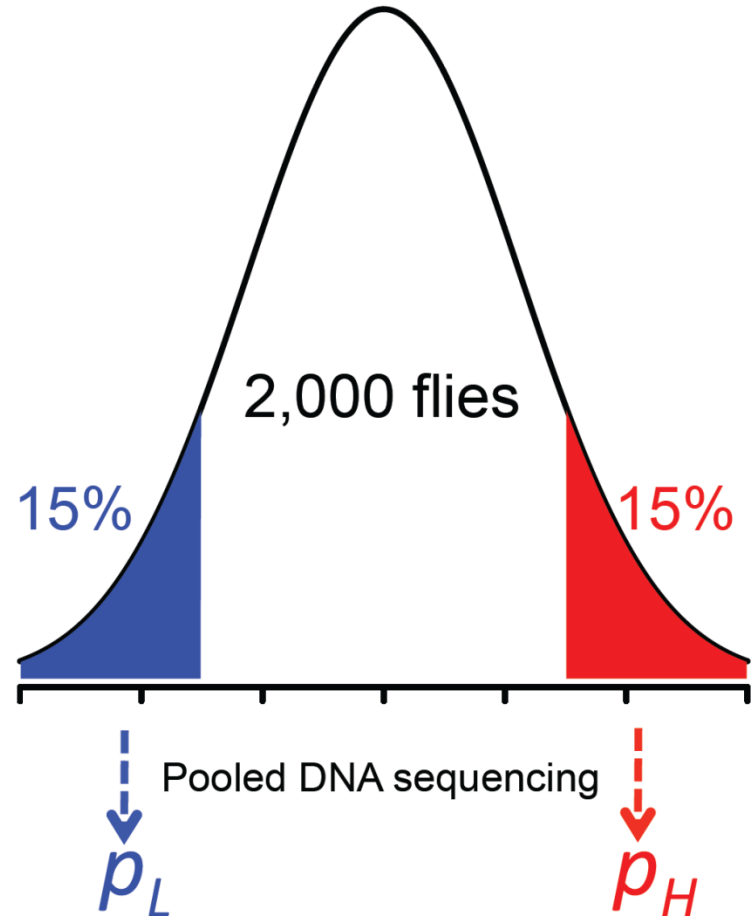


Round robin mating
40 DGRP lines

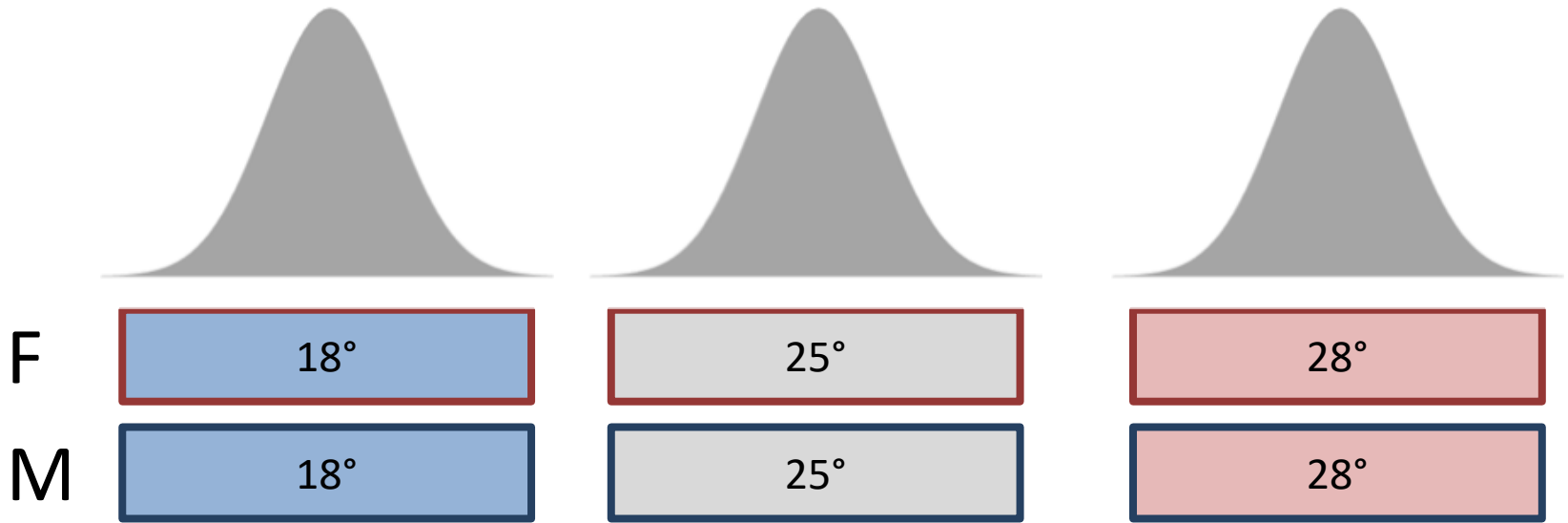
Random mating
170 generations
 $N= 800/\text{generation}$

Extreme QTL Mapping

xQTL



Variation for Lifespan in the AIP



~2000 flies

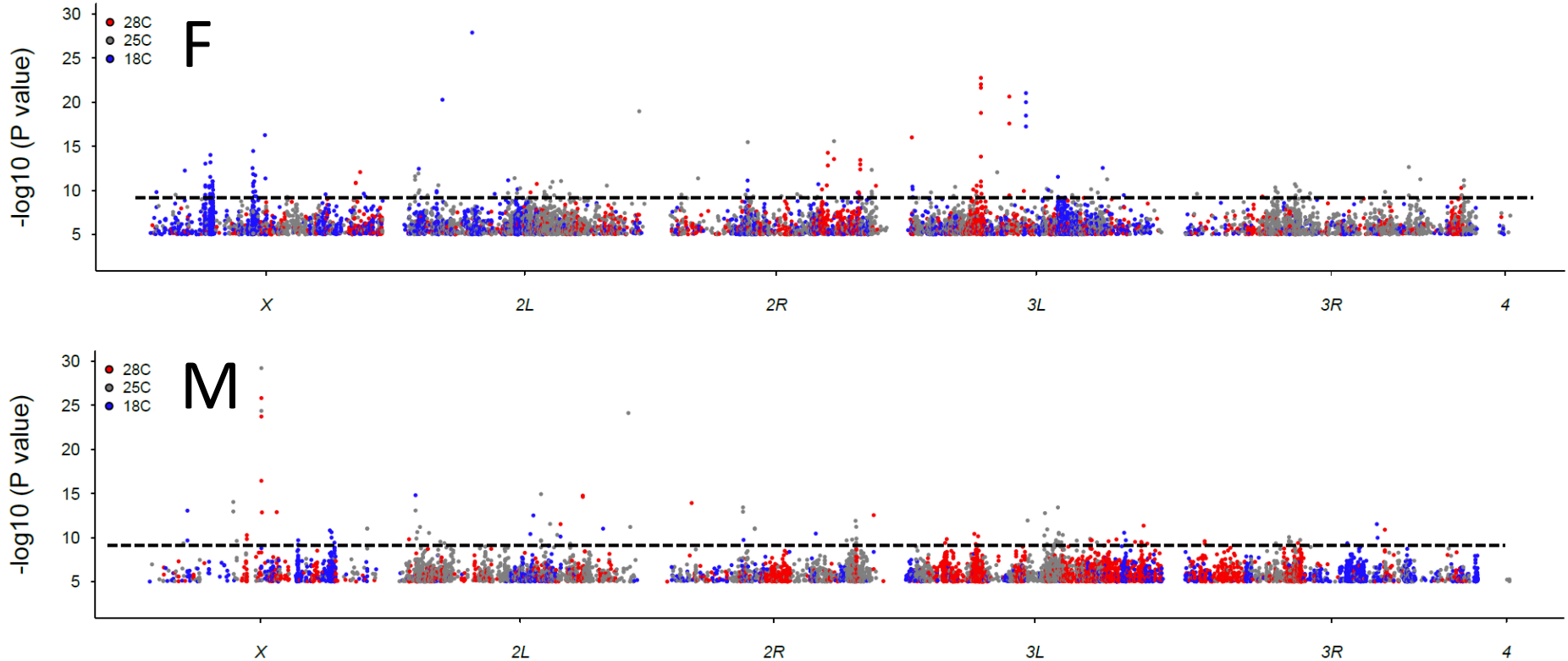
2 sexes

3 environments

2 replicates/sex/environment

~24,000 flies

GWAS for Lifespan in the AIP



2,065 SNPs in or near 1,326 genes significant at $P < 10^{-7}$
Only 14 variants overlap between sexes, thermal environments
All other variants had sex- and environment-specific effects
Massive GSI, GEI

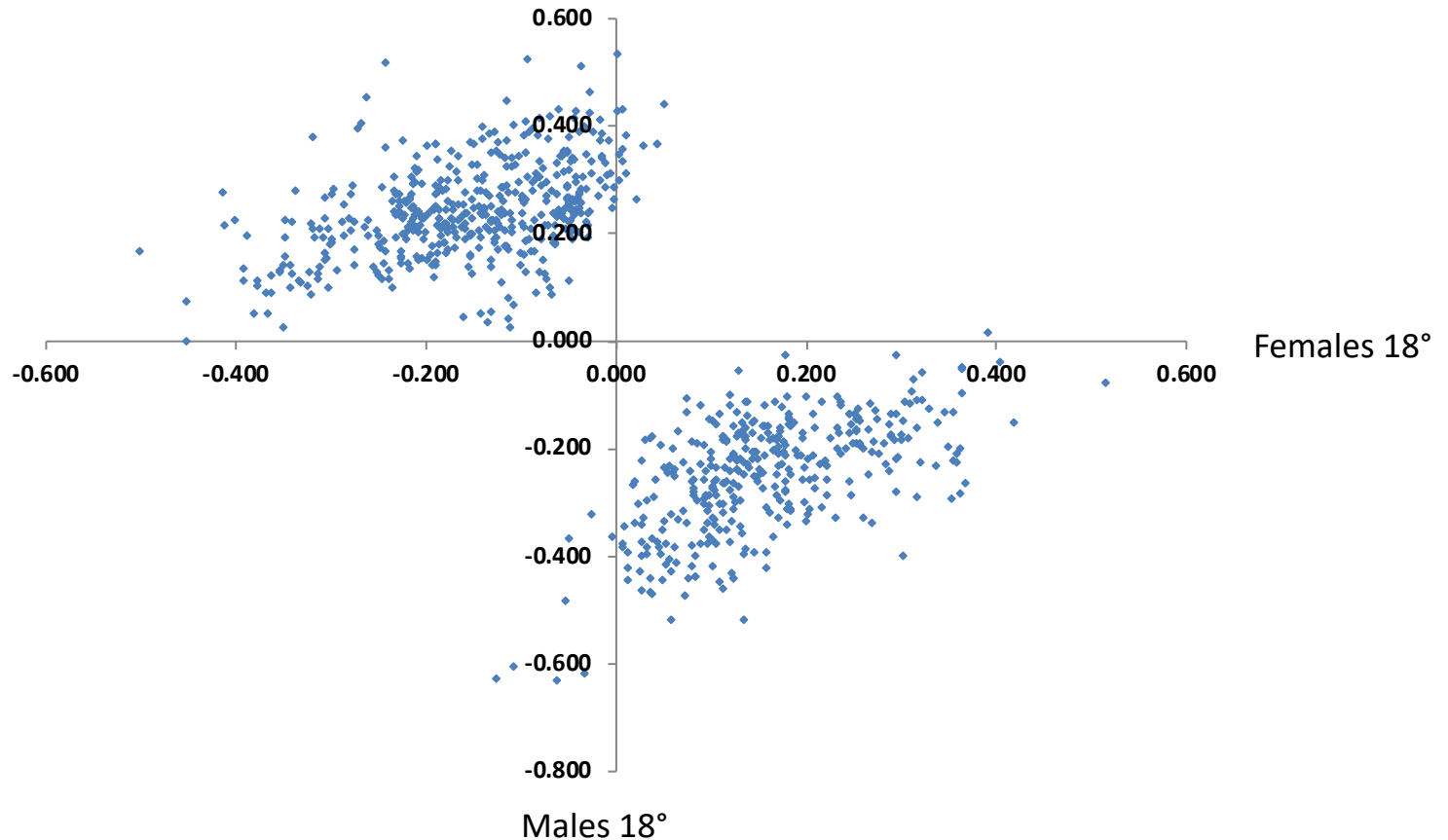
GWAS for GSI, GEI Lifespan in the AIP

2,368 SNPs associated with GSI ($P < 10^{-5}$)

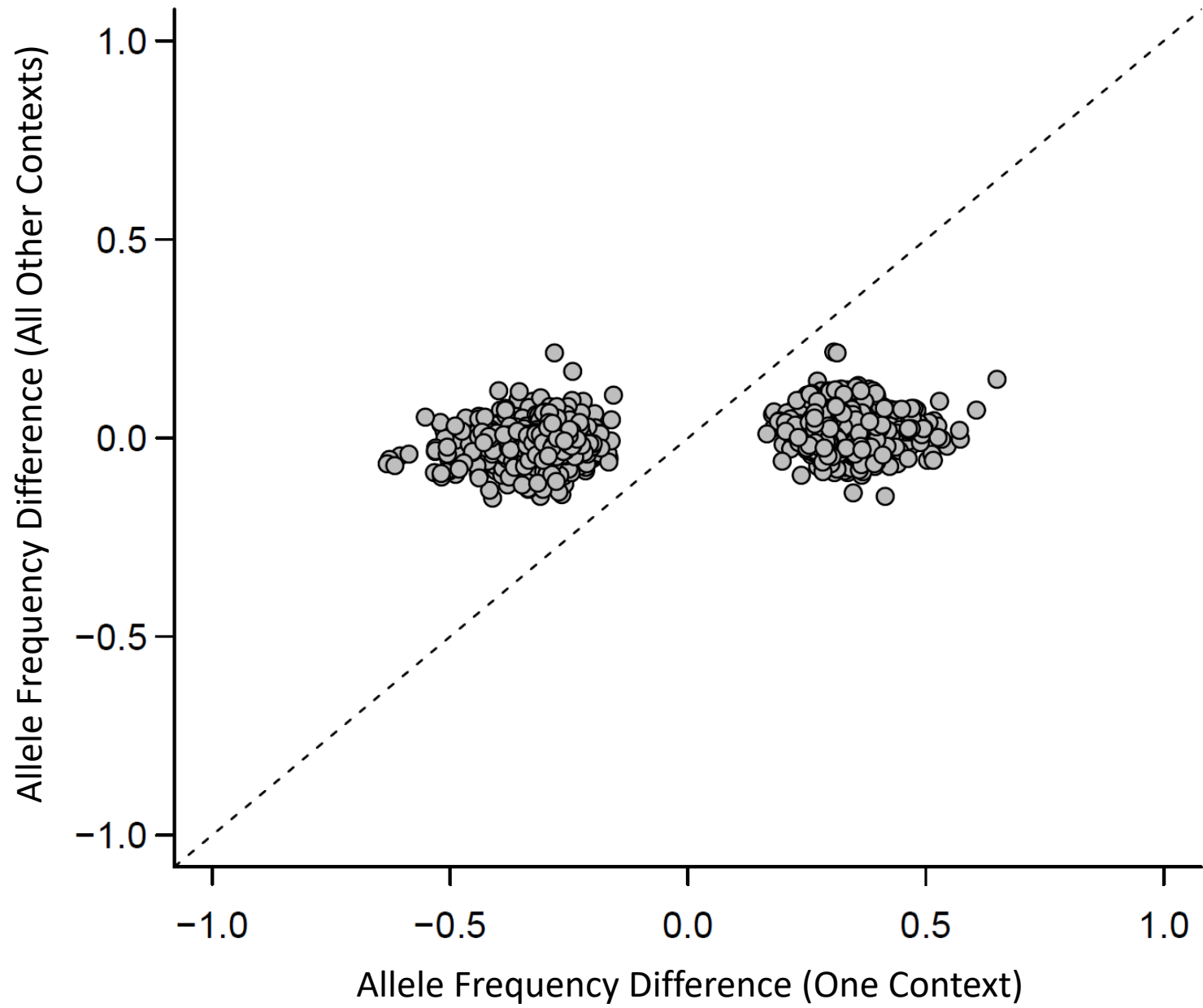
1,619 SNPs associated with GEI ($P < 10^{-7}$)

ALL have opposite effects between sexes or temperatures

Antagonistic pleiotropy pervasive



Take Home Message

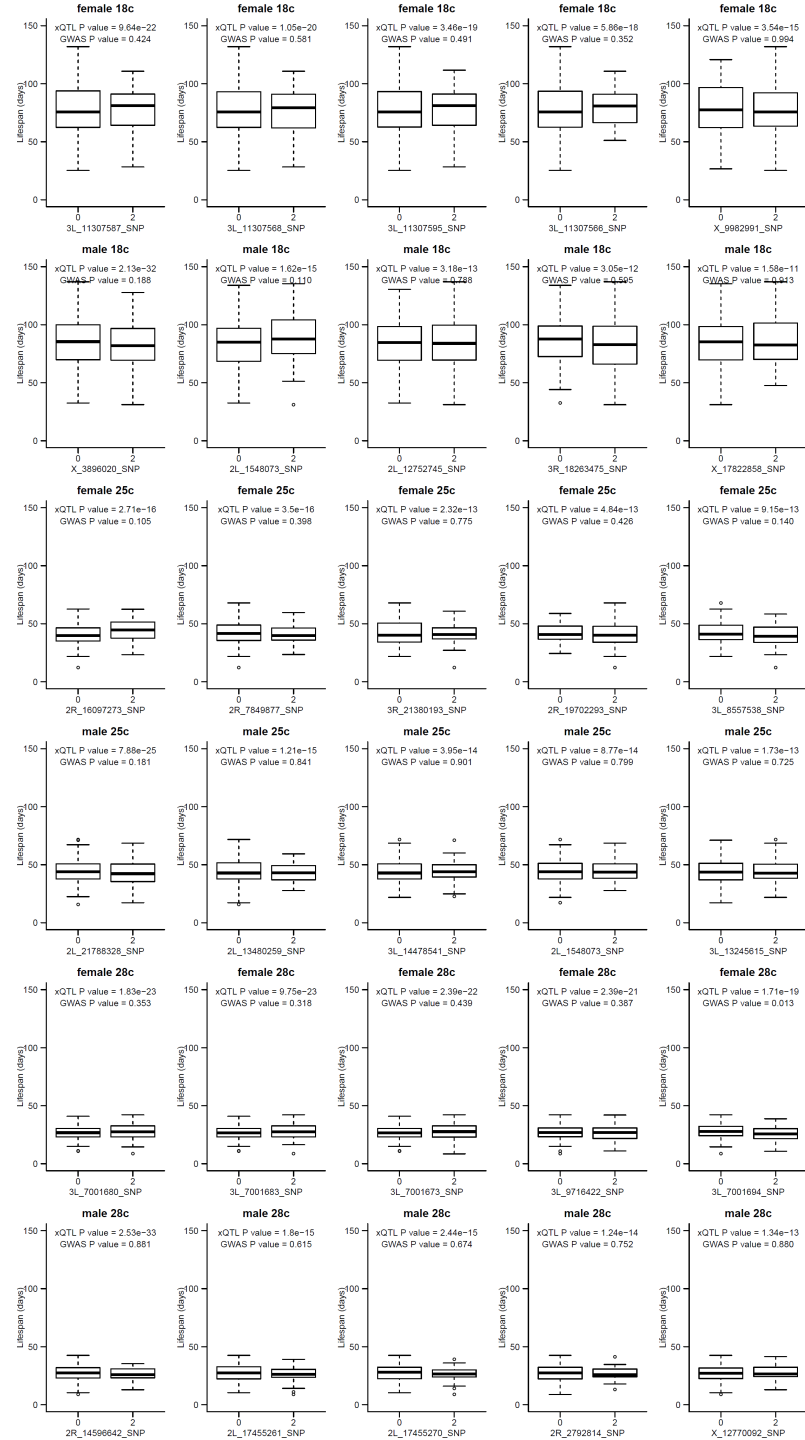


Comparison of Allelic Effects in AIP and DGRP

None of the top variants in the AIP are significant when tested in the DGRP in the same context

Effects of top AIP variants are large within each context

Adequate power to detect effects this large in DGRP

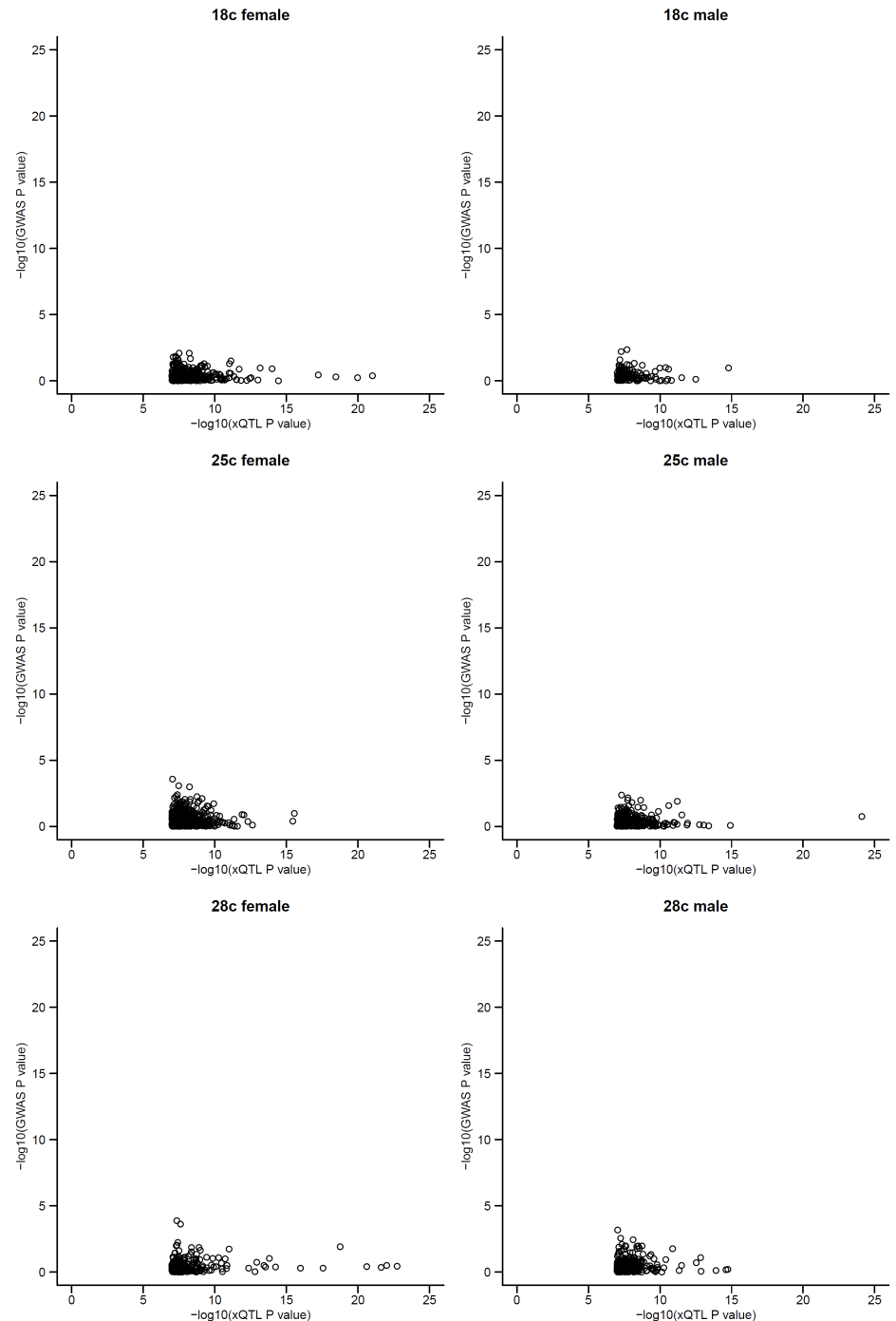


Comparison of Allelic Effects in AIP and DGRP

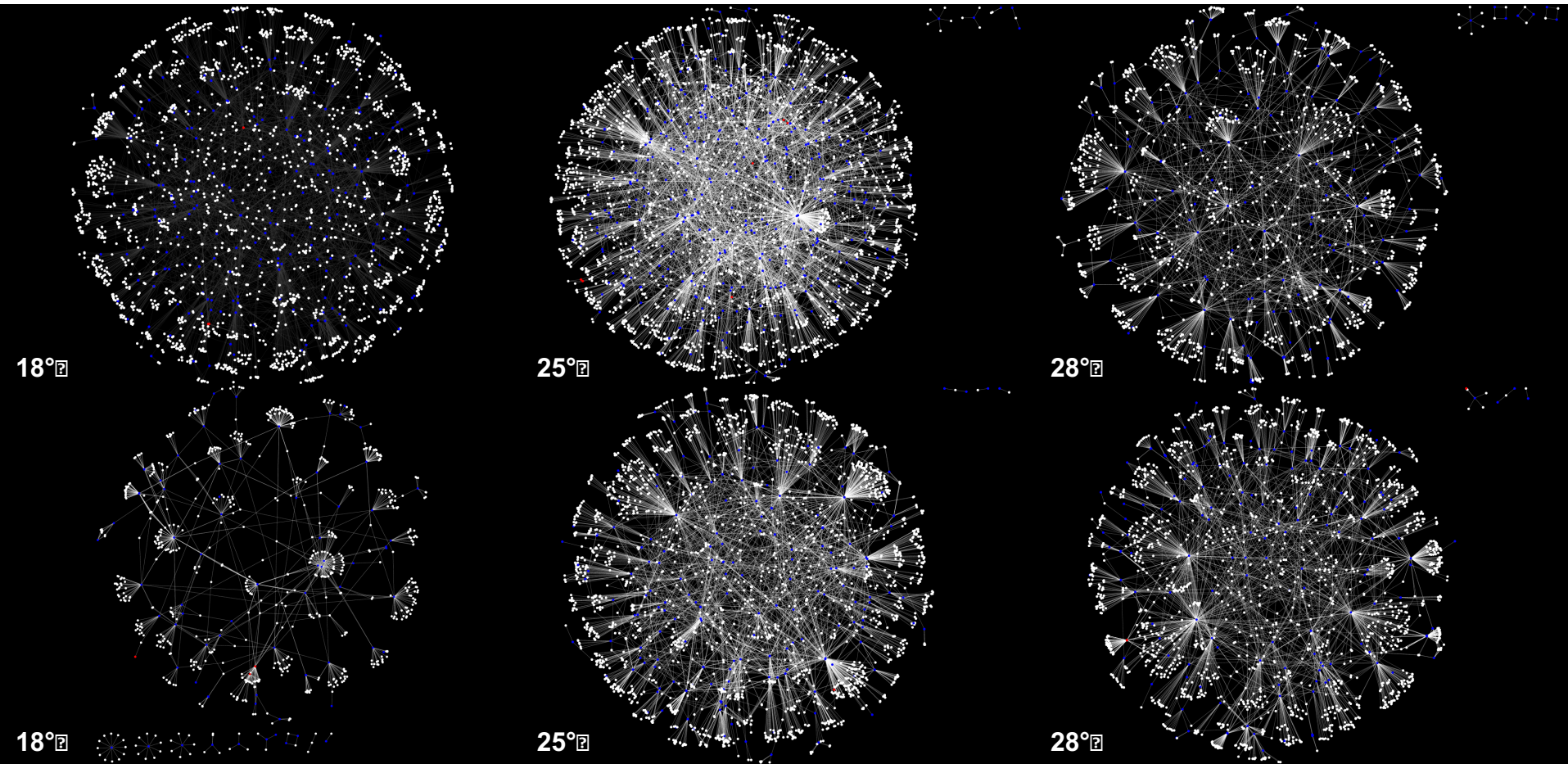
None of the variants ($P < 10^{-7}$) in the AIP are significant when tested in the DGRP in the same context

This could happen if all AIP and DGRP GWAS hits are false positives – not likely for AIP GWAS analyses

This could also happen if there is pervasive epistasis and allele frequencies of interacting alleles are different between the DGRP and AIP – and they are by design



Combine AIP and DGRP Data to Derive Genetic Interaction Networks



Inferred epistatic interactions networks sex- and temperature specific

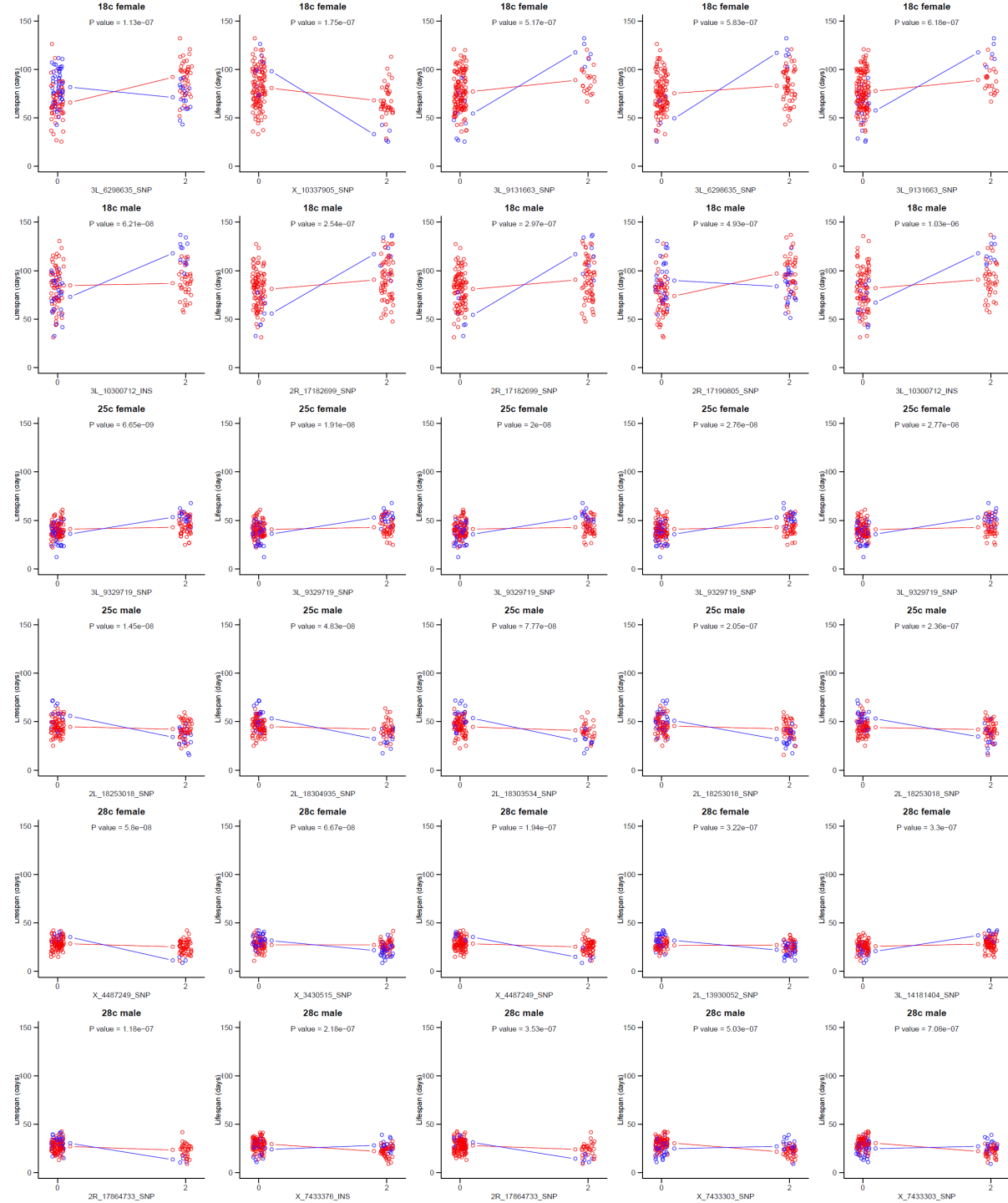
None of the inferred interactions individually significant

Top Epistatic Interactions in Each Context

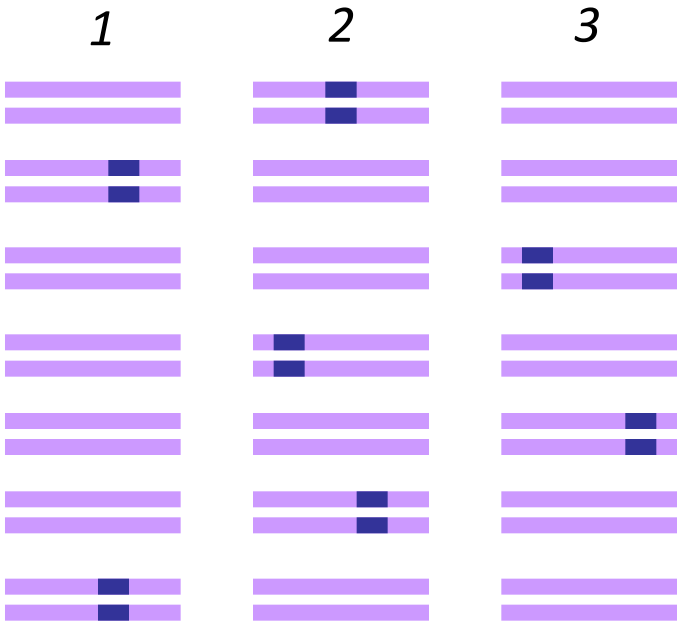
All interactions exhibit change
of rank order in different
genetic backgrounds

This type of interaction
generates no additive genetic
variance when alleles at both
interacting loci are common

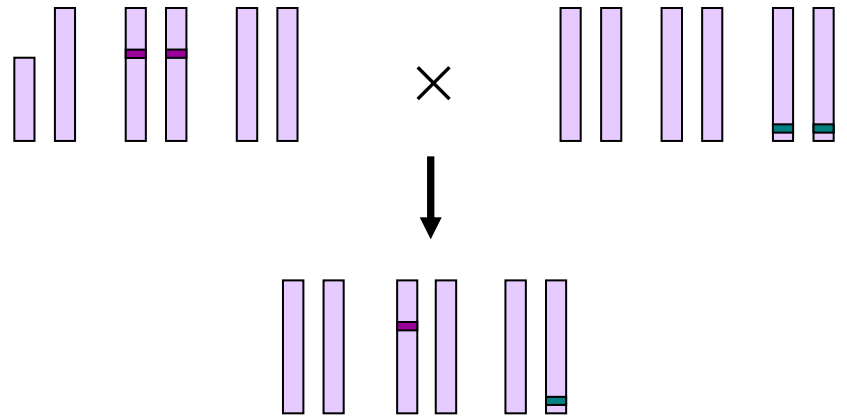
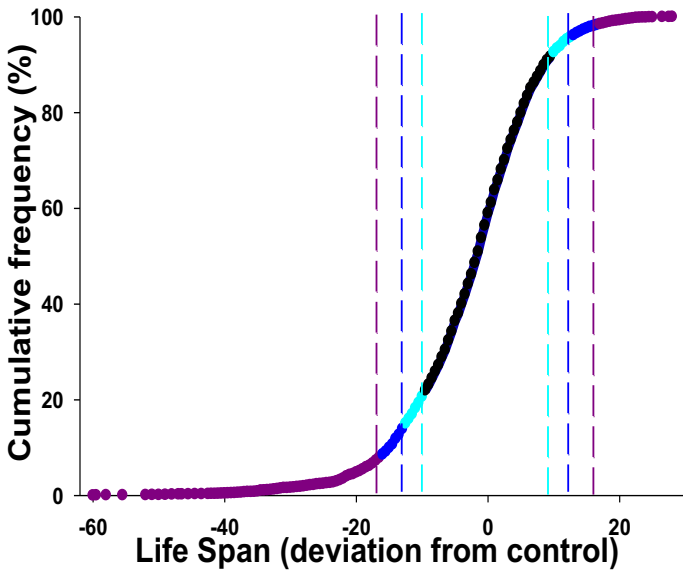
The difference in genetic
variance components between
the DGRP and outbred
populations derived from it
may enhance ability to detect
epistasis if it exists



Induced Mutations Also Exhibit Epistatic Interactions

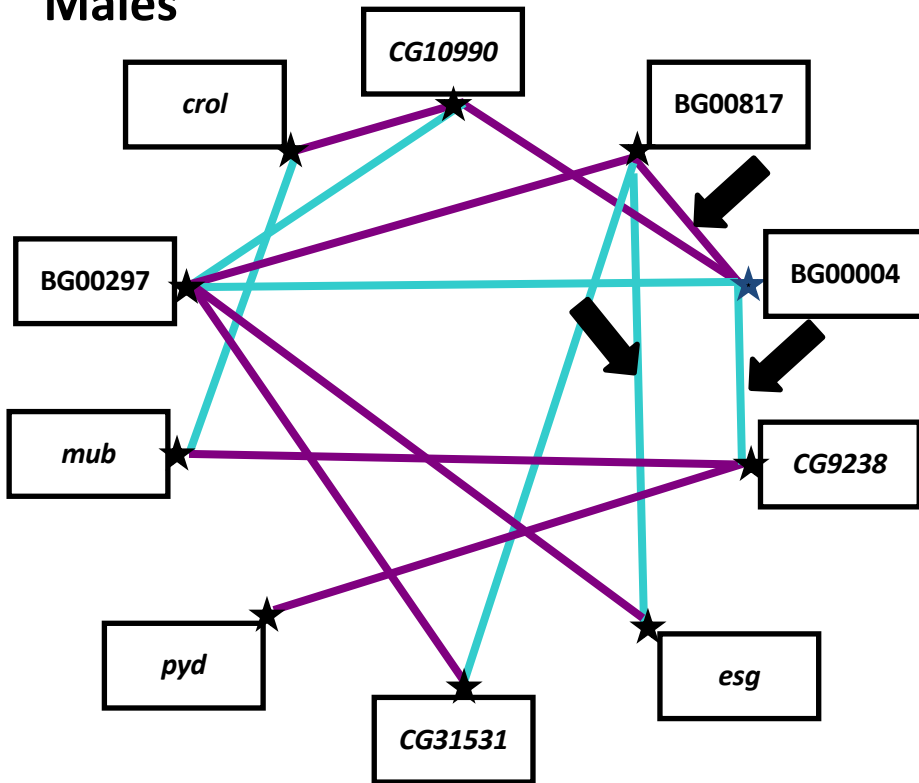


Magwire *et al.* 2010, *PLoS Genetics* 6: e1001037

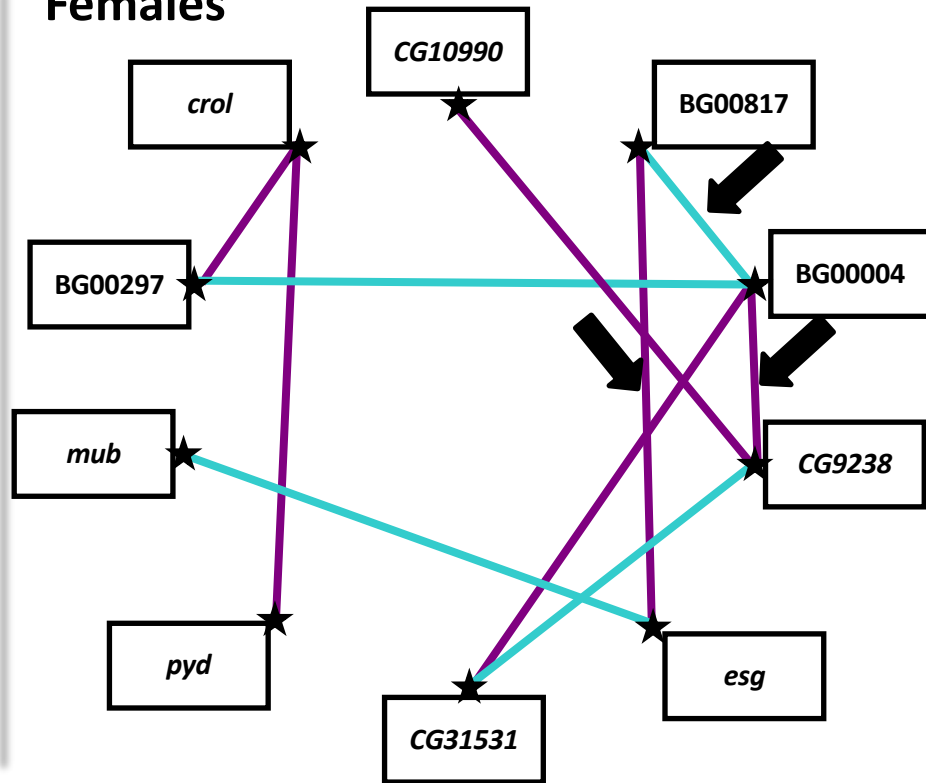


Induced Mutations Also Exhibit Epistatic Interactions

Males

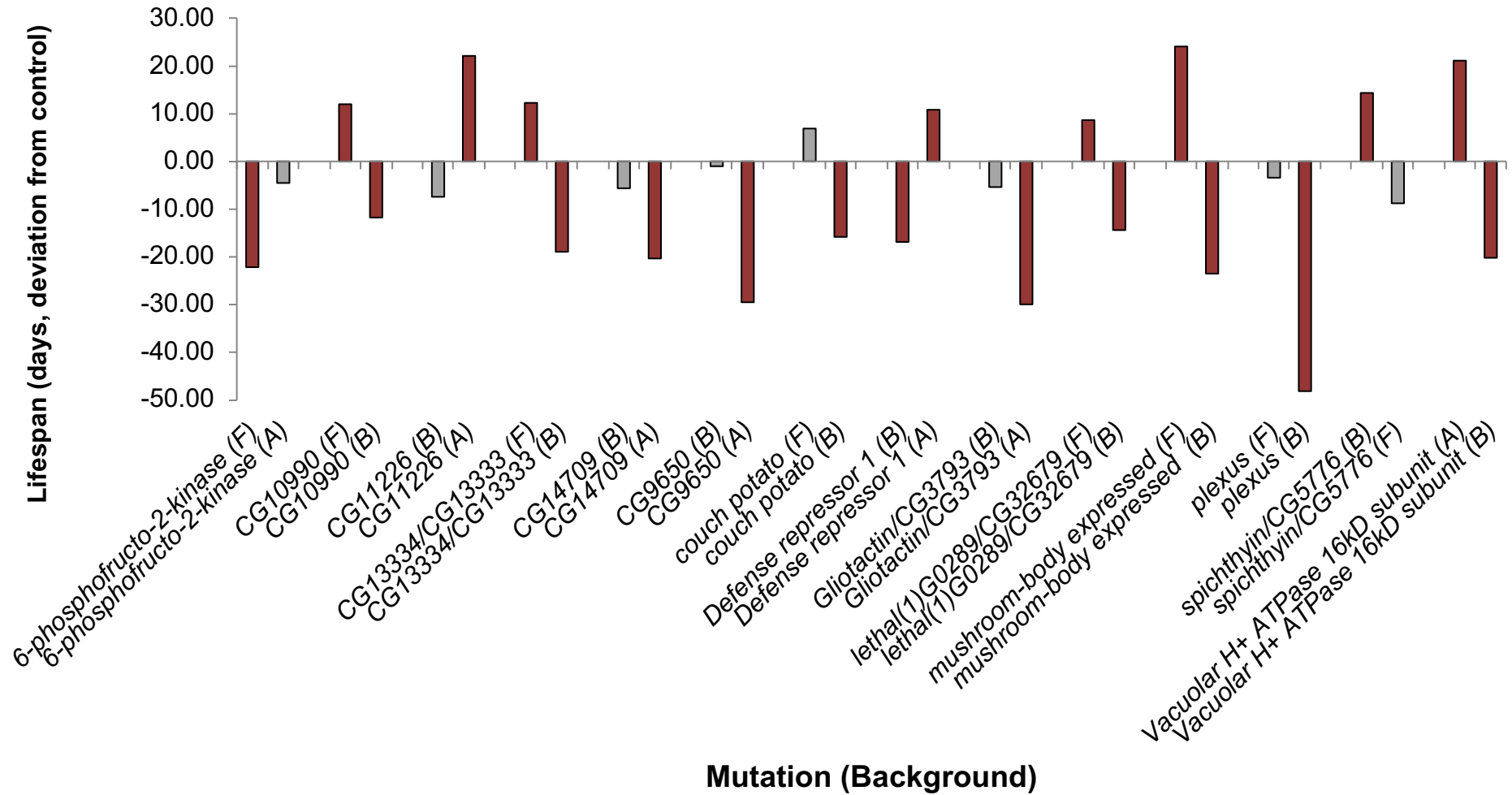


Females



Sex-specific epistasis

Induced Mutations Also Exhibit Epistatic Interactions





The genetic architecture of *Drosophila* lifespan is dominated by sex- and environment-specific variants as well as epistasis

These variants have small effects on lifespan, averaged over both sexes and all environments and may account in part for missing heritability

Effects of variants will not replicate between populations with different allele frequencies

These variants may also 'hide' from natural selection in natural populations experiencing heterogeneous environments, leading to maintenance of variation for lifespan in natural populations

THANK YOU!

Lifespan Measurements

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Epistasis: Effects at One Locus Depend on Allele Frequency Interacting Locus(i)

