ROSLIN

The use of imputed haplotypes to identify lethal recessive effects in pigs

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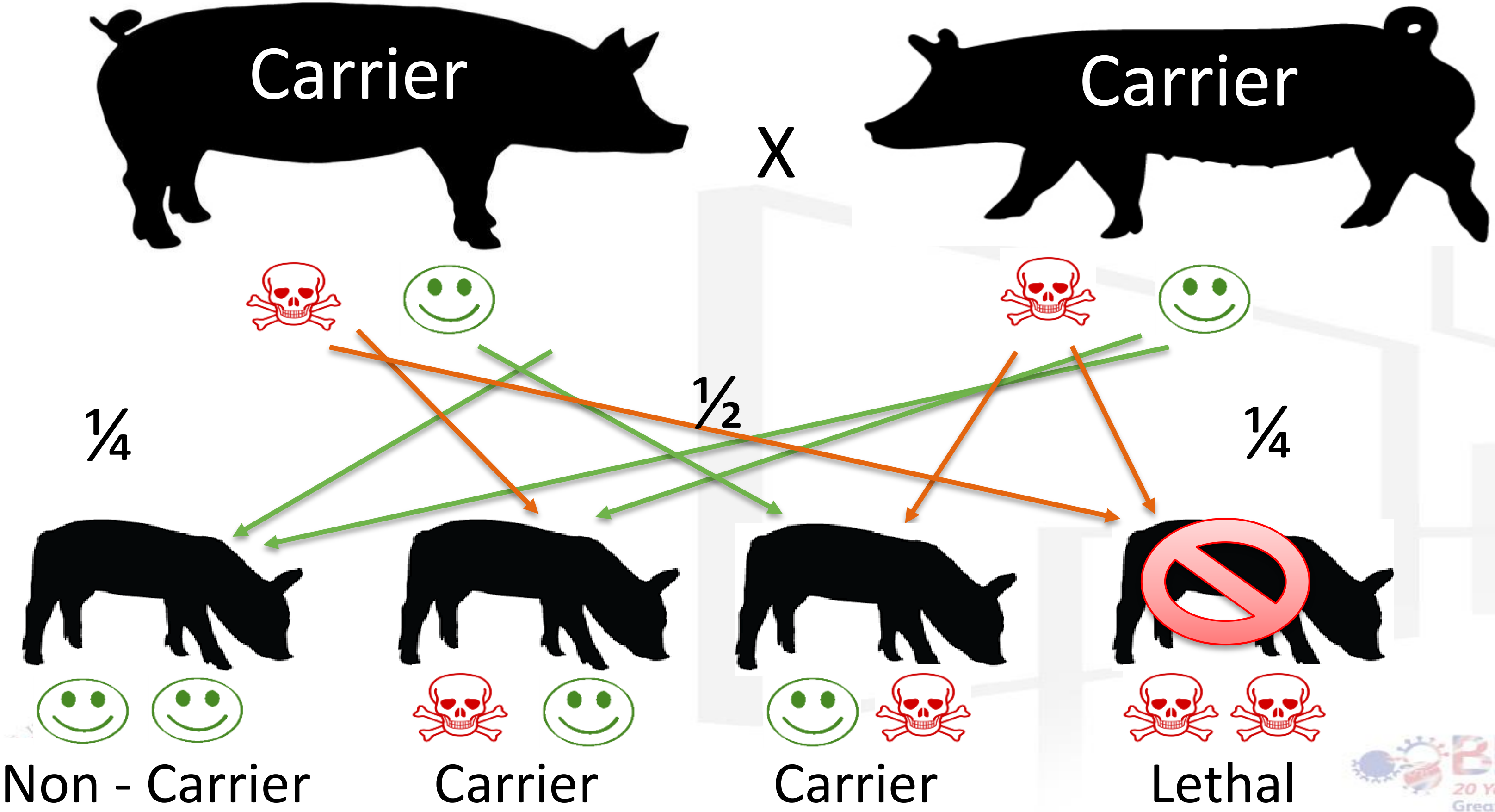
Background

Inheritance of Lethal Recessive Mutations



Carrier X Carrier

¼ ½ ¼

Non - Carrier

Carrier

Carrier

Lethal

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Detection Mutations with Haplotypes

- Haplotypes should increase power to detect lethal mutations
- Previous studies focussed on cattle, mice and humans

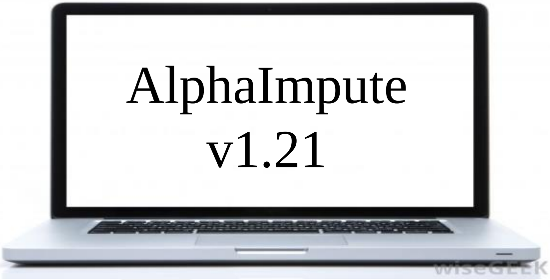
Materials

Materials

5,660 individuals genotyped @ high density (60k)

12,534 individuals genotyped @ low density (402)

23,327 pedigree records



AlphaImpute
v1.21

Hickey et al. (2011)

23,327 individuals haplotyped for 47,704 SNPs

Method

Method

Steps

1. Identify missing homozygous haplotypes
2. Identify candidate haplotypes with a potential lethal effect
3. Formal significance test of top candidate haplotypes

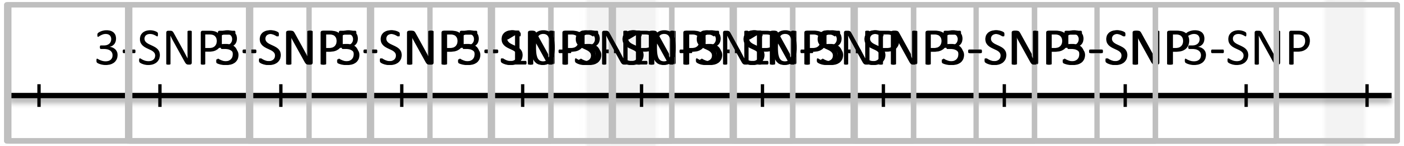
Identifying Missing Homozygotes

Steps

1. Identify missing homozygous haplotypes
2. Identify candidate haplotypes with a potential lethal effect
3. Formal significance test of top candidate haplotypes

Identifying Missing Homozygotes

- Using 3, 5 and 10-SNP length haplotypes, use a sliding window to examine the genome



- Identify those which are **never** homozygous

Preselection of Haplotypes

Steps

1. Identify missing homozygous haplotypes

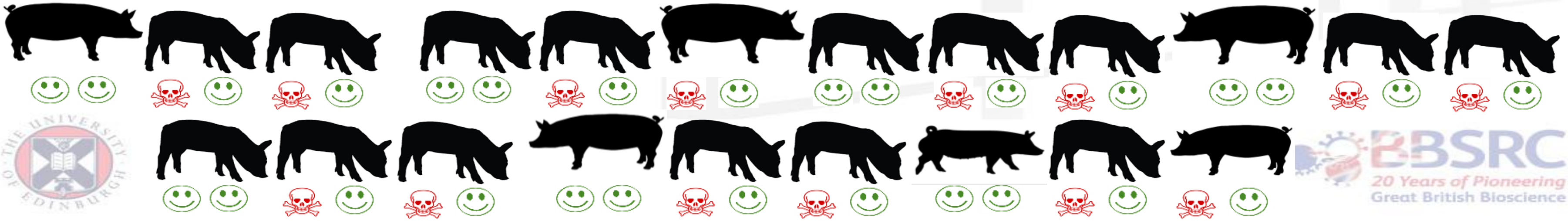
2. Preselect haplotypes with a potential lethal effect
   a. Population-wide Haplotype Frequency
   b. Within Family, Carrier x Carrier
      i. Sire x Dam
      ii. Sire x Maternal Grand Sire (MGS)

Preselection of Haplotypes

Population-wide
Haplotype Frequency

a. Probability of not observing any homozygotes given a haplotype frequency, q, in a population of size, N

$$P = e^{-(q^2 N)}$$

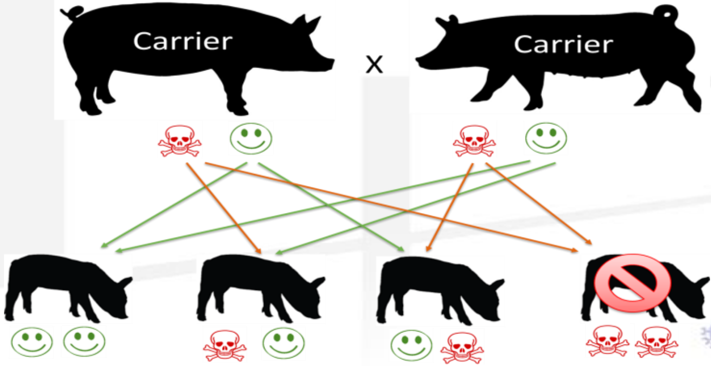


Preselection of Haplotypes

Within Family, Sire x Dam

b. i. Probability of not observing one of the homozygotes from all carrier sire x carrier dam matings, producing a total of n haplotyped offspring

$$P = (2 \text{ x } 0.75^n) - 0.5^n$$

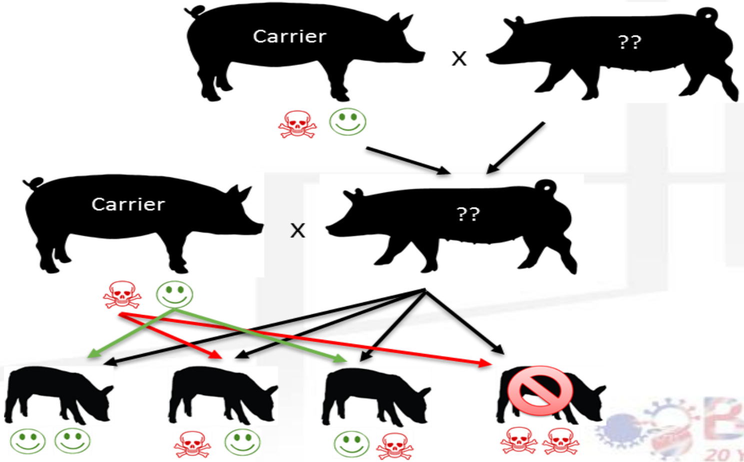


Preselection of Haplotypes

Within Family, Sire x MGS

b. ii. Probability of not observing a minor allele homozygote from all carrier sire x carrier MGS matings, where litter k produced n haplotyped offspring

$$P = \prod_{k=1}^{K}\left(\frac{0.5^{n_k}q + 0.75^{n_k} + (1-q)}{2}\right)$$



Final Preselection of Haplotypes

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- Using the 3 different methods, identify top 3 regions on each chromosome –
    - 3 X 3 X 18 chromosomes = 162 top regions
- Same regions highlighted across methods - 104 top regions
- Examine one haplotype in each of the top regions

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Formal Significance Testing

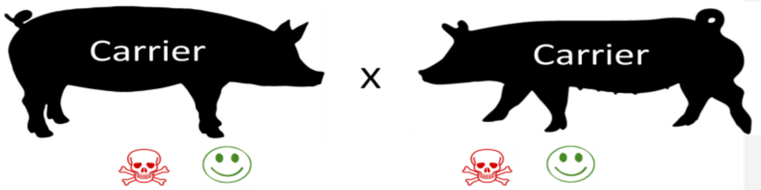
Steps

1. Identify missing homozygous haplotypes
2. Preselect haplotypes with a potential lethal effect
3. Formal significance test of top preselected haplotypes
    - ➢ Phenotypic Analysis Between Families

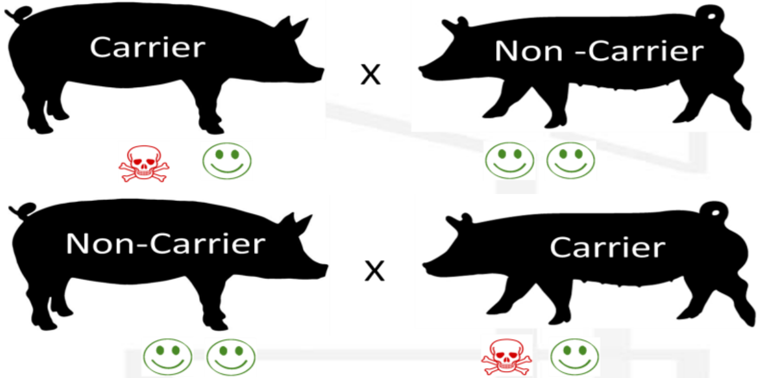
Significant Difference in Litter Size

Phenotypic Analysis, Sire x Dam

Compare Litter Size:



Vs.



(CxC)

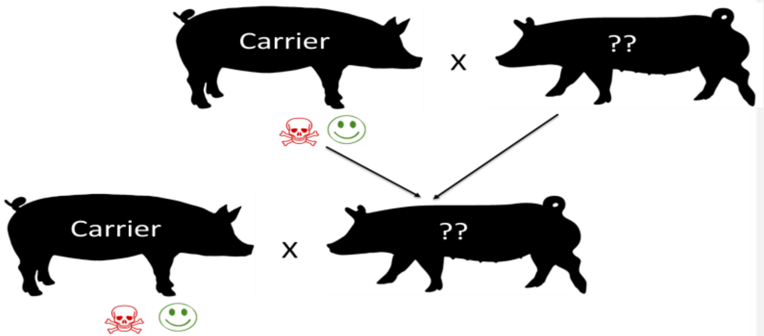
(CxNC)

- ➢ Dam parity, date of birth and pedigree relatedness

Significant Difference in Litter Size

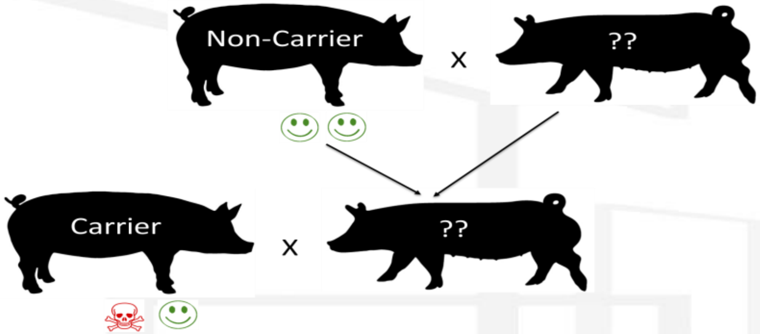
Phenotypic Analysis, Sire x MGS

Compare Litter Size:



(CxC)

Vs.



(CxNC)

- ➢ Dam parity, date of birth and pedigree relatedness

Reduction in Litter Size

Phenotypic Analysis

$$Reduction\ in\ litter\ size\ = \frac{CxNC\ \ - CxC}{CxNC}$$

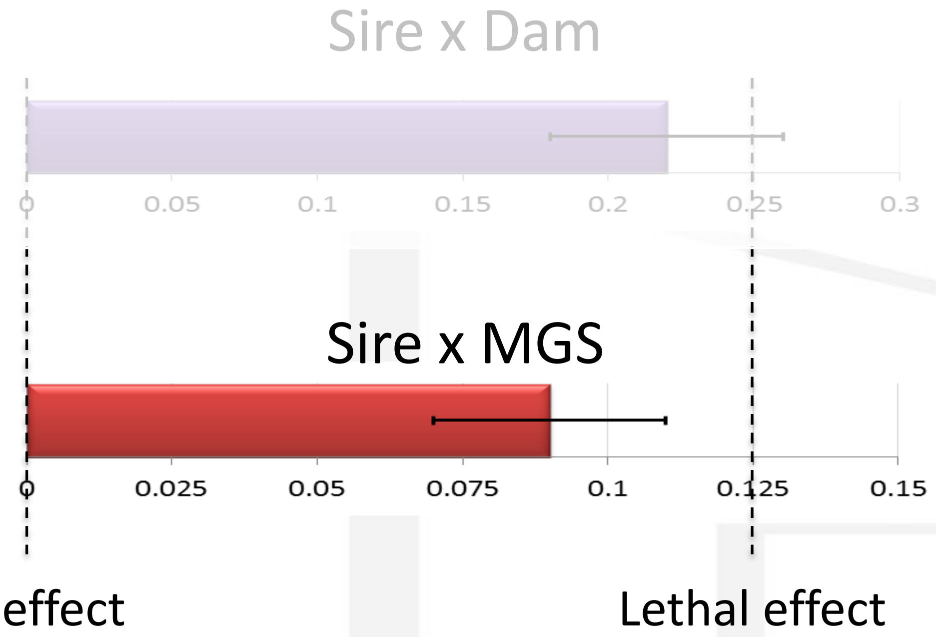
$$= \quad \frac{12 - 9}{12} \quad = \quad 0.25$$

$$95\%\ CI\ of\ the\ reduction = \pm\, 1.96 \sqrt{\left[\frac{\mu_A}{\mu_B}\right]^2 \left[\frac{\sigma_A^2}{\mu_A^2} + \frac{\sigma_B^2}{\mu_B^2} - \frac{2Cov(A,B)}{\mu_A\,\mu_B}\right]}$$

where A relates to (CxNC – CxC) and B relates to CxNC

Stuart et al. (1994)

Reduction Close to Expectation?



- Does the 95% CI overlap the expected lethal effect?

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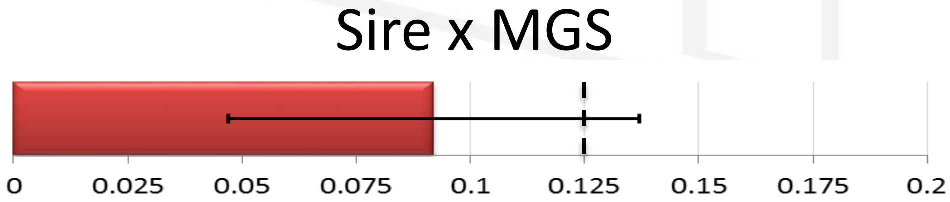
Results

SSC1, 137 Mb

Haplotype Frequency		
Haplotype Frequency	Expected Homozygotes	*P*
0.018	6.14	**2.15×10^{-3} ****

Phenotypic Analysis		
	CxC Litters	*P*
Sire x Dam	6	> 0.05
Sire x MGS	150	**6.99×10^{-5} ****

Within Family		
	Haplotyped Offspring	*P*
Sire x Dam	7	> 0.05
Sire x MGS	8	> 0.05



Sire x MGS

*	$p < 0.05$
**	$p < 0.01$
***	$p < 0.001$

SSC1, 187 Mb

Haplotype Frequency		
Haplotype Frequency	Expected Homozygotes	*P*
0.018	6.75	**1.17×10^{-3} ****

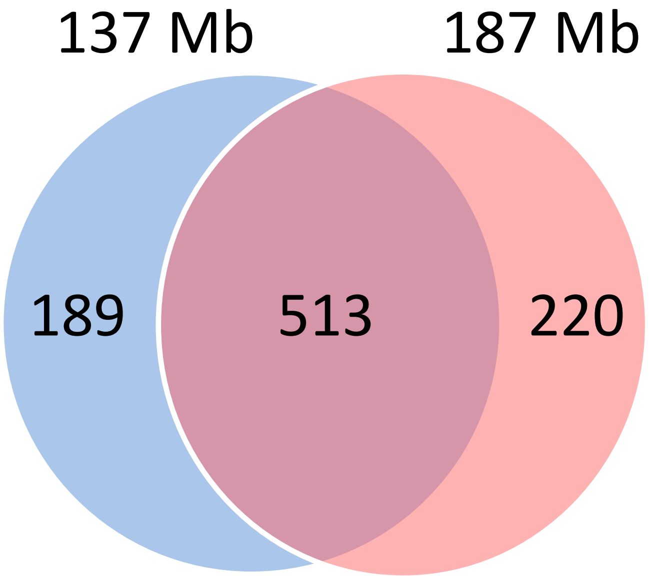
Within Family		
	Haplotyped Offspring	*P*
Sire x Dam	8	> 0.05
Sire x MGS	10	> 0.05

Phenotypic Analysis		
	CxC Litters	*P*
Sire x Dam	9	> 0.05
Sire x MGS	122	**7.11×10^{-3} ****

Sire x MGS

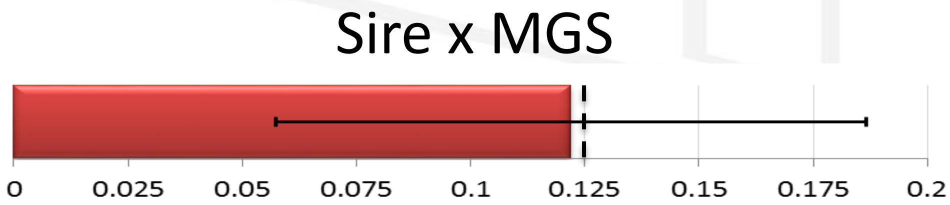
*	$p < 0.05$
**	$p < 0.01$
***	$p < 0.001$

SSC1, 137 & 187 Mb



Number of carriers

Phenotypic Analysis		
	CxC Litters	*P*
Sire x MGS	70	**2.07×10^{-4} *****



*	$p < 0.05$
**	$p < 0.01$
***	$p < 0.001$

SSC6, 24 Mb

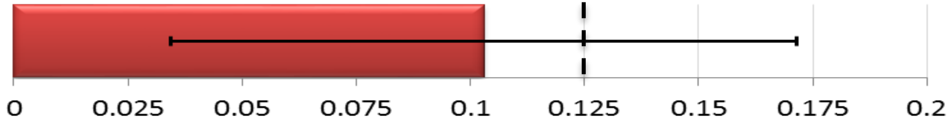
Haplotype Frequency		
Haplotype Frequency	Expected Homozygotes	*P*
0.020	7.15	**7.84×10^{-4} *****

Phenotypic Analysis		
	CxC Litters	*P*
Sire x Dam	8	0.056
Sire x MGS	72	**3.25×10^{-3} ****

Within Family		
	Haplotyped Offspring	*P*
Sire x Dam	16	**0.02 ***
Sire x MGS	7	> 0.05

Sire x Dam

Sire x MGS



*	$p < 0.05$
**	$p < 0.01$
***	$p < 0.001$

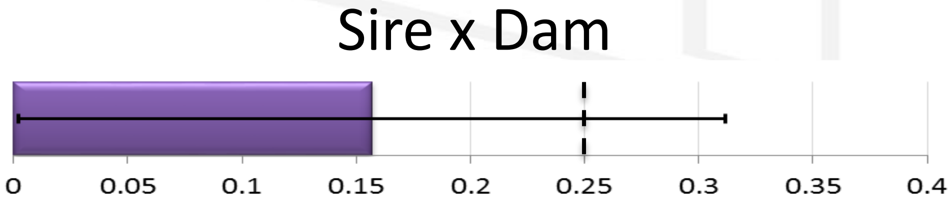
SSC10, 26 Mb

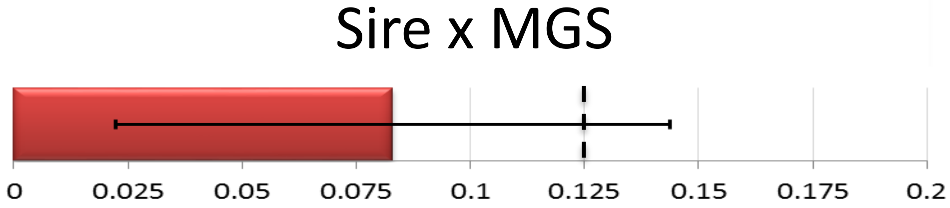
Haplotype Frequency		
Haplotype Frequency	Expected Homozygotes	*P*
0.016	5.24	**5.30×10^{-3} ****

Within Family		
	Haplotyped Offspring	*P*
Sire x Dam	22	**3.57×10^{-3} ****
Sire x MGS	97	**4.74×10^{-6} *****

*	$p < 0.05$
**	$p < 0.01$
***	$p < 0.001$

Phenotypic Analysis		
	CxC Litters	*P*
Sire x Dam	13	**0.048 ***
Sire x MGS	88	**7.69×10^{-3} ****


Sire x Dam


Sire x MGS

SSC14, 60 Mb

Haplotype Frequency

Haplotype Frequency	Expected Homozygotes	*P*
0.017	6.49	**1.77 x 10^{-3} ****

Within Family

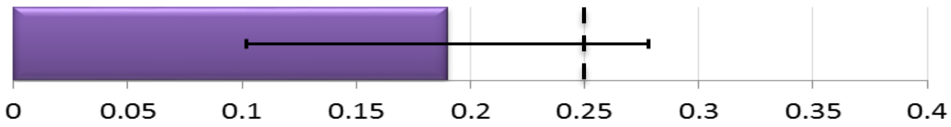
	Haplotyped Offspring	*P*
Sire x Dam	39	**2.68 x 10^{-5} *****
Sire x MGS	48	**8.67 x 10^{-3} ****

*	$p < 0.05$
**	$p < 0.01$
***	$p < 0.001$

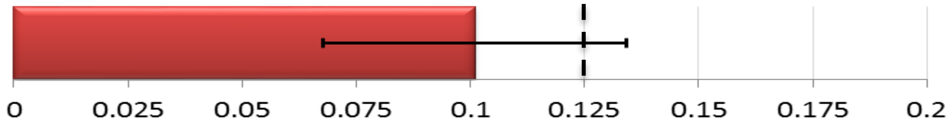
Phenotypic Analysis

	CxC Litters	*P*
Sire x Dam	32	**2.03 x 10^{-5} *****
Sire x MGS	292	**6.28 x 10^{-10} *****

Sire x Dam



Sire x MGS



SSC14, 117 Mb

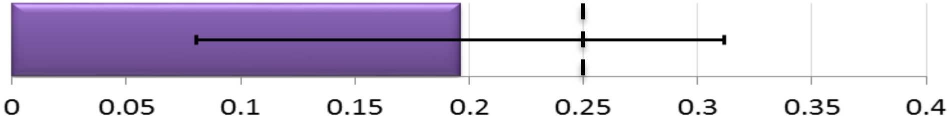
Haplotype Frequency		
Haplotype Frequency	Expected Homozygotes	*P*
0.015	4.2	**0.015 ***

Within Family		
	Haplotyped Offspring	*P*
Sire x Dam	18	**0.011 ***
Sire x MGS	42	**0.019 ***

*	$p < 0.05$
**	$p < 0.01$
***	$p < 0.001$

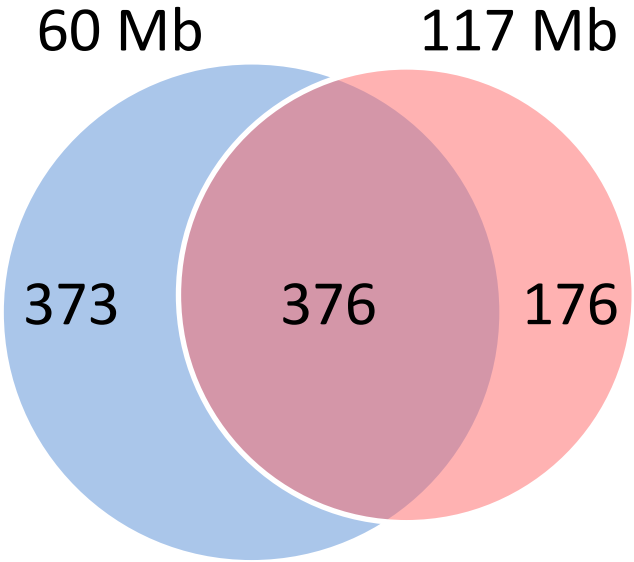
Phenotypic Analysis		
	CxC Litters	*P*
Sire x Dam	19	**9.71×10^{-4} *****
Sire x MGS	204	**1.01×10^{-5} *****

Sire x Dam



Sire x MGS

SSC14, 60 & 117 Mb

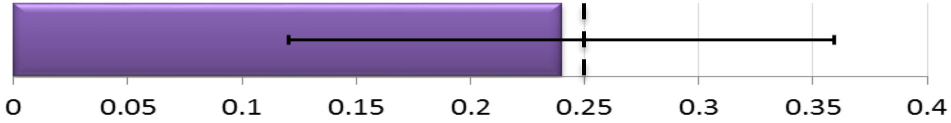


Number of carriers

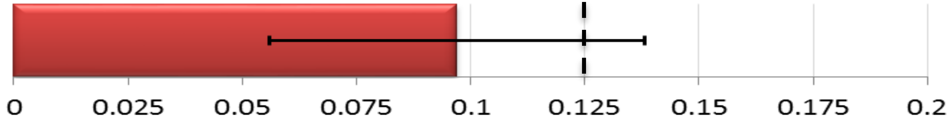
*		$p < 0.05$
**		$p < 0.01$
***		$p < 0.001$

Phenotypic Analysis		
	CxC Litters	*P*
Sire x Dam	17	**1.02×10^{-4} *****
Sire x MGS	178	**2.97×10^{-6} *****

Sire x Dam



Sire x MGS



Conclusion

- Evidence of putative lethal recessive mutations affecting litter size on SSC1, SSC6, SSC10 and SSC14

- Further work – sequence these regions

General Remarks

- Robust framework for identifying carriers of putative lethal recessive mutations
- Reproductive performance improved by avoidance of carrier x carrier matings

Acknowledgements

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John Woolliams
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Genus

PIC

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Pieter Knap
Dave McLaren

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