

GENOME WIDE ASSOCIATION STUDY OF INSECT BITE HYPERSENSITIVITY IN TWO POPULATION OF ICELANDIC HORSES

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WAGENINGENUR

For quality of life

Acknowledgement

- EUROPEAN GRADUATE SCHOOL OF ANIMAL BREEDING AND GENETICS (EGS-ABG)



Objective

- To improve identification and quantification of the chromosomal regions associated with insect bite hypersensitivity (IBH)



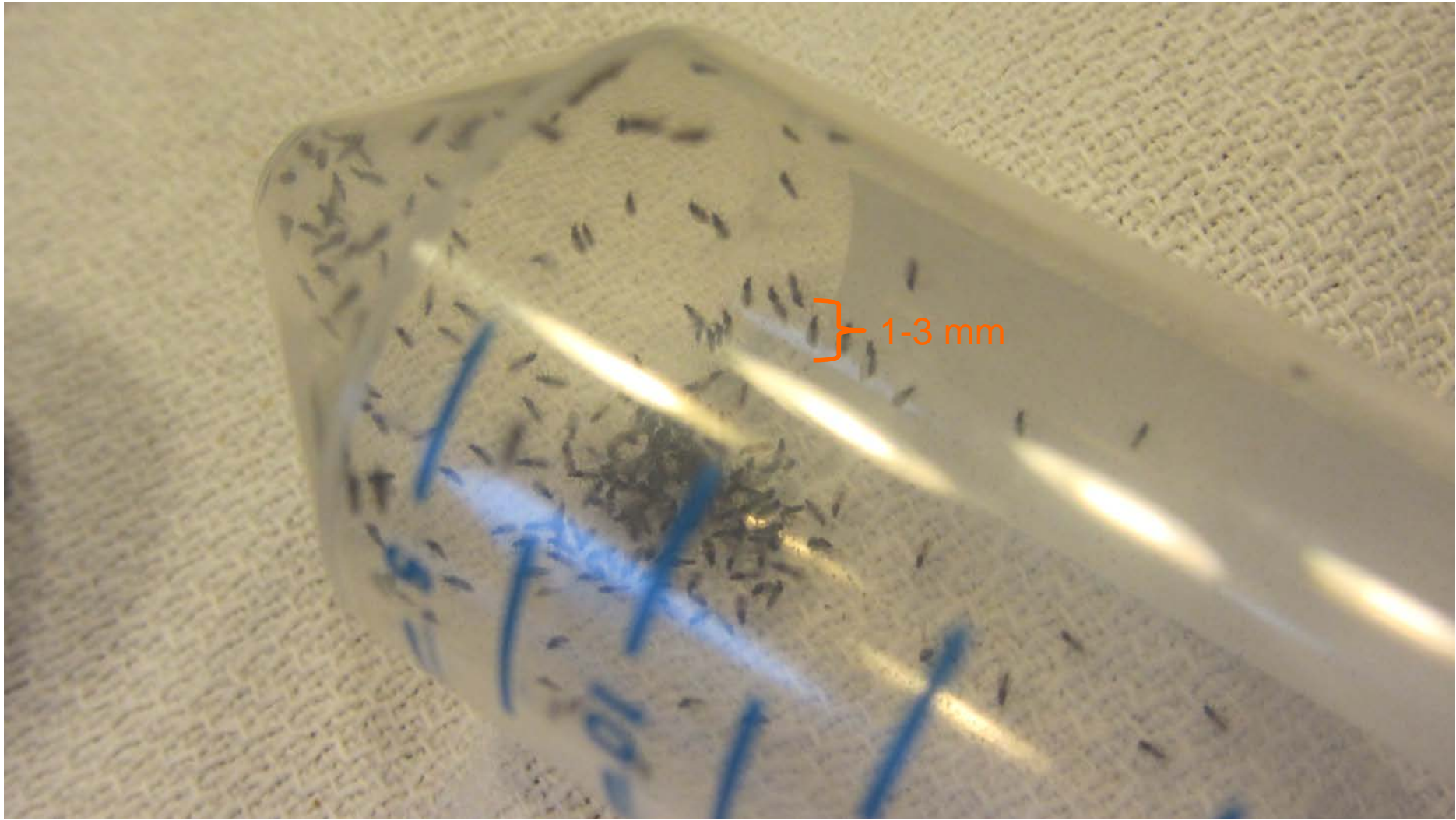
Photo courtesy: Anouk Schurink

Introduction

- Most common skin allergy
- Seasonal, chronic allergic reaction to biting midges especially *Culicoides* species
- Immunoglobulin (Ig) E type I hypersensitivity reaction
- Multifactorial (environment and genetic)
- Prevalence : 3% to 60%
- Heritability: 0.16 to 0.33
- Age at onset: 2 to 4 years
- Preventive measures



Photo courtesy: Anouk Schürink



Culicoides species

Introduction

- Intensely pruritic, hair loss, secondary infections
- Serious welfare problem
- Economic loss



Photo courtesy: Anouk Schurink



Introduction

Previous studies:

Immunogenetics (2012) 64:201–208
DOI 10.1007/s00251-011-0573-1

ORIGINAL PAPER

The same ELA class II risk factors confer equine insect bite hypersensitivity in two distinct populations

Lisa S. Andersson · June E. Swinbune · Jennifer R. S. Meadows · Hans Broström ·
Susanne Eriksson · W. Freddy Fikse · Rebecka Frey · Marie Sundquist ·
Chia T. Tseng · Sofia Mikko · Gabriella Lindgren

Schurink *et al.* *Genetics Selection Evolution* 2012, **44**:31
<http://www.gsejournal.org/content/44/1/31>



RESEARCH

Open Access

Genome-wide association study of insect bite hypersensitivity in two horse populations in the Netherlands

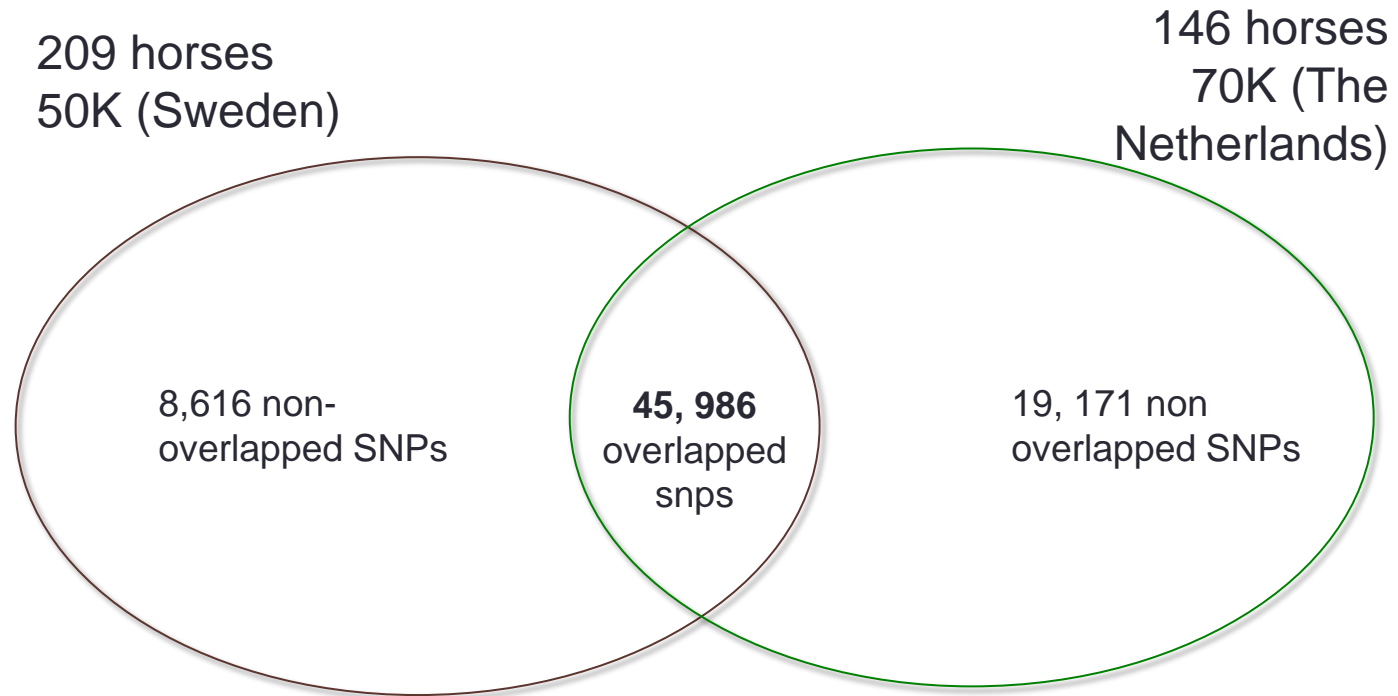
ECA 3, 7, 11, 20, 23

Anouk Schurink^{1*}, Anna Wolc^{2,3}, Bart J Ducro¹, Klaas Frankena⁴, Dorian J Garrick³, Jack CM Dekkers³
and Johan AM van Arendonk¹

Introduction

- Previous studies:
- Immune and allergy related non-MHC (Major Histocompatibility Complex) genes as:
 - CD14 receptor (CD14)
 - interleukin 23 receptor (IL23R)
 - transforming growth factor beta 3 (TGFB3)
 - thymic stromal lympoietin (TSLP)

Materials and Methods



- 355 horses
- 45,986 overlapped single nucleotide polymorphism (SNPs)

Materials and Methods

- Paternal half sib design

	Affected (177)	Unaffected (178)	Total
Male	75	82	157
Female	102	96	198
Age Mean (SD)	12.8 (4.8)	12.4 (5.2)	12.6 (5.0)
Age Range	4-29	4-35	4-35
No. of Sires	95	101	196
No. of Dams	172	172	344

Materials and Methods

355 horses
45, 986 SNPs

GenABEL package (v. 1.7-6) in R (v. 3.0.1)

Quality control :

- SNP call rate: 95%
- Individual call rate: 95%
- Minor allele frequency (MAF) : 5%
- Hardy Weinberg Equilibrium : 1×10^{-8}

Analysis of population stratification

- Identity by state (IBS), genomic kinship matrix

$$f_{i,j} = \sum_k \frac{(x_{i,k} - p_k)(x_{j,k} - p_k)}{(p_k(1 - p_k))}$$

In a pair: i = first animal , j = second animal

$X_{i,k}$ = genotype (0, 1/2 or 1) of "i" animal for SNP

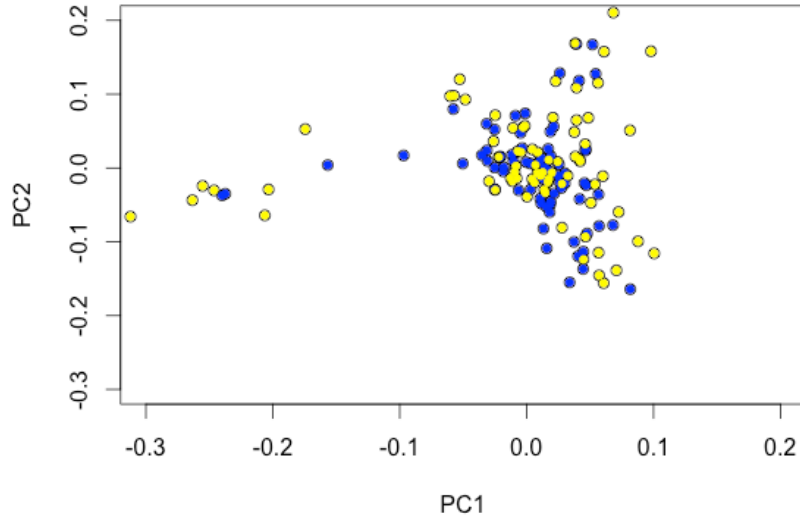
"k"

p_k = frequency of allele top strand allele

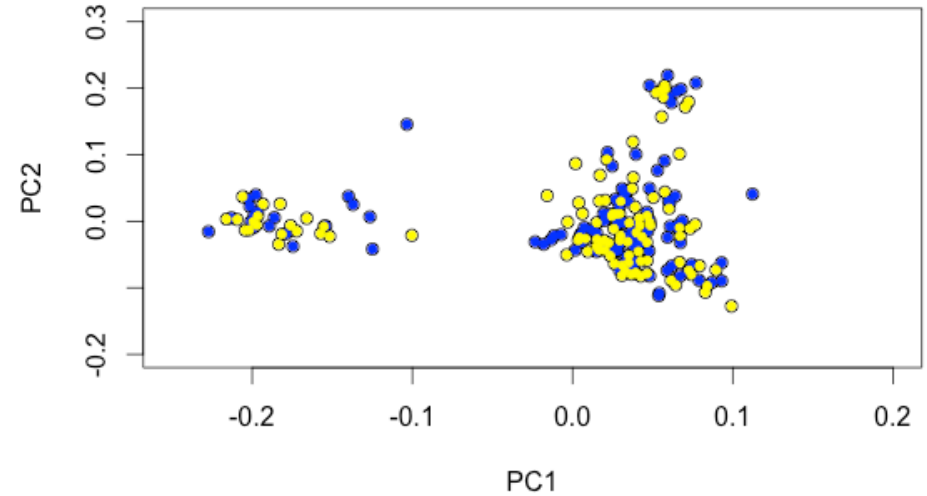
355 horses
31, 997 SNPs

Multi-Dimensional Scaling plot

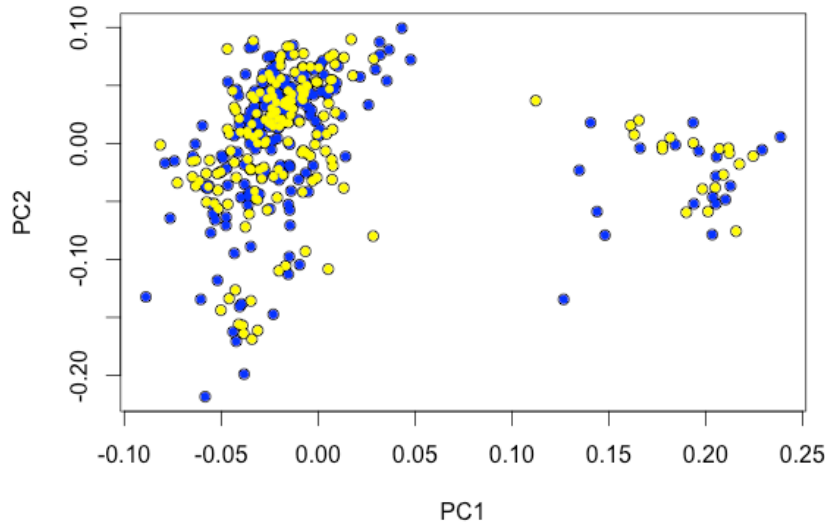
146 horses



209 horses



355 horses

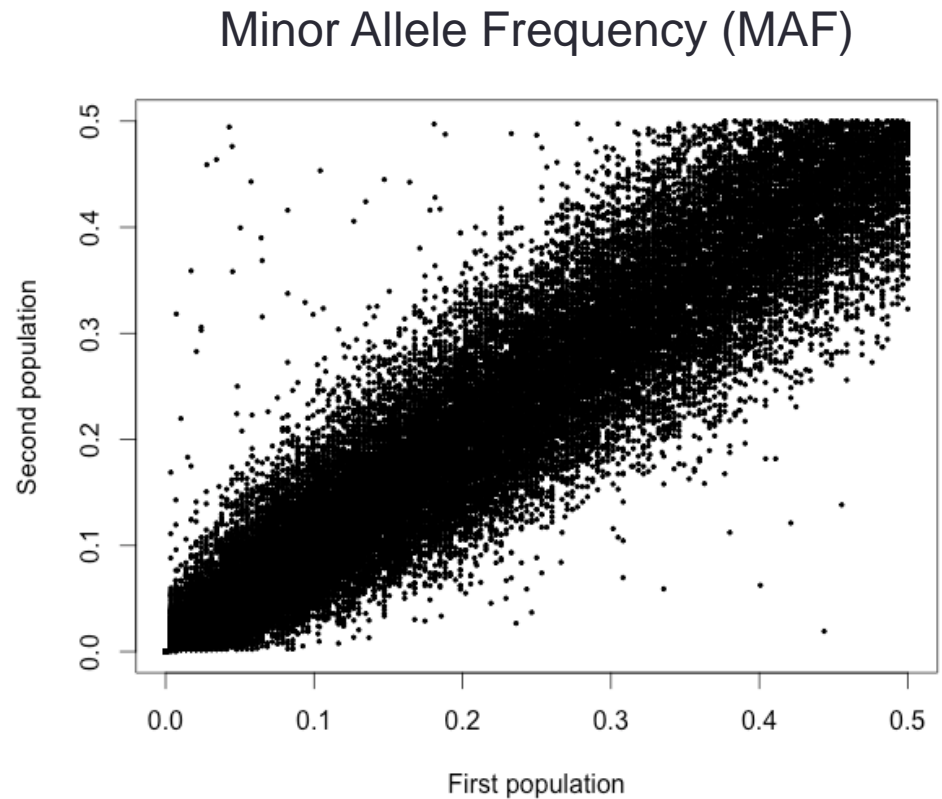


Unaffected horses
Affected horses

PC1 : First principal component
PC2 : Second principal component

Materials and Methods

- **Population Stratification:**
- 43, 006 same allele was a minor allele in both populations
- 1, 618 markers had different allele as a minor allele in two populations
- Genomic control inflation factor (λ), ≈ 0.92 ($se = 1 \times 10^{-4}$)
- Case-control association studies
- 355 horses; 31, 997 SNPs



Materials and Methods

- Association Analysis

- GenSel software (<http://biggs.ansci.iastate.edu/>)
- Bayesian Variable Selection method (Bayes C)

$$\eta = \mu + \sum_{j=1}^K Z_j u_j \delta_j$$

• η = linear predictor related with phenotype through probit link function

μ = mean; K = number of SNPs

At SNP “j”:

Z_j = column vector represents genotype covariate (AA= -10, AB= 0, BB= 10, missing genotypes = average value of SNP)

u_j = random allele substitution effect (normally distributed $\sim N(0, \tilde{\sigma}_u^2)$, $u_j = 0$ when $\delta_j = 0$)

δ_j = random 0/1 variable indicating presence of SNP (probability $\tilde{\lambda}$) or absence of SNP (probability $1-\tilde{\lambda}$) in the model

Results

Windows associated with IBH					Allele frequency of unfavorable allele			
CHR	Mb	No. Of SNPs	%Var	p>Average	SNP	ModelFreq	Affected	Unaffected
1	152	19	0.54	5.5	BIEC2_65299	0.0127	0.69	0.58
1	153	14	1.05	7.0	BIEC2_65455	0.0504	0.82	0.69
3	35	17	0.44	5.1	BIEC2_776764	0.0134	0.57	0.45
4	100	18	0.62	4.1	BIEC2_878810	0.0321	0.57	0.44
6	24	19	0.35	4.5	BIEC2-945312	0.0056	0.46	0.35
9	0	14	0.33	4.0	BIEC2_1067431	0.0102	0.76	0.65
11	0	13	0.54	5.1	BIEC2_133565	0.0146	0.32	0.21
16	66	12	0.32	2.9	BIEC2_356619	0.0107	0.68	0.57
18	32	21	0.39	3.8	BIEC2_410288	0.012	0.43	0.32
19	55	17	0.35	3.6	BIEC2-446571	0.0168	0.84	0.73
22	25	12	0.65	4.6	BIEC2_589497	0.0382	0.79	0.65

p>Average :Percentage of iterations where the window explained more than 0.043% of genetic variance that is an expected variance explained by each window ($100/2317 = 0.043\%$; 2317 non-overlapping number of windows)

Model frequency: proportion of total post burn-in iterations where SNP was included in the model

Results

BIEC2_65455 CHR1 153 MB

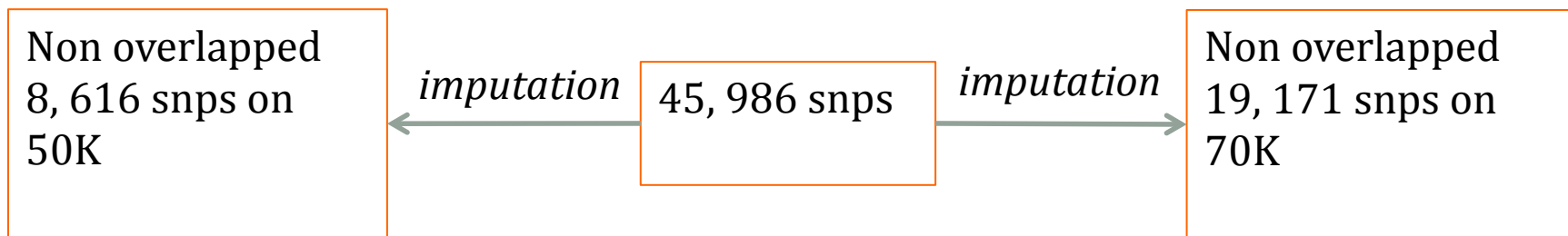
355 horses				Allele frequency		Total frequency	
Genotype	A/A	A/G	G/G	A	G	A	G
Unaffected	87	73	18	0.69	0.31	0.76	0.24
affected	118	55	4	0.82	0.18		
Total	205	128	22				

209 horses							
Genotype	A/A	A/G	G/G	A	G	A	G
Unaffected	60	42	3	0.77	0.23	0.83	0.17
affected	82	20	2	0.88	0.12		
Total	142	62	5				

146 horses							
Genotype	A/A	A/G	G/G	A	G	A	G
Unaffected	27	31	15	0.58	0.42	0.66	0.34
affected	36	35	2	0.73	0.27		
Total	63	66	17				

Discussions

- Overlapping regions with previous studies on chromosomes
 - 1, 3, 4, 7, 15, 18
- Number of markers
- Further analysis with Imputation [BEAGLE (version 3.3)]



- Population structure within reference data set
- Association analysis taking into account imputed SNPs

**THANK YOU
QUESTIONS/ COMMENTS???**