The genetic basis of breed diversification: signatures of selection in pig breeds

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Breed diversification

• Domestication and selective breeding has produced a diversity of pig breeds

• What is the genetic architecture underlying this variation?
History of UK pig breeds

Unique and interesting history of selective breeding

1. Selective breeding from 18th century
   - Breeds-crossed, selection for fat

2. Introgression from Asian breeds
   - Growth, prolificacy, early maturing

   contemporary breeds formed

3. Pig breed societies set up
   - Breeds kept distinct

4. Further selection in 20th century
   - Reproduction, leanness, muscularity, growth
   - Specialisation

“consequence of so much crossing, some well-known breeds have undergone rapid changes; thus, according to Nathusius, the Berkshire breed of 1780 is quite different from that of 1810” (Darwin 1868).
Strong and recent genetic change

- Resultant UK pig breed diversity
  - Numerous breeds
  - Broad and distinct phenotypic diversity
  - High breed genetic differentiation
  - Many commercial breeds originated from UK

- Valuable model to study the genetic basis of phenotypic variation that arose from selection
Selection

- Theory: regions under selection display patterns that depart from neutral expectations
Selection mapping

• Measuring population genetic differentiation ($F_{ST}$)

\[ F_{ST} = \frac{\text{var}(p)}{p(1-p)} \]

• With ‘000s SNPs - can scan genome of populations for signatures of diversifying selection
Detect signatures of selection associated with pig breed diversification by mapping breed genetic differentiation

QUESTIONS:

1. Do signatures of selection overlap regions already associated with phenotypic traits (QTLs and genes)?
2. What types of traits are associated with the signatures of selection?

What does this tell us about the history of selective breeding and genetic characteristics underlying phenotypic diversity in pig breeds?
Porcine Data

- SNPs (PorcineSNP60 chip: 60,000)
  - 13 UK/Europe breeds - 372 individuals
  - 1 Asian breed

- Sequence (10x coverage, Illumina)
  - 12 UK/Europe breeds - 52 individuals
  - 8 Asian breeds - 24 individuals
Analysis pipeline

After quality control 49,260 SNPs remained

For each breed, locus-by-locus $F_{ST}$ estimated

For each breed, moving average estimated using 13-SNP sliding window

For each breed, a 99th percentile was imposed on the $F_{ST}$ distribution

*Breed specific signatures of diversifying selection*
RESULTS
Coat colour: *KITLG*

in BERKSHIRE: black animal with 6 white points

- *KITLG* is involved in melanocyte production
  - Investigated for role in pig colouration (Hadjiconstantouras et al. 2008; Okumura et al., 2010)
  - Pigmentation in mice, humans
Variation at *KITLG*

Differentiation region on SSC5 (98.0-99.0 Mb) searched for sequence variants unique to Berkshire

- Key variants found:
  - 1 SNP on the 3’-UTR of *KITLG* in Berkshire
    - not in other European breeds; in 1 Asian breed, the Jiangquahai
  - 2 non-synonymous variants in *KITLG* in Berkshire
    - 3/50 European and 16/24 Asian individuals, respectively
      - Similar results by Okamura et al (2008)

- An Asian origin of *KITLG* in Berkshire

- *MC1R* implicated in Berkshire coat phenotype
  - $E^P$ allele: 2bp insertion

- Interactions between the different coat loci?
Coat colour: *EDNRB*

In GLOUCESTERSHIRE OLD SPOTS: white animal with black spots

- **EDNRB** involved in melanocyte development
  - White coat-spotting (mice, horses)

Signal of $F_{ST}$: 53.5 – 55.5 Mb
### Variation at **EDNRB**

#### Key variants found:
- 2 non-synonymous changes in the exon of **EDNRB**

**Genetic differentiation region on SSC11 (53.5-55.5 Mb) searched for sequence variants unique to Gloucestershire Old Spots**

<table>
<thead>
<tr>
<th>Residue 17</th>
<th>European breeds</th>
<th>Asian breeds</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leucine</td>
<td>Gloucestershire Old Spots</td>
<td>Xiang, Jiangquahai</td>
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<tr>
<td>Phenylalanine</td>
<td>All other European breeds</td>
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**Signal peptide**

**N-terminal extracellular domain**
Mutations in \textit{EDNRB} lead to reduced expression or partial/complete loss-of-function

- Hirschsprung’s disease
EDNRB and pig spotting

• **MC1R** – $E^P$ allele: 2bp insertion
  - Other European coat phenotypes also associated with $E^P$ allele
  - At low frequency or absent in Asian breeds

• Melanocortin – Endothelin signalling
  - Complex melanocortin-endothelin signalling in cats (Kaelin et al 2012)
  - **MC1R** variant interacts with partial loss-of-function **EDNRB** to give Gloucestershire Old Spots its spots?
### Ear phenotype variation

<table>
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<tr>
<th>Prick-eared breeds</th>
<th>Intermediate-eared breeds</th>
<th>Flat-eared breeds</th>
</tr>
</thead>
<tbody>
<tr>
<td>Berkshire</td>
<td>Duroc</td>
<td>British Saddleback</td>
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<tr>
<td>Hampshire</td>
<td>Landrace</td>
<td>Gloucestershire</td>
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<tr>
<td>Large White</td>
<td>Welsh</td>
<td>Old Spots</td>
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<tr>
<td>Middle White</td>
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<td>Large Black</td>
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<tr>
<td>Pietrain</td>
<td></td>
<td>Mangalica</td>
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<tr>
<td>Tamworth</td>
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</tbody>
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- Assess genetic divergence between the different ear phenotypes:

\[ F_{ST} : \]

- Prick-eared breeds vs flat-eared breeds
- Prick-eared breeds vs intermediate-eared breeds
- Intermediate-eared breeds vs flat-eared breeds
Signals for ear phenotype

- Significant QTL on SSC7 overlaps the signal
- Significant QTL on SSC5 is ~10Mb upstream of the signal
Signal on SSC5

Associated with contrast between prick or intermediate ears and large flat ears

- Syntenic to a region in the dog genome associated with ear morphology (Boyko et al. 2010, Vaysse et al. 2011)
Variation at SCC5 signal

Differentiation region on SSC5 (31.0-34.0 Mb) searched for variants shared by flat-eared breeds vs prick-eared breeds:

- No non-synonymous differences
- Variants in non-protein coding sequence
  - SNPs in regulatory elements or miRNA genes may be responsible?
Selection mapping of QTLs in DUROC

Fatty acid composition QTL in Duroc (Uemoto et al 2012)

GWAS study found a significant SNP in this region (Yang et al 2013)

Genes with a role of fatty acid synthesis found in the region: SCD (120.90-121.30 Mb); EVOLVL3 (123.08-123.083 Mb)

- Duroc has a unique meat quality composition
  - high intramuscular fat content
  - high concentrations of saturated and mono-unsaturated fatty acids
Conclusions

• Pig breeds display signatures of selection associated with highly visible phenotypic differences
  
  • Gloucestershire Old Spots and Berkshire selected for distinct coat phenotypes
    – *EDNRB* amino acid differences
    – *KITLG* variation
  
  • 3 genomic regions are associated with ear phenotype variation in pigs

• Influence of Asian alleles on the UK phenotypic diversity
  
  • Asian breeds influenced highly visible phenotypic differences as well as production traits
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Thank you ... Questions?

• Journal article on this study:


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