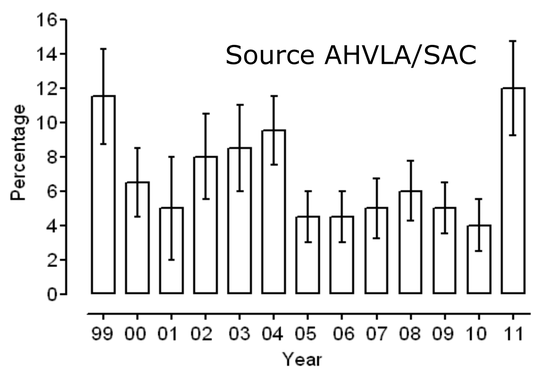
Genetics of cobalt deficiency in sheep

AH Brassington[1], J Xu[1], Clare CE[1], O Matika[2], WY Kwong[1], D Wragg[2], NR Kendall[1],R Talbot[2], DA Barrett[1], AL Archibald[2], RD Emes[1] and KD Sinclair[1]

[1]University of Nottingham, Sutton Bonington, Leicestershire, LE12 5RD
[2]Roslin Institute, University of Edinburgh, Easter Bush, Edinburgh, EH25 9RG

Introduction

- Trace element deficiencies such as cobalt can impair animal productivity, fertility and health.
- Cobalt (Co) is an essential micronutrient in ruminant diets required for the intra-ruminal synthesis of vitamin B_{12}
- Susceptibility to Co deficiency varies with respect to:
  - (i) Geographical location
  - (ii) Diet
  - **(iii) Genetics**
- Prevalence of Co deficiency in UK: **Mean 7%**



Hypothesis

Inter-individual and inter-breed variability in susceptibility to cobalt deficiency arises due to single-nucleotide polymorphisms (SNPs) in genes that regulate:

- B_{12} absorption and storage
- B_{12} metabolic and epigenetic pathways

Goals

- Identify novel genetic variants and thereby contribute to available genomic resources
- Develop a SNP chip to identify strains of sheep sensitive to Co/B_{12} deficiency
- Improved diagnosis and treatment of Co deficiency in sheep
  - Novel strategies for targeted Co/B_{12} supplementation
- Ability to select for Co/B_{12} tolerance within breeding programmes

Approach

1. SNP discovery

Identify SNPs in genes encoding enzymes involved in B_{12} related metabolic pathways

2. Functional significance of SNPs

Identify SNPs leading to modifications in B_{12} metabolism in sheep (Fig.1)

- Genotyping
- Comprehensive quantitative metabolic profiling
- Bioinformatic analysis to identify functionally significant SNPs

3. Proof of concept study

Demonstrate that selection on the basis of identified SNPs leads to improvements in lamb feed intake and growth

- 90 pure-bred Texels
- Fed a cobalt and sulphur deficient diet
- Plasma/serum measurements of B12 (Fig.2A) and key intermediary metabolites (Fig.2B and C)
  - Homocysteine (Hcy)
  - Methylmalonic acid (MMA)
- Metabolomic and bioinformatic analysis of liver samples
- Confirm and extend list of functionally important SNPs
- Retrospectively identify genetically sensitive lambs
- Produce list of allele variants associated with cobalt deficiency
- Identify 'low-risk' and 'high-risk' allele combinations for cobalt deficiency

4. Breed comparison

- Nasal swab collection of DNA from ~200 sheep
- 3 British breeds
  - Suffolk (61)
  - Bluefaced Leicester (59)
  - Swaledale (70)
- Genotyping and phylogenetic analysis (Fig.3)
- Between and within breed comparison of SNPs
- Prevalence of 'risk' allele combinations within breeds

Preliminary findings

- **26** functionally significant SNPs in **15** metabolic genes
- **21** functionally significant SNPs in **12** epigenetic genes

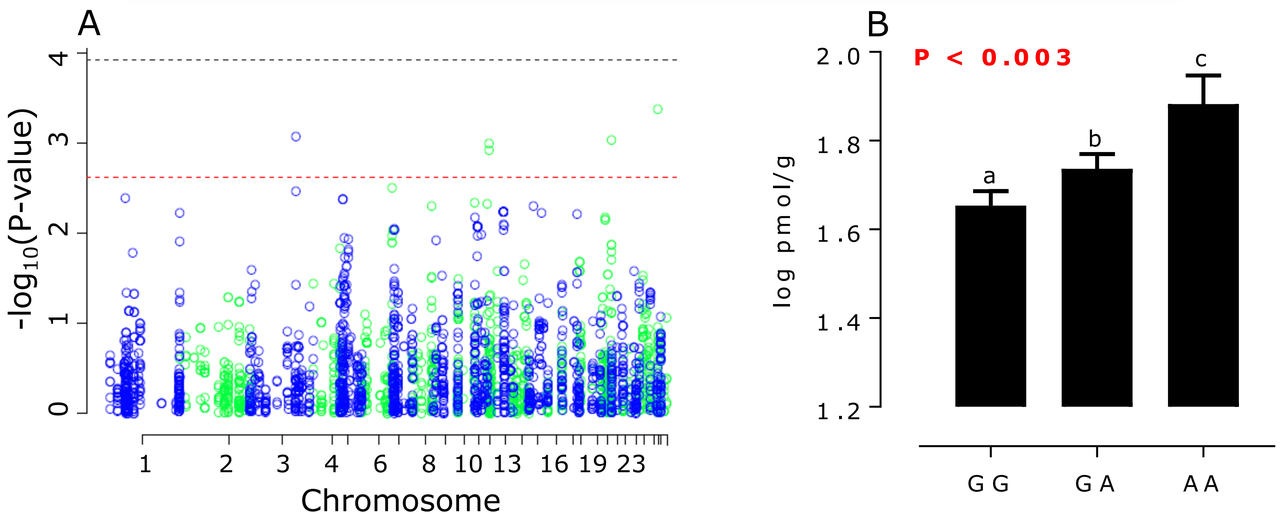


Figure 1. Example of **(A)** a Manhattan plot from genetic quantitative trait association analyses showing a significant association between Hcy and SNPs. **(B)** Histogram of liver Hcy concentrations with homozygous and heterozygous genotypes for gene *SUOX* at SNP dbrs159869938 (missense variant).

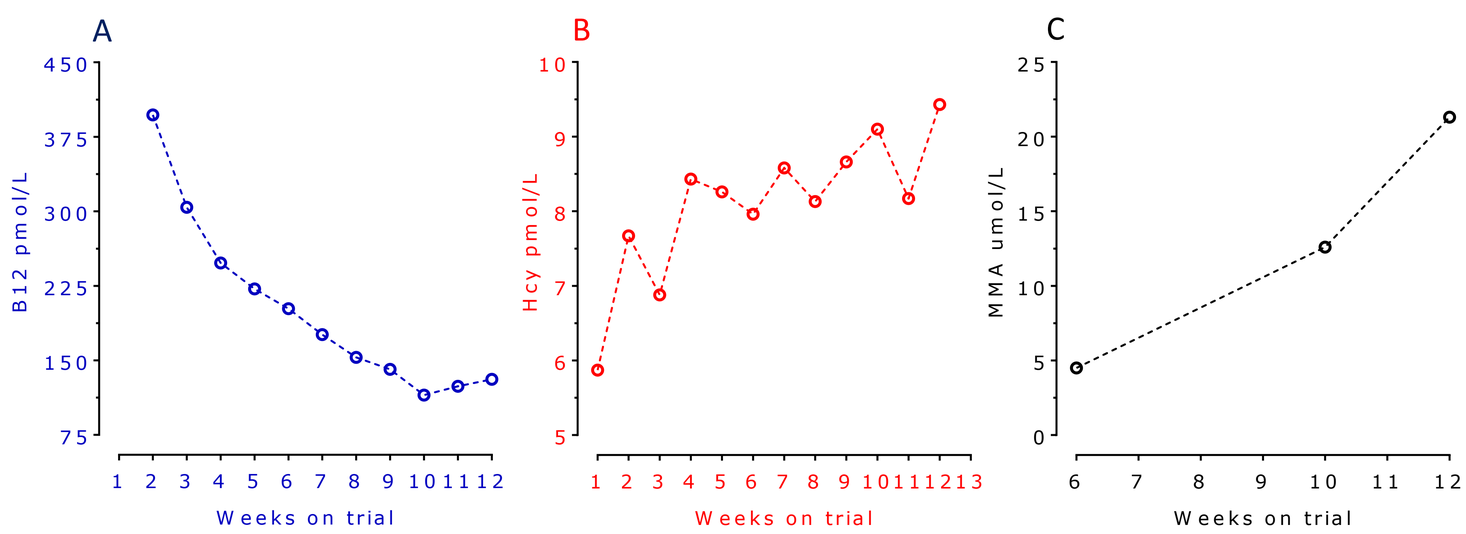


Figure 2. B12 (A), Hcy (B) and MMA (C) concentrations in sheep plasma.

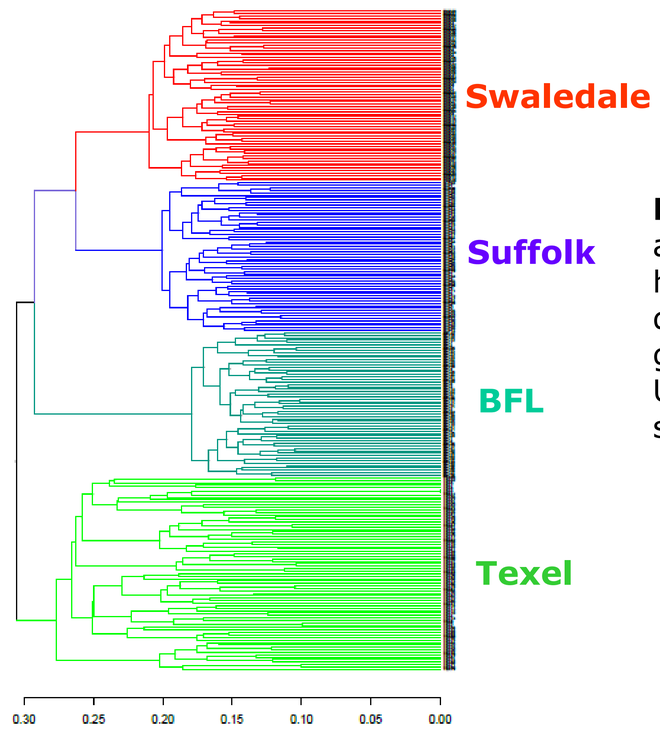


Figure 3. Phylogenetic analyses indicating a high degree of breed divergence for our 256 genes; with the three UK breeds clustering separately from Texels.