



Using runs of homozygosity to locate a new fatal recessive mutation in a rare breed of cattle

Geoff Pollott and Claire Wathes

Introduction

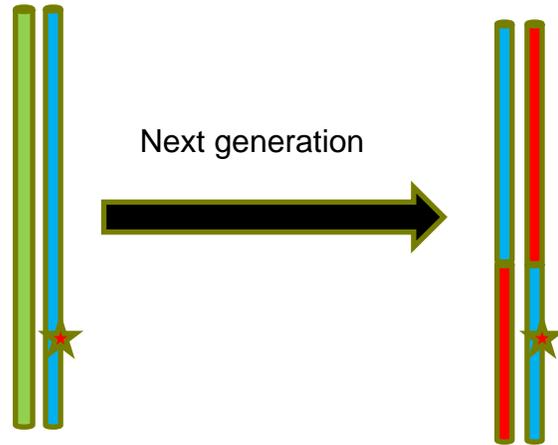
- Unexplainable peri-natal calf mortality noticed in Irish Moiled calves (rare breed)
- Segregation analysis indicated the likelihood of an autosomal recessive condition
- This work describes an attempt to locate the mutation causing this condition
- Uses SNP-chip data and a 'runs of homozygosity' (ROH) method of analysis

Development of autosomal recessive offspring



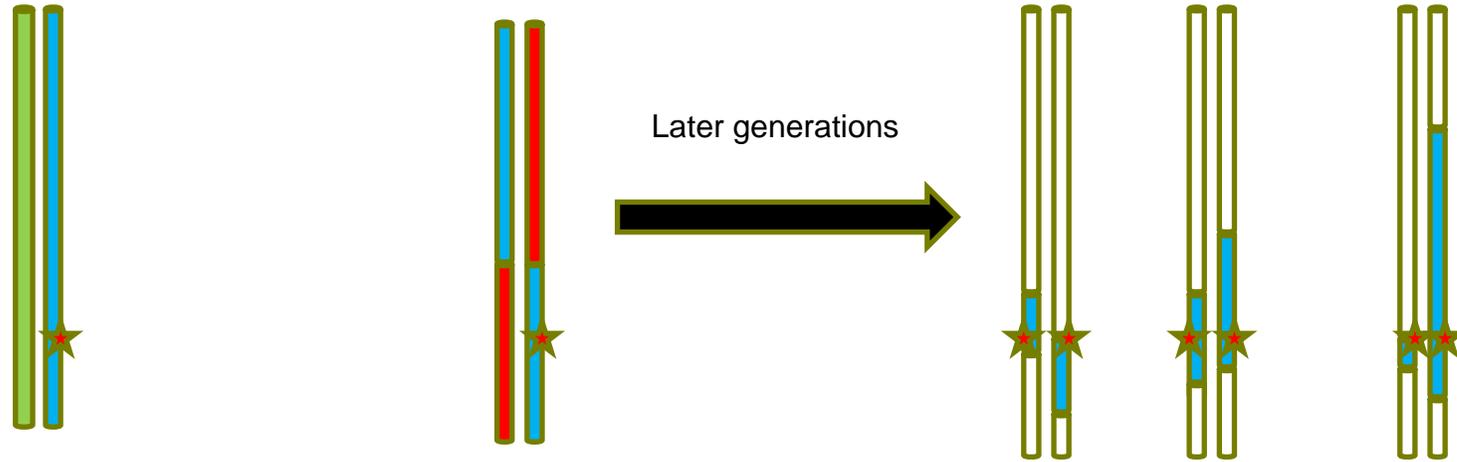
Mutation occurs on one of the autosomes

Development of autosomal recessive offspring



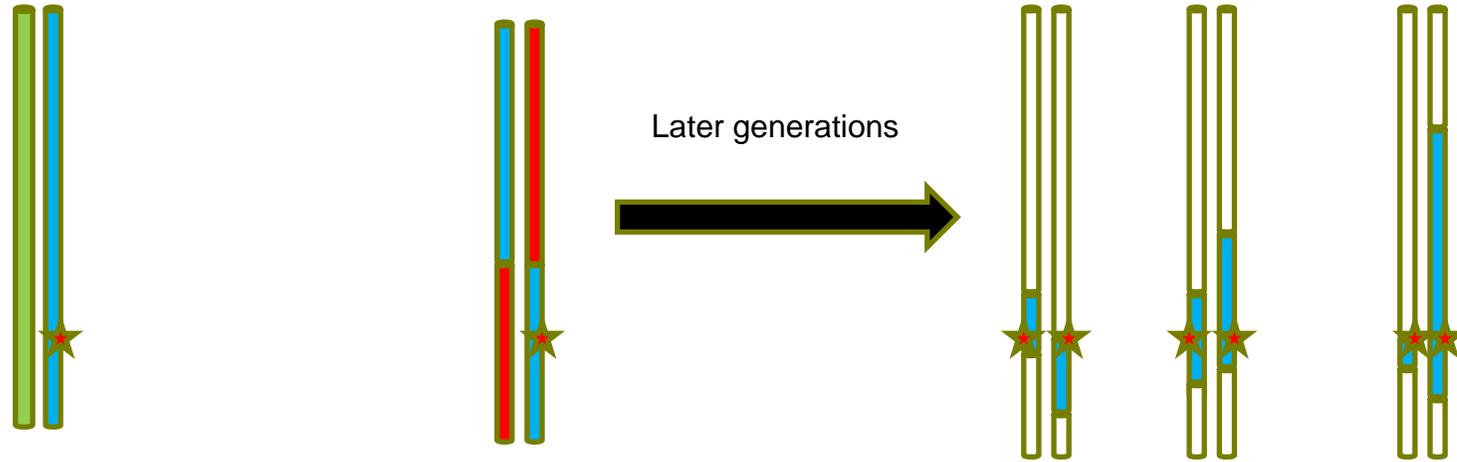
All offspring inheriting the mutation will have some of the original 'blue' DNA around it

Development of autosomal recessive offspring



Affected animals will appear with two copies of the mutation plus a variable amount of homozygous 'blue' DNA from the original mutated chromosome

Development of autosomal recessive offspring



Sections of overlapping 'blue' chromosome will be autozygous. Throughout the 'population' of affected animals, DNA closest to the site of the mutation will be more homozygous than DNA further away

Autozygosity by difference method (ABD)

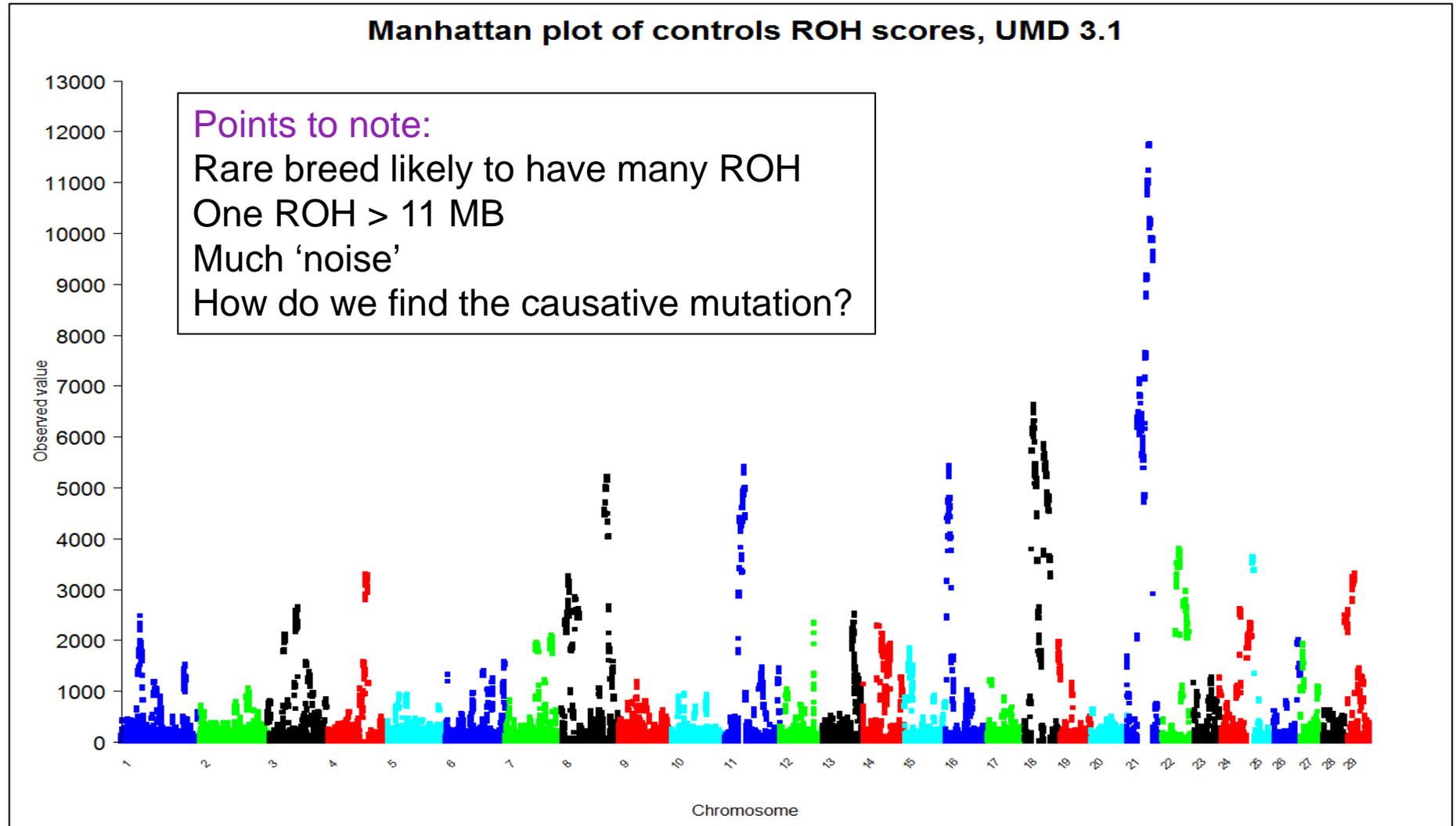
- Limitations of chi-squared, ASSHOM and ASSIST (Charlier et al, 2006) previously discussed
- ABD method devised based on ROH in cases and controls
- Reports mean ROH at each SNP marker in cases and controls separately, and their difference
- Genomic region with the largest difference likely to be location of the causal mutation
- 'Significance' of the results estimated by 10,000 permutations of randomly selected animals

Dataset

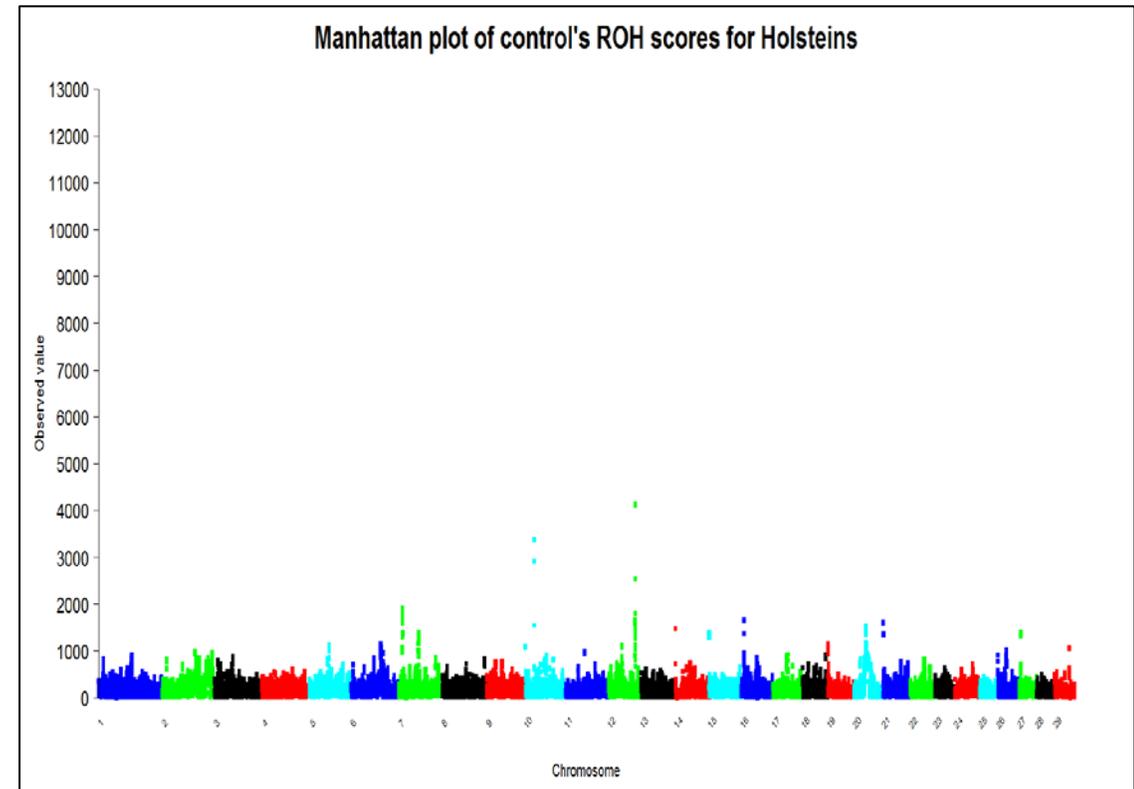
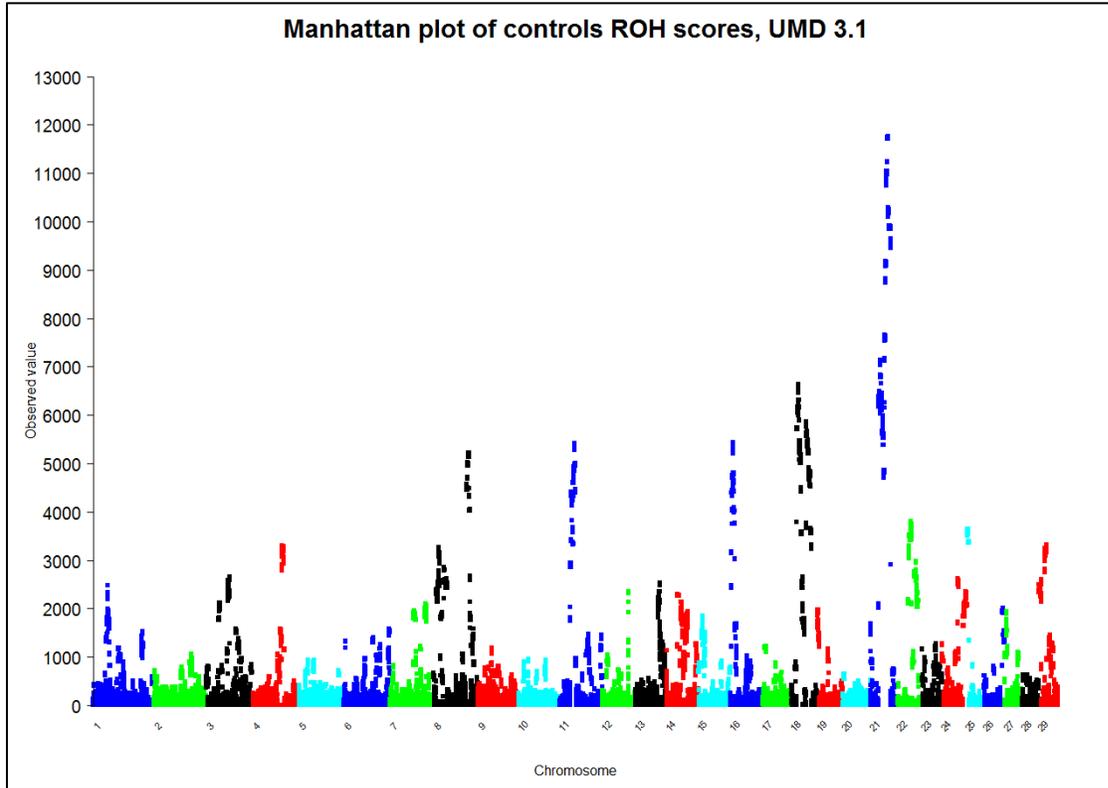


- Tail-hairs were collected from all animals analysed
- DNA was extracted from the accompanying follicles
- Data from 68 animals (24 dead cases, 44 live controls) was available using the Illumina BovineHD Beadchip.
- After quality control 580,071 SNP on autosomes available for analysis
- Results expressed as length of ROH in kilobases (KB)

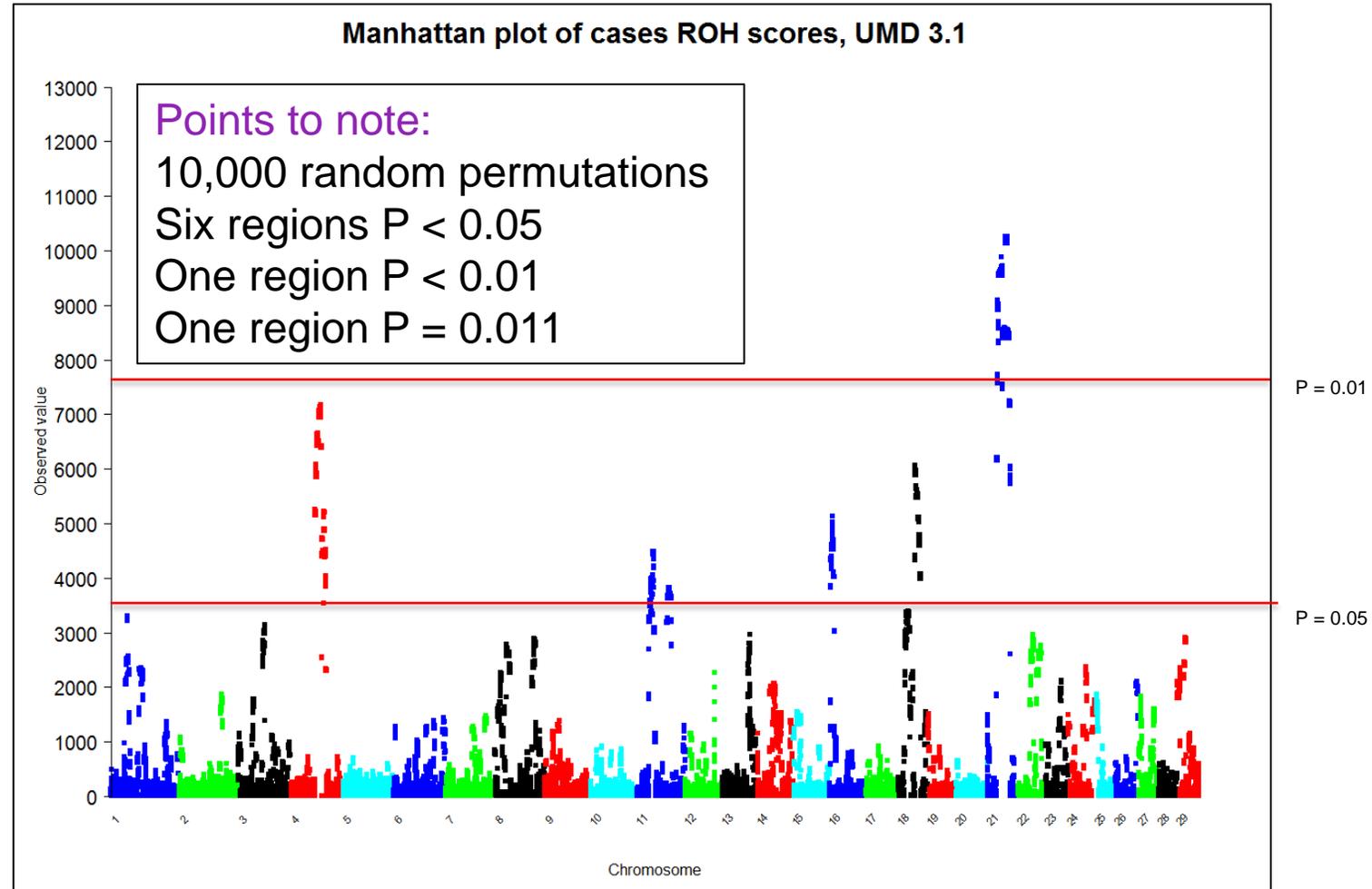
ROH in controls (KB)



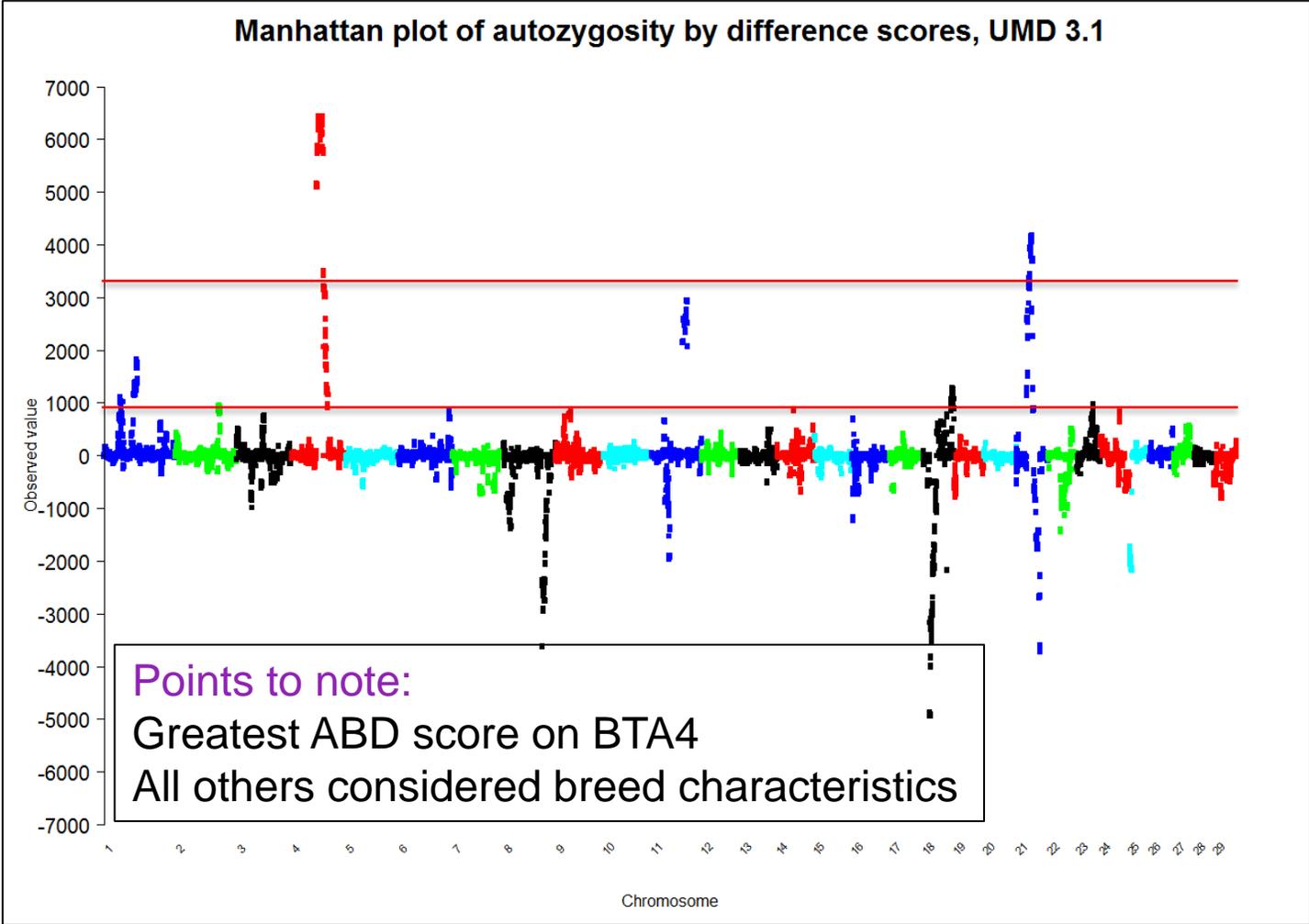
Holstein comparison



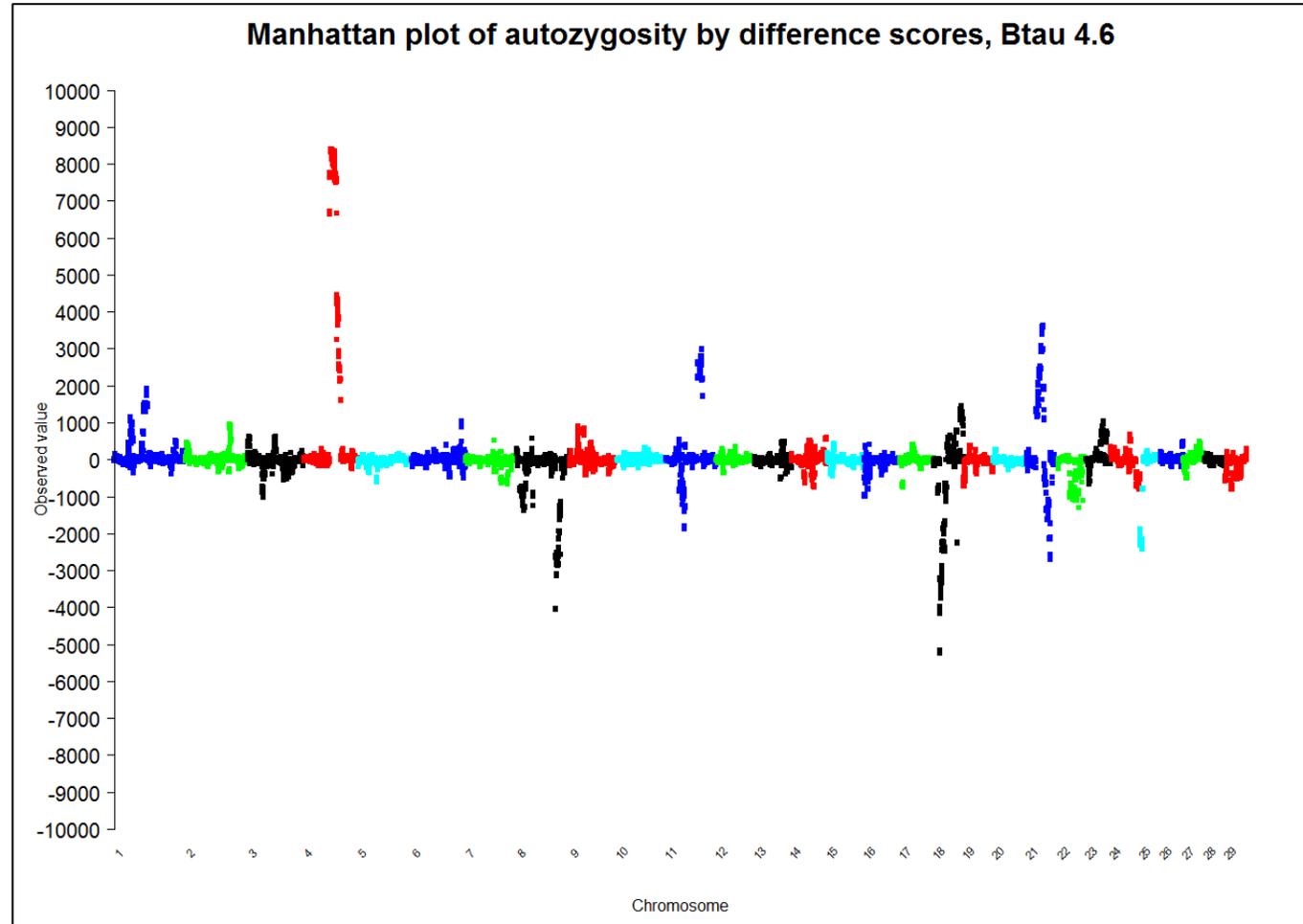
ROH in cases (KB)



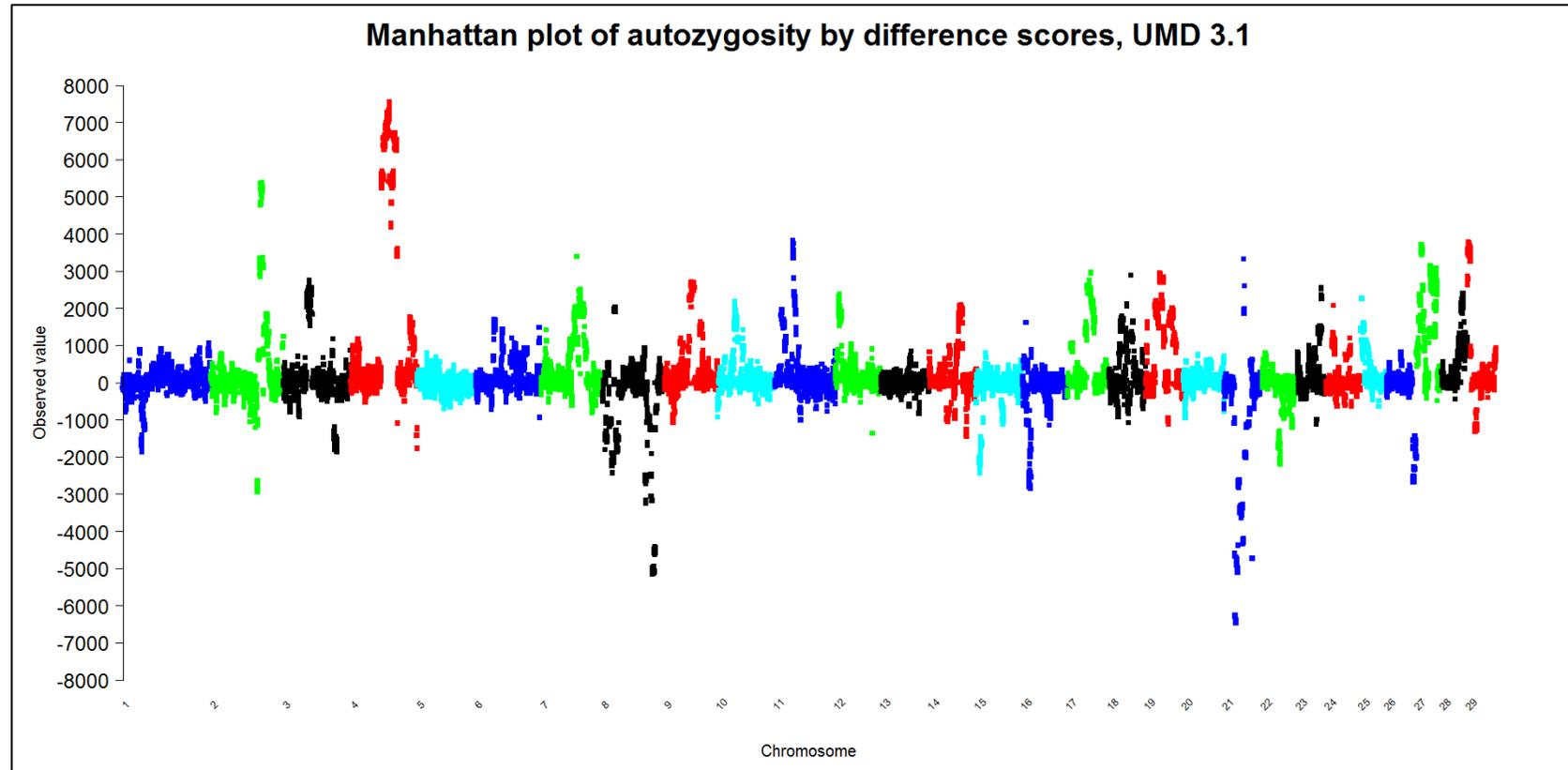
ABD scores (KB)



ABD scores (KB) using Btau 4.6



Results check – small dataset of from 50K SNP chip



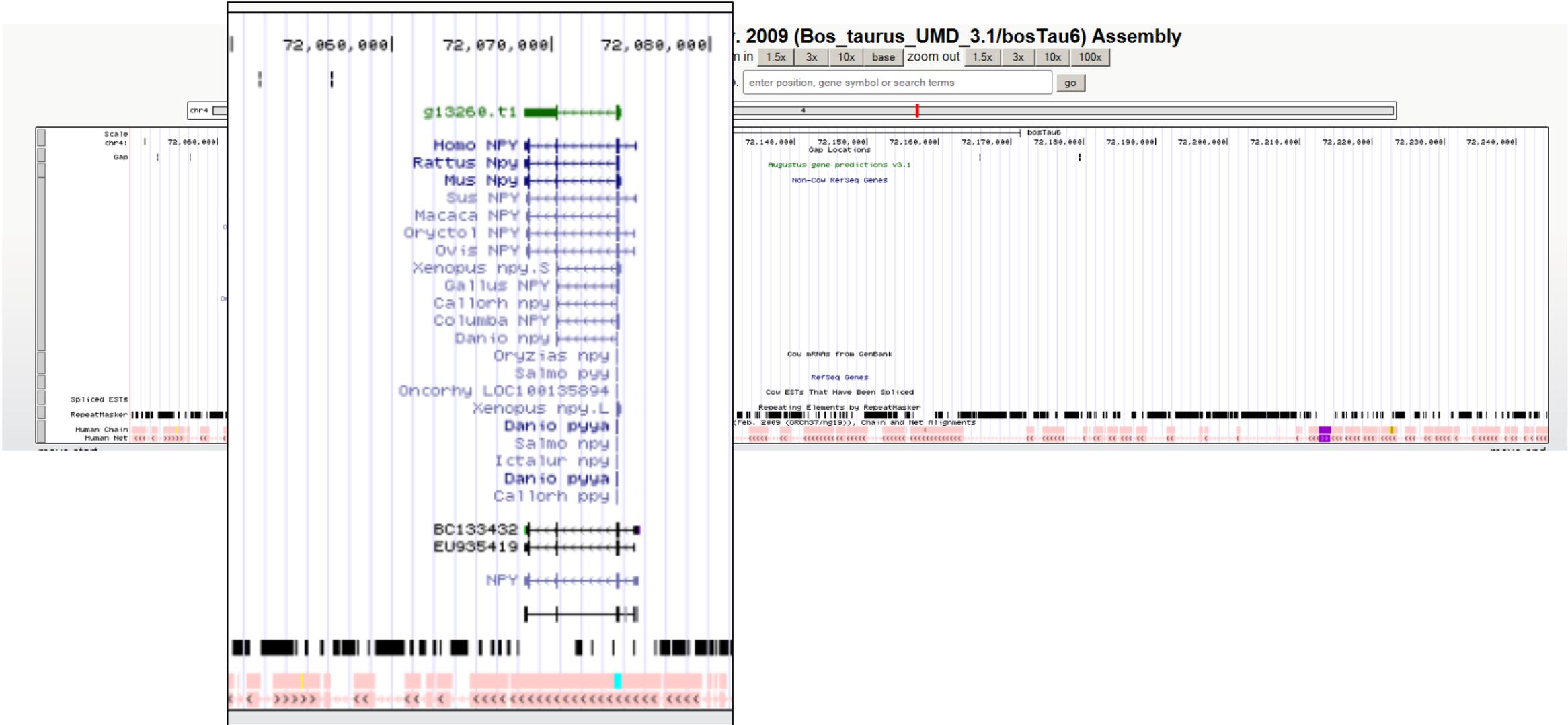
Issues with field samples

- › Phenotyping – did all dead calves die of the mutation?
- › Sample security – were all samples correctly identified?

ABD method

- Reasonably robust to mis-phenotyping and sample misidentification
- Level of these issues must be low

Genes in region with longest ROH in cases



NPY gene

- › Involved with leptin-NPY and insulin signalling pathways
- › Farmers have reported 'diabetes-like' symptoms in dying calves
- › No evidence from standard post-mortems
- › Sequencing of NPY exons showed no difference

Future work



- › Whole genome sequencing (WGS) to identify actual sequence changes in genomic regions so far identified
- › Apply ABD method to WGS data
- › Develop test for Irish Moiled animals
- › Breed the condition out

Acknowledgements



- Irish Moiled Cattle Society
- Rare Breeds Survival Trust
- Former Genesis-Faraday 'Spark' award