Bovine tuberculosis: herd-level surveillance of *Mycobacterium bovis* genotypes in Northern Ireland (2003-2008)

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Introduction Bovine TB is a respiratory disease caused by *Mycobacterium bovis* infection. It is the most complex and difficult multi-species, endemic disease facing government, the veterinary profession and the farming industry in the UK (Reynolds 2006). The maintenance and spread of infection within and between populations of cattle and populations of wildlife is poorly understood, with investigations hampered by difficulties in accurately defining epidemiologically-linked cases. The recent development of *M. bovis* genotyping schemes, based on recording DNA repeat copy number variation (variable number tandem repeats or VNTRs) offers highly-discriminating tools for investigating the spread of *M. bovis* (Skuce *et al* 2005, Smith *et al* 2006). Systematic application of *M. bovis* genotyping in conjunction with tracing of cattle movements and wildlife surveillance can be expected to provide valuable insights into disease source and spread. The objective of this study was to investigate, by means of structured herd-level surveillance, the genotype diversity, distribution and structure of the current *M. bovis* population.

Materials and methods New, culture-confirmed bovine TB herd incidents were enlisted on a rolling 365 day period (2003-2008). *M. bovis* was isolated and confirmed using standard procedures. The first (disclosing) *M. bovis* isolate per herd was subjected to genotyping (Skuce *et al* 2005). VNTR profiling, spoligotyping, nomenclature, reference isolates and quality control were as described (Skuce *et al* 2005). Animal-level data (movements and test records) were derived from the Animal and Public Health Information System (APHIS, Houston 2001). *M. bovis* isolates were geo-referenced to their final herd using MapInfo Professional v7.5. To investigate potential associations between *M. bovis* genotype and location and inter-annual differences in frequency, contingency tables were analysed using Chi-square tests with Monte Carlo simulation and by test for trend statistics.

Results 175 genetically-distinct *M. bovis* genotypes were identified in 8,630 isolates from 6,609 herds. On average 73 genotypes were identified every year, with 29 genotypes present in all 6 years. *M. bovis* genotypes showed striking geographical localisation and significant association (P<0.001) to regions. However, genotypes were also trans-located significant distances from their normal 'home range'. Whilst the frequency of most *M. bovis* genotypes was relatively stable over the survey period, significant differences (P<0.001) were observed for some genotypes in years 2003 to 2008, indicating that the *M. bovis* population was not entirely static. Despite regions being dominated by geographically-localised genotypes, significant and exploitable (in outbreak investigation) local diversity was still present.

Conclusions Significant genotype diversity was disclosed in the sampled Northern Ireland *M. bovis* population. The population was highly geographically structured, with different *M. bovis* genotypes tending to cluster significantly in distinct regions. This suggested that most sources tended to be local and relatively stable. Whilst the frequency of most *M. bovis* genotypes was relatively stable over the survey period, expansion and contraction of some genotypes was evident. *M. bovis* genotyping, in conjunction with comprehensive cattle movement databases and wildlife surveillance, offers a powerful tool for investigating bovine TB source, maintenance and spread. The population structure and the performance characteristics of *M. bovis* genotyping support its use to answer detailed epidemiological questions of direct policy relevance.

References
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