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## Phylogenetic analysis of Mycobacterium avium subsp. paratuberculosis in Australia

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n behalf of Animal Health Australia, Agriculture Victoria has been the responsible authority for the Australian Johne's Disease Reference Laboratory for the last 18 years. The reference laboratory has the largest collection of Mycobacterium avium subsp. paratuberculosis (MAP) isolates in Australia, holding 291 isolates collected over the last 30 years. Here we report genome sequencing of 231 representative isolates from the Australian MAP collection. To determine where Australian isolates are positioned in a global context and to assess the reliability of typing methods, phylogenetic analysis based on whole genome single nucleotide polymorphism (SNP) profiling, IS1311 genotyping, LSP gene analysis and average nucleotide identity were conducted on all isolates. Phylogenetic analysis of SNPs identified in the MAP core genome revealed 8 distinct clades within the Type C strains and 5 distinct clades within the Type S strains of MAP. There were fewer than 20 SNP differences across the core genome among some adjacent clades highlighting the monomorphic nature of MAP and suggesting that isolates from within a clade have risen from a common ancestor. Australian sheep strains clustered most closely to a sheep strain from Scotland (MAPMRIO103) and the Australian Bison strains clustered most closely to US bison type strains. No distinct phylogeographic clustering of MAP was observed in this study. IS1311 PCR and Restriction Enzyme Analysis (REA) intermittently generated incorrect results when compared to Long Sequence Polymorphism (LSP) analysis, whole genome SNP-based phylogenetic analysis, IS1311 sequence alignment and average nucleotide identity (ANI). These alternative methods generated consistent Map typing results. A published SNP based assay for genotyping Map was found to be unsuitable for differentiating between Australian and international strain types of Map.

### **Recent Publications**

- 1. Hodgeman RM, Mann R, Savin K, Djitro N, Rochford S and Rodoni B. (2021). Molecular characterization of Mycobacterium avium subsp. Paratuberculosis in Australia. BMC Microbiology, 21, 101.
- 2. Milne RJ, Poiani A, Coulson G, Hodgeman, R.2004. Faecal Escherichia coli and Chlamydophila psittaci in the Superb Lyrebird Menura novaehollandiae: host sex and age effects, Acta Ornithologica 39: 111-120(10).
- 3. Muller J, Gwozdz J, Hodgeman R, Ainsworth, C, Kluver P, Czarnecki JC, Warner S, Fegan M. 2015. Diagnostic Performance characteristics of a rapid field test for Anthrax in Cattle, Prev Vet Med, 120:277-82.

#### **Biography**

Rachel Hodgeman has over 25 years' experience in both food and veterinary microbiology. She has been the leader of the National Johne's Disease Reference Laboratory for Australia for the last 10 years. She established the diagnostic bacteriology laboratory for Agriculture Victoria to the highest quality standards of ISO17025 and obtained NATA accreditation for that laboratory in 2002. She has worked on several research projects including validation of qPCRs for diagnostic use, studying bacterial diseases of oysters, Leptospirosis in horses, pathogen identification for exporting meat to Singapore and studying psittacosis in Lyrebirds. She is researching John's disease as part of her PhD. She is studying the genetic diversity of MAP in Australia using comparative genomics in order to develop new and more accurate diagnostic identification and typing methods.

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