

Use of whole genome sequencing to understand the phylogenetic structure and epidemiology of *Mycobacterium bovis* in the United States

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National slaughter inspection is the primary method of detecting bovine tuberculosis (BTB) in cattle in the United States. Skin tests are administered for surveillance and investigation purposes. BTB continues to be sporadically detected in cattle and farmed cervids. It is unknown whether herds are infected by new introductions from animals or humans, or low levels of residual infection. Understanding the infection source is critical for managing program efficiency.

Lower resolution genotyping methods such as spoligotyping have been used for several years. Whole genome sequencing (WGS) was implemented in 2012, and performed on isolates from 1990 to the present. Consensus single nucleotide polymorphisms (SNP) were identified and used to classify strains, along with phylogenetic tree analysis.

875 *Mycobacterium bovis* isolates recovered within the U.S. were sequenced. These isolates separated into 23 major phylogenetic groups, with different strain types within groups. During 1998-2014 there were 129 BTB infected cattle and cervid herds and one feedlot. An isolate was not available for one herd. In addition, 18 cattle cases were found through slaughter surveillance where BTB was not confirmed in the herd of origin, for a total of 147 herds/cases. Of these, 78 strain types occurred in the U.S. prior to 1995, including the Michigan endemic area, (64 herds) and farmed cervid outbreaks (14 herds/cases). The remaining 68 herds/cases and the feedlot were infected with 43 strain types. Of these, 30 herds/cases were within 15 SNP of isolates from imported cattle. An additional 10 herds/cases were more distantly related to imported cattle. The source is not apparent for the remaining 28 herds/cases. Outside of known outbreaks, most cattle herd isolates are unique with limited spread to other herds. Imported cattle appear to be a source of introduction; however, limited imported cattle surveillance may underestimate this relationship. Other possible sources include low level residual infection or new introductions from infected humans. WGS is a powerful new tool in understanding the transmission dynamics of BTB in the United States that assist in prioritizing and directing local investigations.