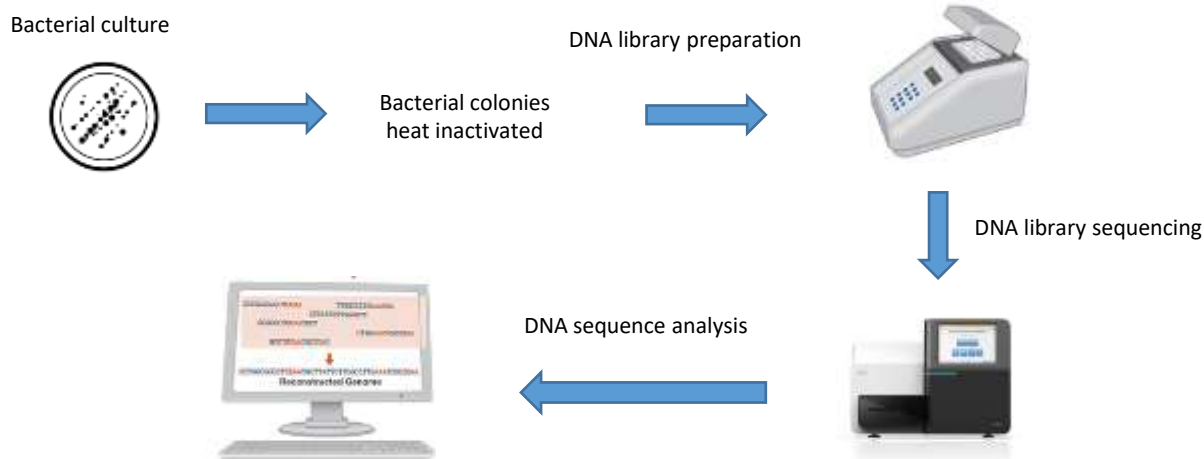


# Whole genome sequencing of *M. bovis*

## What is whole genome sequencing (WGS)?

Genetic material (DNA) is found in all living organisms, including bacteria. Each *M. bovis* bacterium contains unique DNA which carries the genetic instructions for its development, function, growth and reproduction. WGS is a molecular technique used to characterise the entire DNA content of an organism.



## How is WGS better than genotyping?

Genotyping was the method of genetic typing previously used by the Animal & Plant Health Agency (APHA) to characterise different strains of *M. bovis*. WGS replaced genotyping in April 2021 and offers considerable benefits. The table below compares the two molecular techniques:

Genotyping	Whole genome sequencing
Used a combination of two techniques; spoligotyping and variable number tandem repeat (VNTR) typing. These techniques target different regions of the bacterium's DNA sequence to characterise it.	Instead of targeting specific regions of the <i>M. bovis</i> genome, WGS analyses its entire DNA sequence.
Targeted specific regions of the bacterium's DNA sequence, looking for differences between isolates. It only allowed limited differentiation ('granularity') between <i>M. bovis</i> strains.	Sequencing the entire bacterial genome allows greater and more accurate discrimination between <i>M. bovis</i> strains.
Could distinguish between local sources of TB infection and breakdowns caused by introductions of infected cattle moved over long distances ('purchased' infection). It was more difficult to distinguish transmission routes between cattle herds infected with the same genotype located within the 'home-range' of that genotype (the geographical area where it's commonly found).	Allows assessment of genetic relatedness between isolates, providing additional information on evolutionary relationships. APHA can use this information to distinguish local sources of TB infection, identify transmission pathways between cattle herds, and where sufficient data exists, between cattle herds and wildlife.
Genotyping, like all genetic typing methods, first required the bacterium to be isolated by microbiological culture.	Like genotyping, WGS can only be carried out once <i>M. bovis</i> has been cultured, however the process is quicker than genotyping and costs about the same.

## How is WGS used in the control of bovine TB?



When a TB incident occurs, APHA field vets carry out a disease investigation to gather information about how TB infection could have entered the herd and whether it could have spread. To support this investigation, microbiological culture of *M. bovis* is attempted from tissue samples taken from the carcasses of slaughtered animals at post-mortem inspection. If *M. bovis* is cultured in the laboratory, genetic analysis is carried out to determine the particular strain of the bacterium.

- APHA uses WGS to characterise isolates of *M. bovis* cultured from cattle slaughtered for TB control. This provides information about the genetic relatedness of *M. bovis* strains, where they have come from and how they have evolved.
- WGS is an important tool used by APHA for investigating TB breakdowns and studying the spread of TB in the local and national cattle population, and the factors that affect it over time.
- At farm level, WGS helps APHA field vets to identify the most likely source of TB infection for a breakdown herd, and also whether it has spread to other cattle herds. Once APHA is aware of the likely origin of a breakdown, they can advise farmers on measures they could take to reduce the risk of further infection entering the herd.
- Records of historic *M. bovis* genotypes are still available to APHA staff involved in the control of bTB even though WGS is being used instead.
- WGS has only been used by APHA since June 2017, and not many historic *M. bovis* isolates have been sequenced. This means that the current WGS database which new isolates are compared against is limited, however this will improve over time now that WGS is being used routinely.
- Although WGS can provide more information about genetic relatedness of isolates than genotyping, there are limits to the inferences that can be made when interpreting WGS data. For example it's often not possible to determine the direction of transmission of *M. bovis* between different species.

## WGS and TB surveillance in wildlife

WGS is used to analyse *M. bovis* isolates from wildlife species (e.g. road killed badgers) and compare them with isolates from local TB breakdowns in cattle herds and other farmed animals. This allows APHA to assess whether cattle and badgers from the same geographical area are affected by the same strain of the TB bacterium, and to assess the spread of *M. bovis* strains between geographical areas and across time. For instance, WGS is being used to support TB surveillance in cattle and badgers and understand the epidemiology of bTB in the confirmed hotspot area in east Cumbria, England (HS21), where a link between cattle and badger TB infection was first identified in 2017. Visit [gov.uk](http://gov.uk)<sup>1</sup> to find out more about TB surveillance in wildlife in England.



### Reference

1. <https://www.gov.uk/government/publications/bovine-tb-surveillance-in-wildlife-in-england>