The Interferon Gamma test (IFNγ)

Bovine TB is a slowly developing disease cause by the bacteria *Mycobacterium bovis* (*M. bovis*). Identifying infected cattle requires the use of diagnostic tests, as it is rare for cattle to show any clinical signs until the later stages of infection. The interferon gamma (IFNγ) or 'gamma' test is a supplementary blood test used alongside the tuberculin skin test for the detection of TB in cattle in the UK.

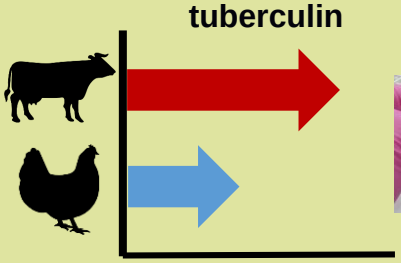
How does the test work?

The gamma test is carried out in the laboratory using freshly collected blood samples. The test measures the immune response of cattle, essentially using the same reaction as the skin test, but by challenging immune cells directly rather than challenging the live animal. Blood is stimulated with bovine and avian tuberculin and levels of interferon gamma (IFNγ) are measured. Interferon gamma is a cytokine (protein messenger) released by white blood cells (a key part of the immune system).

Avian tuberculin: made from *Mycobacterium avium*, which infects birds. Used alongside bovine tuberculin to distinguish between cattle infected with *M. bovis* and those which react to other environmental mycobacteria.

Bovine tuberculin: made from *M. bovis.* Triggers an immune response in cattle infected with *M. bovis* or similar *Mycobacteria.* Contains specific antigens (molecules which trigger an immune response) not present in environmental mycobacteria.

Cattle that have bovine TB will release greater amounts of IFNγ in response to bovine tuberculin compared to avian tuberculin



The test result

There are several possible outcomes for each blood sample subjected to the gamma test;

- **Positive** – the animal has failed the test and is classified as a reactor (gamma reactor)
- **Negative** – the animal has passed the test
- **Re-sample / Rejected** – the blood sample failed the quality controls for the test or was not tested (respectively). The laboratory will request a new sample from the same animal

How accurate is gamma test?

The accuracy of a test is usually measured in terms of **'sensitivity'** (probability that an infected animal is correctly identified) and **'specificity'** (probability that an uninfected animal is correctly identified). Data for Great Britain estimates that gamma test **specificity is 96.5%** and **sensitivity of around 90%** [1,2], but it is possible that is lower or higher than this under certain situations [3] :

Sensitivity = 90%

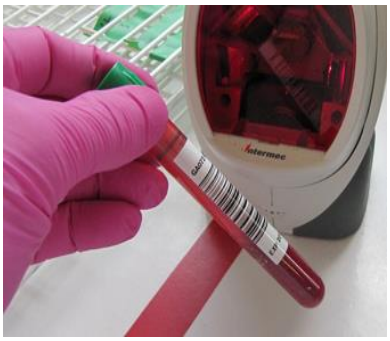
90% means that on average the test will identify 9 out of 10 infected cattle, meaning 1 will be missed

Specificity = 96.5%

This means an average of one false positive result for every 30 uninfected cattle tested.

1 in 30

How are blood samples collected?

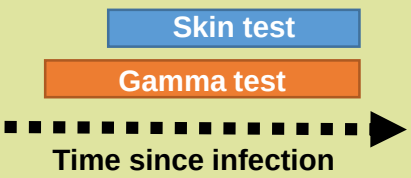


Blood samples are usually taken from the tail vein, or sometimes the neck and then placed for transport in specially designed temperature controlled packing to ensure the samples arrive in good condition at the laboratory. As with the skin test, suitable animal handling facilities are required. Government-funded gamma testing is currently only carried out by APHA staff, usually trained Animal Health Officers working in pairs. For private testing, blood samples can be collected by private vets (with prior approval from APHA) and submitted to APHA for analysis at the owner's expense.

What are the advantages of the gamma test?

Higher sensitivity – more likely to identify infected cattle than the skin test [1,2]

Identifies animals at an earlier stage of infection [4]



Identifies some animals missed by the skin test [5-8]

These benefits come at a cost, as the gamma test is more likely to produce 'false positive' results than the skin test (false +ve rate of 1 in 5000 cattle tested). This is one reason why the gamma test is used in herds which are believed to have a higher risk of TB, such as those which have already reacted to the skin test.

Why not leave gamma reactors in the herd?

Some animals react to the gamma test but not the skin test. Research has shown that these animals, if left in the herd, are much more likely to become skin test reactors in the future than gamma test negative animals [5-7] and a significant number of them will have gross visible lesions at post mortem [8]. For these reasons all gamma reactors are removed and slaughtered to reduce the chances of disease remaining in the herd.

'Non Visible Lesion' (NVL) reactors

Relatively few gamma reactors will have visible lesions at slaughter. This may be because animals are at an early stage of infection (visible lesions have not developed), or because slaughterhouse inspection has a low sensitivity (small lesions are not found). However, in a small proportion of cases this may be because the animals are false positives.

Where can I find more info?

For more information on testing and other TB topics visit www.tbhub.co.uk. This sheet was produced as a part of a Knowledge exchange project funded by NERC. For more info contact a.robertson@exeter.ac.uk or visit www.tbknowledgeexchange.co.uk.

Studies referenced

1. **de la Rua-Domenech et al. (2006)** Res. Vet. Science
2. **Schiller et al. (2009)** Clin. Vaccine Immunol.
3. **Nunez-Garcia et al. (2018)** Preventive Veterinary Medicine
4. **Pollock et al. (2005)** Veterinary Immunology and Immunopathology
5. **Lahuerta-Marin et al. (2015)** Veterinary Research
6. **Gormley et al. 2006** Veterinary Microbiology
7. **Coad et al. (2008)** Veterinary Record
8. **Clegg et al. (2017)** Preventative veterinary medicine.

NERC SCIENCE OF THE ENVIRONMENT

UNIVERSITY OF EXETER

Animal & Plant Health Agency

Department for Environment Food & Rural Affairs