

# ANIMAL HEALTH UPDATES

*Productivity Systems - Animal Health Group*

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## Disease - Tuberculosis (TB) in Bovines

Tuberculosis (TB) in cattle has different manifestations depending on the species of Mycobacteria involved.

### A. TB caused by *Mycobacterium bovis*.

*M.bovis* is the specific cause of bovine tuberculosis in cattle which has been classified under the "cattle diseases" notifiable to the Office International des Epizooties (OIE). All age groups and species are susceptible but infection is more prevalent in cattle and pigs. Infected cattle lose 10-25% of their productive efficiency. It is also an important zoonotic disease, particularly in immunocompromised individuals. Infection in human occurs mainly through consumption of infected milk. Spread can also occur by inhalation in people who are in close contact with infected cattle.

### Occurrence

All species, including human, and all age groups are susceptible to *M.bovis*. Cattle, goat and pig are most susceptible. Sheep and horse show high natural resistance.

### Sources of infection

Infected cattle are the main source of infection to other cattle. Organisms are excreted in the ex-

haled air, sputum, milk, urine, vaginal and uterine discharges and discharges from open peripheral lymph nodes. Cattle in early stages of disease may also excrete organisms in nasal and tracheal mucous, before any lesions are visible. Some wildlife and feral animals act as reservoir of infection for cattle which makes the eradication programme more difficult.

### Transmission

Inhalation is the most common portal of entry in cattle. Infection by ingestion is possible at pasture when faeces contaminate the feed and drinking water, but a large infective dose is required. Environmental contamination of pasture is not of major importance in epidemiology of the disease in cattle.

Drinking of infected milk by the young is a common means of transmission in the areas where the disease is endemic.

Sexual transmission may occur during coitus. Infected semen, insemination guns or uterine pipettes during artificial insemination are also implicated in transmission. Intramammary infection can occur if teat siphons or milking machine cups are contaminated.

### Risk factors

Poor housing predisposes animals to the disease. The closer the animals are in contact, the greater is the chance that the disease

will be transmitted. Zebu cattle are thought to be much more resistant than European cattle.

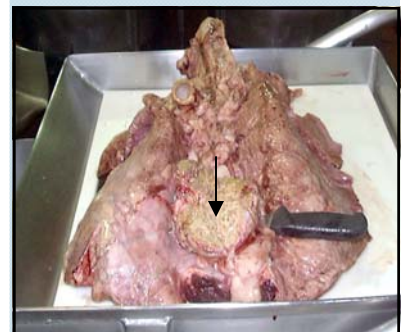
### Pathogenesis

Infection spreads in two stages:

**1.Primary complex :** This usually forms at the point of entry and local lymph node when infection is by inhalation. Bacteria pass from this primary focus which is in the respiratory tract in 90-95% of cases in cattle. In calves fed with tuberculous milk, the primary complex is likely to be in the pharyngeal or mesenteric lymph nodes.

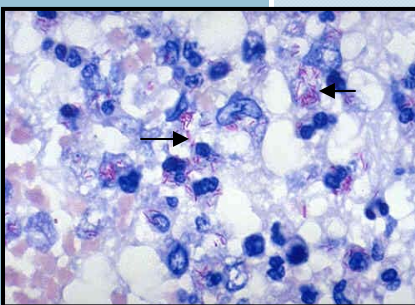
**2.Post primary dissemination:** This may take the form of acute miliary TB, discrete nodular lesions in various organs, or chronic organ TB caused by endogenous or exogenous reinfection. Lesions in the lungs in cattle occur in the caudal lobes in 90% of cases.

### Clinical Findings



TB lesion in the lung

Source : [www.michigan.gov](http://www.michigan.gov)



Acid fast stained Mycobacteria in a tissue section

Source : <http://bepast.org>



## Disease - Tuberculosis (TB) in Bovines (from page 1)

Progressive emaciation and fluctuating temperature become more pronounced after calving. Pulmonary involvement is characterized by chronic cough which is never loud, occurring only once or twice at a time and is low, suppressed and moist. Cough is easily stimulated by squeezing the pharynx or by exercise and is most common in the morning or cold weather. Infertility or recurrent abortion may occur late in pregnancy with lesions similar to Brucellosis occurring on placenta.

Tuberculous mastitis is of major importance due to its zoonotic potential, spread to calves and difficulty of differentiating it from other forms of mastitis. Udder is usually characterized by marked induration and hypertrophy.

### Diagnosis

The OIE has listed certain tests for identification of bovine TB infection.

#### 1. Delayed Type hypersensitivity test (DTH):

This is the only OIE prescribed test for detection of bovine TB. It is an allergic test wherein 0.1ml of Purified Protein Derivative (PPD) is injected intradermally on the side of the neck. The dose of PPD injected should not be lower than 2000 IU. The reaction is read 72 hrs after injection. An increase in skin thickness of over 4 mm is considered positive. The same person should measure the skin before the injection and 72 hours after injection. Bovine PPD is available at Indian Veterinary Research Institute (IVRI), Izatnagar, UP.

#### False positive reactions :

Various reasons for false positive reactions are:

⇒Animal sensitized to other mycobacterial allergens (*M.avium subspecies paratuberculosis* (MAP) and other non-pathogenic mycobacteria) and by ingestion of *M.avium* through water contaminated by birds which are infected with avian TB.

⇒Animals sensitized to other allergens eg. *Nocardia farcinus* in bovine farcy.

Tests in addition to DTH should also be used to reduce false positives (ELISA, culture).

#### False negative reactions :

False negative reactions could be due to the following:

⇒Advanced cases of tuberculosis

⇒Early cases until 6 weeks after infection

⇒Cows that have calved within the preceding 6 weeks

⇒Animals desensitized by tuberculin administration during the preceding 8 - 60 days.

⇒Old cattle

⇒Low potency tuberculin or bacterial contamination of tuberculin

#### 2. Comparative tuberculin test

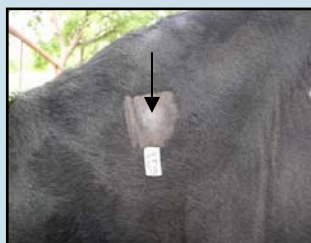
This test is generally used to follow-up on Bovine PPD positive reactors on DTH when infection with *M.avium* or *MAP* is suspected or a probability. This should not be done during 60 days following the initial DTH. It is recommended as a primary test when infection with *M.avium* or *MAP* is a probability.

When infection with *M.avium* is suspected or probable, Avian and Bovine PPD (dose as for DTH) are injected simultaneously into two separate sites on the same side of the neck 12 cm apart. The reading is taken after 72 hrs, the greater of the two reactions indicate the organism responsible.

Avian PPD is currently not available.

The same procedure is followed with *MAP* PPD (Johnin) and Bovine PPD when infection with *Johne's bacillus* is suspected or a probability. Johnin is available at IVRI, Izatnagar, UP.

The transitory positive reaction to bovine PPD on DTH that may occur in cattle due to *M.tuberculosis* infection in their attendants cannot be differentiated from *M.bovis* infection by the comparative tuberculin test.



An animal positive on Delayed Type Hypersensitivity Test

3

Other

#### tests

**a. Bacteriological examination:** May comprise demonstration of acid-fast bacilli by microscopy, isolation of bacteria on selective media and their subsequent identification by cultural and biochemical tests, DNA probes and PCR techniques (Available at IIL, Hyderabad and IVRI, Izatnagar, UP).

**b. Blood-based tests :** Lymphocyte proliferation assay (LPA), Gamma interferon (IFN- $\gamma$ ) assay (both from whole blood) and Enzyme Linked Immunosorbent Assay (ELISA) from serum. These are usually used as ancillary tests to confirm or negate the results of DTH.

#### Samples for confirmation of diagnosis

⇒**Bacteriology:** Affected lymph nodes, lung, granulomas from viscera. (culture, PCR)

⇒**Histology :** Formalin fixed samples of the above tissues (Microscopy)

#### Steps to control bovine TB in farms

⇒Feed and water troughs to be cleaned and thoroughly disinfected periodically with hot, 5 % phenol or cresol disinfectant.

⇒Suspicious reactors being held for retesting are to be isolated from the remainder of the herd.

⇒Calves are to be fed only with pasteurized milk.

⇒Farm attendants should be checked periodically as they could be a source of *M.tuberculosis* infection.

⇒Testing protocols must be adhered to at the time of purchase, quarantine and introduction of animals into the herd.

⇒Adequate fencing should be provided around the farm to prevent entry of other animals, which could be a source.

⇒When overall incidence of tuberculosis is 5% or less, compulsory testing and culling of reactors is the most satisfactory method of eradication.

⇒Transmission to humans can be significantly reduced by pasteurization of milk.

#### Requirements for a disease free herd

OIE has made certain recommenda-



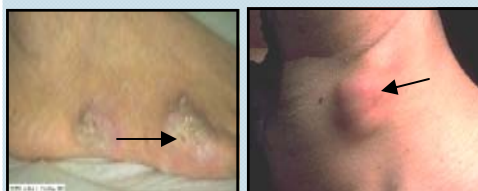
tions to manage human and animal health risks associated with *M.bovis*. These are important if the farm or area intends to qualify as a 'compartment' under OIE norms for purposes of international trade:

- ⇒No clinical signs of TB in animals
- ⇒Animals over 6 weeks of age has shown negative result to at least 2 TB (DTH) tests carried out at intervals of 6 months, the first test being performed 6 months after culling of last affected animal.
- ⇒Negative test result to annual TB testing to ensure continuing absence.
- ⇒New cattle to be procured from TB free herd or at least isolated and 2 TB tests carried out at 6 months interval with negative result.
- ⇒99.9% of animals in the 'compartment' free from TB for 3 consecutive years.

#### **B. Tuberculosis caused by *M. avium* and other atypical mycobacteria**

Tuberculosis caused by these is not a major disease problem but infected animals react to DTH, creating difficulty in *M.bovis* tuberculosis eradication programmes. Infected cattle are potential sources of infection of *M.avium* complex to humans.

Organisms of *M.avium* complex are ubiquitous and have been isolated from animal bedding in high numbers . Ingestion appears to be the normal infection route. The comparative tuberculin testing is used to differenti-



**Tuberculous lesions on foot and neck in humans**

Source : [www.visualsunlimited.com](http://www.visualsunlimited.com)  
<http://matrix.ucdavis.edu>

ate it from *M.bovis* infections.

#### **C. Tuberculosis caused by *M. tuberculosis***

Most cases are transitory and removal of tuberculous human from environment usually results in disappearance of positive reactors in cattle.

Reactors are most common in young

stock.

#### **D. Skin Tuberculosis**

It is caused by non-pathogenic mycobacteria. Chronic indurative lesions of skin occur usually on the lower limbs. Lumps of 1-2 cm in diameter appear below the skin and often occur in chains. Iatrogenic lesions may be caused by vaccines that produce subcutaneous granulomas, colonized by non-pathogenic mycobacteria. Cutaneous abrasions and tick bites are probable portals of entry for the organism.

The affected animals may give a suspicious or positive reaction to tuberculin test when they are in fact free of tuberculosis. In herds with tuberculosis, reactors having lesions of skin tuberculosis are usually culled, but in free herds, a reactor with skin tuberculosis is considered as non-specific and is retained provided it is negative on retest.

#### **Therapeutic effect of Nisin in subclinical mastitis**

Researchers from Department of Veterinary Medicine, College of Animal Sciences, Zhejiang University, China, evaluated an antimicrobial peptide, Nisin, in the treatment of subclinical mastitis in lactating cows. The report published in the July 2007 edition of Antimicrobial Agents and Chemotherapy indicated that nisin therapy caused bacteriological cure rate of 90.1% for *Streptococcus agalactiae* (10 of 11), 50% for *Staphylococcus aureus* (7 of 14), 58.8% for coagulase negative staphylococci (7 of 17), and 65.2% for all cases (30 of 46). Meanwhile, only 15.9% (7 of 44) spontaneously recovered in the untreated cows. The number of mammary quarters with N-acetyl-beta-D-glucosaminidase (NAGase) activity and high milk SCC were significantly decreased after Nisin-treatment. The therapeutic effects on bovine subclinical mastitis, as well as its safety in humans, makes nisin to be a good candidate for treatment of subclinical mastitis. Nisin is produced commercially by fermentation of natural substrates, including milk, using *Lactococcus lactis*.

Source: [www.ncbi.nlm.nih.gov](http://www.ncbi.nlm.nih.gov)

#### **Dairy food intake reduces risk of prostate cancer**

Researchers from Fred Hutchinson Cancer Research Center, University of Washington, Roswell Park Cancer Institute, and Swedish Medical Center and Swedish Cancer Institute, report that smokers may benefit from boosting dairy consumption and reducing omega-6 intakes. The new research, published in the *Journal of Nutrition*, reports that an increased intake of dairy foods may cut risk of prostate cancer in smokers by 40%. They also report that an increased intake of omega-6 fatty acids may increase the risk of prostate cancer, adding to an ever-growing body of research suggesting the omega-6-rich diet may be detrimental to prostate health.

Researchers based their result on the study of dietary habits of 12,025 men for a period of 11 years.

Source: [www.nutraingredients.com](http://www.nutraingredients.com)

#### **'LipoBridge' technology to break blood brain barrier**

According to scientists, 99 per cent of all potential Central Nervous system (CNS) drugs are larger than 500 Daltons, and thus automatically rejected by the blood brain barrier (BBB) which simply will not allow molecules this large to pass. However LipoBridge technology may be able to change all that according to its developers, Genzyme.

In LipoBridge technology, the therapeutic active agent is formulated with short chain oligoglycerolipids to facilitate transport across the blood brain barrier. The LipoBridge formulation interferes with the barrier, temporarily opening up the tight junctions and allowing drug compounds to cross to the CNS. The BBB remains open for a very short period of time thus reducing the risk of other unwanted molecules crossing over. The BBB also remains intact following the drug delivery.

Source : [www.drugresearcher.com](http://www.drugresearcher.com)



**OIE - Significant animal diseases reported to OIE in July'07**

Sl.No	Disease Outbreak	Countries reporting
1	Foot and Mouth Disease	Kyrgyzstan
2	Blue Tongue	Belgium, France, Spain, Germany
3	Highly Pathogenic Avian Influenza	France, Germany, India
4	Peste des Petites Ruminants (PPR)	China, Uganda
5	New Castle Disease	Honduras, Estonia, Chile
6	African Swine Fever	Burkina Faso
7	Low Pathogenic Avian Influenza	USA (Source : <a href="http://www.oie.int">www.oie.int</a> )

**'Superbug' strain of MRSA detected**

In the Netherlands, Methicillin Resistant *Staphylococcus aureus* (MRSA) strain has been found in 20 per cent of pork, 21 per cent of chicken and 3 per cent of beef on sale to the public, the UK's Soil Association stated.

The association warned that MRSA found in farm animals have already transferred to farmers, farm-workers and their families in the Netherlands, causing serious health impacts.

About 40 per cent of pigs and 50 per cent of pig farmers in the Netherlands have been found to carry farm-animal MRSA, the Soil Association stated.

Dutch scientists and government officials blame the MRSA new strain in farm animals on the high levels of antibiotics used in intensive livestock farming, according to the association.

MRSA is already a high-profile, persistent problem in many UK hospitals.

Source: [www.foodproductiondaily.com](http://www.foodproductiondaily.com)

**MRSA transmission from cows to humans**

Scientists from Central Veterinary Institute, Budapest, Hungary, isolated methicillin-resistant *Staphylococcus aureus* (MRSA) from cows with subclinical mastitis and from a person who worked with these animals which they claim is the first documented case of direct transmission of MRSA between cows and humans. The bovine and human strains were indistinguishable by phenotyping and genotyping methods. This was reported in the April 2007 edition of Emerging Infectious Diseases Journal

Source : [www.ncbi.nlm.nih.gov](http://www.ncbi.nlm.nih.gov)

**Biofilm susceptibility test for *S.aureus* mastitis**

Various studies have shown that there is a discrepancy between the results of this standard susceptibility test and the actual cure rate of the applied antimicrobial treatment. Increasing evidence suggests that biofilm formation by *S.aureus* is associated with this problem. The currently available antimicrobial susceptibility assays for bacteria growing in biofilms, are not considered reliable enough for routine application. Researchers from Department of Veterinary Pharmacy and Toxicology, Faculty of Veterinary Medicine, University of Utrecht, Netherlands, developed a susceptibility test for bacteria growing in biofilm, suitable for routine testing of the antimicrobial susceptibility of *S.aureus*. The results showed clear differences between strains and various antimicrobial agents with respect to the effect of longer duration of the antimicrobial challenge on the eradication of *S.aureus* growing in biofilm. This was reported in the May 2007 edition of Veterinary Microbiology journal.

Source : [www.ncbi.nlm.nih.gov](http://www.ncbi.nlm.nih.gov)

**Lactoferrin to fight antibiotic resistant pathogens**

Lactoferrin is present in cow's milk and this protein appears to have many biological functions, including antibacterial and anti-inflammatory activities.

Researchers from Agriculture and Agri-Food Canada, Dairy and Swine Research and Development Centre, Canada, evaluated the efficacy of intramammary treatments containing penicillin G and (or) bovine Lf (bLf) to cure chronic mastitis caused by a clinical isolate of *S.aureus* highly resistant to beta-lactam antibiotics. Bacterial cure rate was null for control quarters, 11.1% for bLf, 9.1% for penicillin and 45.5% for the combination in a group where mastitis was induced. While studying the effect on chronic mastitis acquired in previous lactation, bacterial cure rate was higher for the bLf + penicillin combination (33.3%) compared to penicillin alone (12.5%). In conclusion, bLf added to penicillin is an effective combination for the treatment of stable *S.aureus* infections resistant to beta-lactam antibiotics. The report was published in Journal of Animal Sciences, June'07.

Source : [www.ncbi.nlm.nih.gov](http://www.ncbi.nlm.nih.gov)

**Brain cancer vaccine launched in Switzerland**

The company, Northwest Biotherapeutics (NWBT) received approval from the Swiss Institute of Public Health - to market DCVax-Brain, a vaccine for the treatment of patients with brain cancer. DCVax-Brain and DCVax-Prostate are personalised, made by combining a patient's own master immune cells (dendritic cells) with cancer biomarkers derived from or displayed by the patient's own tumor. The vaccines work by mobilising the full spectrum of immune response, rather than just single immune agents such as antibodies or T cells alone. Source : [www.drugresearcher.com](http://www.drugresearcher.com)

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