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Bovine tuberculosis and its zoonotic importance

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Zoonotic diseases are naturally transmitted from animals to humans (WHO, 2020. Tuberculosis is one of the most important and widespread zoonotic diseases. Despite slow reductions in the annual burden of active human tuberculosis (TB) cases, zoonotic TB (zTB) remains a poorly monitored and an important unaddressed global problem. Zoonotic tuberculosis (TB), caused by *Mycobacterium bovis*. *M.bovis* is a member of the Mycobacterium tuberculosis complex (Mtbc), which includes *M.tuberculosis, M.caprae, M.microti, M.africanum, M.canetti, M.pinnipedii,* and *M.bovis.* These belongs to a single genus *Mycobacterium* within the family *Mycobacteriaceae* and order *Actinomycetales* (Cocito *et al* 1994). The genus Mycobacteria are obligate intracellular pathogen, aerobic, non-spore forming, non-motile, rod shaped and acid fast bacilli which include diverse species ranging from environmental saprophytes and opportunistic invaders, and all are slow growers, they take several weeks to months for growth. Mycobacterial cell wall is rich in lipids such as mycolic acids (long-chain fatty acids C78-C90), waxes, and phospholipids. Unlike most of the organisms in this group, *M.bovis* has a broad host range, including cattle, cervids, badgers, humans, and many other animals.

Economic burden:

Zoonotic TB poses a significant risk, particularly in low- and middle-income countries where livestock rearing practices and raw dairy consumption are common. The global prevalence of human TB due to *M.bovis* has been estimated at 3.1%. Of all human TB cases, 2.1% and 9.4% are pulmonary and extra-pulmonary TB cases respectively (Cosivi et al 1998). Many factors like poverty, high population density, social stigma, poor public awareness, and overwhelmed health systems lack of resources for TB transmission prevention and treatment were acting stumbling blocks to control and eradicate bovine TB in developing countries. Based on a random-effects (RE) meta-regression model, the analysis revealed a pooled prevalence estimate of 7.3% (95% CI: 5.6, 9.5), indicating that there may be an estimated 21.8 million (95% CI: 16.6, 28.4) infected cattle in India a population greater than the total number of dairy cows in the United States (Srinivasan et al 2018).

Transmission:

1. Ingestion of Infected milk, dairy products, or meat can transmit the bacteria.
2. Close contact with infected animals or their bodily fluids can also lead to transmission.

3. Inhalation of airborne droplets containing Mycobacteria from animals with pulmonary TB, especially in crowded and less-ventilated settings, can be a potential route of transmission.

Pathogenesis:

The primary portal of entry for *M.bovis* among all species is respiratory, other common routes include oral, congenital, or entry through open wounds (Good *et al*., 2011). When the organism gains entry through the mucous membrane or alveolar spaces, the immune system responds to the bacterial cell wall and activates the inflammatory process for phagocytosis. This makes macrophages and neutrophils accumulate at the sites of infections (Arentz and Hawn, 2007). Muramyl dipeptide (from peptidoglycan) complexes with mycolic acids present in cell wall can cause granuloma formation, phospholipids induce caseous necrosis.

Symptoms: Animals

- Affected animals may become weak, sluggish and have a general lack of energy. A progressive loss of body condition and appetite is a common sign. Some animals may exhibit a low-grade, fluctuating fever.
- The swelling of Supramammary Lymph Nodes and other lymph nodes, particularly in the head, neck, and chest areas, is a characteristic sign.
- Intermittent Diarrhea or Constipation. In some cases, a chronic, moist cough and difficulty breathing may occur.
- In later stages, animals may become severely thin and emaciated. The affected udder quarter may become progressively hardened.

Symptoms: Human beings

Pulmonary TB (Affecting the Lungs):

- Persistent cough, coughing up blood or mucus, chest pain, a persistent fever, especially in the evening, or chills, night sweats, feeling tired or weak all the time, weight loss, and loss of appetite

Extrapulmonary TB (Affecting Other Organs):

- TB can affect other organs besides the lungs, and symptoms will depend on the affected organ. Swollen lymph nodes, abdominal pain, joint or bone pain, confusion or headache, seizures

Lesions:

TB can cause lesions in the lungs, lymph nodes, liver, spleen, and other organs. Abscesses, or pockets of infection, can develop in various tissues and organs.

Diagnosis:

- Based pathognomonic symptoms and lesions
- The tuberculin test, based on a delayed-type hypersensitivity to mycobacterial tuberculin, is the standard ante-mortem test in cattle. Reactivity in cattle is usually detectable 30 to 50 days after infection (Monaghan et al., 1994).
- Two main methods of tuberculin testing are employed:
    a. Single intradermal (caudal fold) test: 0.1 ml of bovine PPD is injected intradermally into the caudal fold of the tail. The injection site is examined 72 hours later, and a positive reaction is characterized by a hard or oedematous swelling.
    b. Comparative intradermal test: 0.1 ml of avian PPD and 0.1 ml of bovine PPD are injected intradermally into separate clipped sites on the side of the neck about 12 cm apart.
- Skin thickness at the injection sites is measured with calipers before injection of tuberculins and after 72 hours. An increase in skin thickness at the

injection site of bovine PPD which exceeds that at the avian PPD injection site by 4 mm or more is interpreted as evidence of infection and the animal is termed a reactor.

- Blood-based tests that have been developed for use in conjunction with the tuberculin test include: – Gamma interferon assay. This test identifies animals at a slightly earlier stage of infection than the tuberculin test and is approved as a supplementary test for cattle in the EU, USA and New Zealand.
- ELISA is a low-cost method of detection for antibodies and it can be used in countries with a high prevalence of bovine TB with large number of animals with chronic disease.
- Lymphocyte transformation and related assays
- Specimens suitable for laboratory examination: Lymph nodes, tissue lesions, aspirates and milk.
- Stained tissue sections usually reveal typical patterns of tubercle formation.
- **Isolation of *M. bovis*:** Decontamination of specimens to eliminate fast-growing contaminating bacteria. Ground-up specimens are treated for up to 30 minutes with 2% to 4% sodium hydroxide or 5% oxalic acid, followed by neutralization of the alkali or acid.
- After the centrifugation, sediment is used for inoculation into Lowenstein - Jensen medium/without glycerol and containing 0.4% sodium pyruvate, or Ogawa egg yolk media/ Middlebrook 7H10 agar are inoculated with the centrifuged deposit and incubated aerobically at 37° C for up to 8 weeks.
    - Growth rate and colonial appearance Positive ZN staining of bacilli in smears from colonies. The Biochemical profile will help confirm Mycobacteria.
    - Septi-check FB system- used for recovery of Mycobacteria.
    - Continuous automated Mycobacterial liquid culture system
    - The MB/Bac T system
    - BACTEC 60 TB sytem: God standard stem for confirmation of Mycobacteria.

Control Measures:

In 2020, the RNTCP was renamed as the National TB Elimination Program (NTEP) to emphasize the aim of the Government of India to eliminate TB in India by 2025, five years ahead of the global targets of 2030. The SDG targets for TB (base line 2015) are: 80% reduction in incidence. 90% reduction in mortality in 2020.

a. Proper food hygiene practices, including pasteurization of milk, are crucial for preventing transmission.
b. Early detection and control of bovine TB in animals remains the mainstay of reducing zoonotic TB risk.
c. Addressing zoonotic TB requires a "One Health" approach, recognizing the interdependence of human and animal health sectors.
d. Stronger inter-sectoral collaboration between veterinary and medical professionals is essential for effective control.

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