Johne’s Disease (Paratuberculosis)

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Johne’s (Yo-nees) disease is a contagious, chronic, and usually fatal infection that affects primarily the small intestine of ruminants. Johne’s disease is caused by Mycobacterium avium subspecies paratuberculosis (M. avium subsp. paratuberculosis), a hardy bacterium related to the agents of leprosy and TB. Johne’s disease is found worldwide.

The disease, discovered by Heinrich A. Johne (1839-1910), a German bacteriologist and veterinarian, in 1905, an acid-fast bacillus, often abbreviated MAP. MAP is akin to, but distinct from, Mycobacterium tuberculosis, the main cause of tuberculosis in humans, and Mycobacterium bovis, the main cause of tuberculosis in cattle and occasionally also in humans. MAP is 99% genetically related to Mycobacterium avium, but has different phenotypic characteristics, such as:

- slower growth
- requires the addition of an iron transport chemical known as mycobactin when grown in vitro
- forms a rough colony when grown on a solid agar medium
- Infects mammals instead of birds

Also, the environmental distribution of MAP is markedly different from that of M. avium, which can produce mycobactin, so can grow and multiply outside the body.

Because few herds have instituted biosecurity programs, infection continues to spread. Although infection seems less widely distributed in beef and goat herds and sheep flocks, Johne’s is nonetheless of critical significance to all producers.

Johne’s disease can have severe economic impacts on infected herds. Identifying and protecting noninfected herds and flocks will provide a source of breeding stock and replacement animals for others and help to reduce the national prevalence of the disease.

Zoonosis

MAP is capable of causing Johne’s-like symptoms in humans, though difficulty in testing for MAP infection presents a diagnostic hurdle. Clinical similarities are seen between Johne's disease in ruminants and inflammatory bowel disease in humans, and Mycobacterium avium, but has different phenotypic characteristics, such as:

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Clinical Signs and Stages

In cattle, the main signs of paratuberculosis are diarrhea and wasting. Most cases are seen in 2- to 6-year-old animals. The initial signs can be subtle, and may be limited to weight loss, decreased milk production, or roughening of the hair coat. The diarrhea is usually thick, without blood, mucus, or epithelial debris, and may be intermittent. Several weeks after the onset of diarrhea, a soft swelling may occur under the jaw. Known as "bottle jaw" or intermandibular edema, this symptom is due to protein loss from the bloodstream into the digestive tract. Paratuberculosis is progressive; affected animals become increasingly emaciated and usually die as the result of dehydration and severe cachexia.

Signs are rarely evident until two or more years after the initial infection, which usually occurs shortly after birth. Animals are most susceptible to the infection in the first year of life.
Newborns most often become infected by swallowing small amounts of infected manure from the birthing environment or udder of the mother. In addition, newborns may become infected while in the uterus or by swallowing bacteria passed in milk and colostrum. Animals exposed at an older age, or exposed to a very small dose of bacteria at a young age, are not likely to develop clinical disease until they are much older than two years.

The clinical signs are similar in other ruminants. In sheep and goats, the wool or hair is often damaged and easily shed, and diarrhea is uncommon. In deer, paratuberculosis can progress rapidly. Intestinal disease has also been reported in rabbits and nonhuman primates.

Unlike cattle and sheep, infections in deer often present with clinical illness in animals under one year of age.

In cattle, signs of Johne’s disease include weight loss and diarrhoea with normal appetite. Several weeks after the onset of diarrhoea, a soft swelling may occur under the jaw. This intermandibular oedema, or “bottle jaw,” is due to protein loss from the bloodstream into the digestive tract. Animals at this stage of the disease will not live very long—perhaps a few weeks at most.

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In sheep and goats, the clinical signs are harder to spot. The intestines become thick and less efficient at absorbing nutrients. Affected sheep continue to eat but lose weight and “waste away.” Although the disease causes diarrhoea in cattle, less than 20 percent of sheep show diarrhoea. In up to 70 percent of sheep, the disease may remain at subclinical levels, where individual animals never show signs of the disease but shed the agent in their feces and infect other sheep and contaminate the environment. In goats, weight loss, poor performance and occasionally clumpy feces are all that is seen. Affected animals usually show sign before they are 1 year of age.

Johne’s disease is generally described as having four stages:

**Stage I: Silent, subclinical, nondetectable infection.** Typically, this stage occurs in calves, heifers, and young stock under 2 years of age or animals exposed at an older age. Current tests (including fecal culture and serological tests) cannot detect infection in animals that young. Research to develop new tests to detect the disease in such animals is ongoing. This stage progresses slowly over many months or years to Stage II. It is possible that some animals recover from this early phase of infection.

**Stage II: Subclinical shedders.** This stage usually occurs in heifers or older animals. Animals appear healthy but are shedding *M. avium* subsp. *paratuberculosis* in their manure at levels high enough to be detected. Current blood tests are not reliable to detect Johne’s in animals at this stage. These animals pose a major but often hidden threat of infection to other animals through contamination of the environment. Stage II animals may or may not progress over time to Stage III.

**Stage III: Clinical Johne’s disease.** Animals in this stage have advanced infection, and clinical signs are often brought on by stress. Clinical signs at this stage include acute or intermittent diarrhoea, weight loss despite a normal appetite, and decreased milk production. Some animals appear to recover but often relapse in the next stressful period. Most of these animals are shedding billions of Johne’s-causing organisms, and fecal organism detection tests give positive results. Many animals are positive on serologic tests as well. Clinical signs may last days to weeks before the animals progress to Stage IV.

**Stage IV: Emaciated animals with fluid diarrhoea.** This is the terminal stage of the disease in which animals become extremely thin and develop bottle jaw. Animals culled to
slaughter in this stage may not pass inspection for human consumption due to disseminated infection.

In the typical herd, for every animal in Stage IV, many other cattle are infected. For every obvious case of Johne's disease (Stage IV) among dairy cattle on the farm, 15 to 25 other animals are likely infected. The clinical case represents only the "tip of the iceberg" of Johne's infection.

In other ruminant species, the progression of the disease may occur more rapidly with weight loss as the only visible sign of infection.

**Epidemiology**

In an endemic herd, only a minority of the animals develops clinical signs; most animals either eliminate the infection or become asymptomatic carriers. The mortality rate is about 1%, but up to 50% of the animals in the herd can be asymptptomatically infected, resulting in losses in production. Once the symptoms appear, paratuberculosis is progressive and affected animals eventually die. The percentage of asymptomatic carriers that develop overt disease is unknown. Johne's disease usually enters a herd when healthy but infected animals (Stage I or II) are introduced. Cattle are most susceptible to the infection in the first year of life. Calves most often become infected by swallowing small amounts of infected manure from the calving environment or udder of the cow. In addition, calves can become infected while in the uterus or by swallowing bacteria passed in milk and colostrum. Studies have shown that up to 25 percent of calves are infected in utero if the cow is in Stage III of the disease. Calves may become infected by exposure to contaminated manure any time in the first year of life (e.g., from manure splatter to calves raised near adult cows).

Cattle of any age can become infected, though some age resistance does occur. This age resistance can be overcome by high doses of bacteria over time from sources such as manure-contaminated feed bunks or water sources. All ruminants are susceptible to Johne's disease. In addition, all infected animals shed the organism through feces, thereby creating a possible route of exposure.

**Pathogenesis**

The primary site targeted by Johne's disease is the lower part of the intestine known as the ileum. The wall of the ileum contains a large number of pockets of lymphoid tissue known as Peyer's patches that lie just beneath the interior surface of the intestine. Peyer's patches are clusters of macrophages and lymphocytes organized much like lymph nodes. Covering Peyer's

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patches are a layer of cells called M cells. These cells function to sample the content of the lumen of the intestines and pass antigens (bacteria) through to the underlying cells of the Peyer's patch to "show" these antigens to the macrophages and lymphocytes. This is a means of "educating" the cells in a young animal about its environment, and is a protective mechanism designed to help the animal become immune to pathogens in its environment.

Unfortunately, when M cells bring *M. avium* subsp. *paratuberculosis* to the Peyer's patch, the bacteria find an ideal place for growth. Macrophages in Peyer's patches engulf *M. avium* subsp. *paratuberculosis* for the purpose of destroying the foreign invader, but for reasons yet unclear, these macrophages fail to do this. After phagocytosis by tissue macrophages in Peyer's patches, the bacterium is confined within phagosomes and phagolysosomes. It appears to be able to disrupt phagosome-lysosome fusion and if fusion occurs, block the degradative actions of lysosomal enzymes and molecules via the structure and composition of its cell envelope and through the production of peroxidases. Inside a macrophage, *M. avium* subsp. *paratuberculosis* multiplies until it eventually kills the cell, spreads, and infects other nearby cells. In time, other parts of the ileum and other regions of the body are teeming with millions of the mycobacteria. How *M. avium* subsp. *paratuberculosis* neutralizes or evades the normally efficient bacterial killing mechanisms of the macrophages is unknown, although the unusually resistant cell wall of mycobacteria likely plays an important role.

A model for granuloma dynamics of bovine paratuberculosis. MAP crosses the intestinal barrier via M cell or enterocyte transcytosis (1) and is subsequently taken up by macrophages in a predominantly tolerizing (2a) or a pro-inflammatory (2b) lamina propria compartment, the state of which may be determined by interplay between different dendritic cells (DC) subsets and enterocytes in combination with antigens present at that particular time. Subsequently the granuloma will develop into a pluribacillary (2a) or a pauci-bacillary (2b) lesion respectively. Depending on reaching bursting capacity due to bacterial replication (3a) or the end of the natural lifespan of macrophages or non-MAP related causes of cell death (3b) the infected macrophages will die and release MAP and MAP antigens into the lamina propria. Free MAP will enter the intestinal lumen via fluid streams and/or will be taken up by macrophages and DC migrating to the lumen thus leading to shedding of MAP in feces (4). Cellular debris and free MAP antigens from the lesion will be cleared and lead to the formation of scar tissue characterized by multinucleated giant cells and essential devoid of MAP. MAP and MAP antigen may spread to different sites in the intestine and restart formation of a lesion or enter the afferent lymph (6) and migrate to the draining lymph node causing lymph node lesions or activation of T and B cells when taken up and properly processed by antigen presenting cells either on route or in the lymph node (7). Activated T cells and B cell derived antibodies as well as monocytes will enter the intestine via the arterio-venous capillary bed (8).


The animal's immune system reacts to the *M. avium* subsp. *paratuberculosis* invasion by recruiting more macrophages and lymphocytes to the site of the infection. The lymphocytes release a variety of chemicals signals, called cytokines, in an attempt to increase the bacterial killing power of the macrophages. Macrophages fuse together, forming large cells, called multinucleated giant cells, in an apparent attempt to kill the mycobacteria. Infiltration of infected tissues with millions of lymphocytes and macrophages leads to visible thickening of the
Tests for Johne’s disease (JD) can be divided into two categories: those that detect the organism and those that assess the host response to infection. The first category includes fecal smear and acid-fast stain, culture, and polymerase chain reaction (PCR) tests. There are no tests of metabolic products or unique antigens of Map. The second category, detection of host response, includes clinical signs in combination with gross and microscopic pathology and immunologic markers of infection, which include antibody response to Map (serology), delayed-type hypersensitivity (DTH) reaction, lymphocyte proliferation, and increased cytokine (IFN-γ) production. In the live animal, fecal organism detection tests (culture and polymerase chain
reaction methods (PCR)) are the most accurate diagnostic test. However, on a herd basis only about 40 percent of infected cattle will be disclosed by even the most sensitive fecal culture technique. The sensitivity of fecal culture is low because some infected cattle (Stages I and II) do not shed the agent in their manure or because some animals shed the agent only intermittently and can be missed at testing time.

Intestinal granuloma associated with *Map* (Note Mø and lymphocytis infiltration)
Diagnostic test performance depends on the stage of disease. JD in dairy cattle is clinically categorized into four stages. In Stage 1, animals are infected, asymptomatic, and no organisms are detected in feces. In Stage 2, animals are asymptomatic, but organisms can be detected in feces. Stage 3 animals are symptomatic with weight loss and diarrhea. Stage 4 is advanced clinical disease, animals are symptomatic with signs of lethargy, emaciation and profuse diarrhea. Diagnostic tests will generally tend to perform better in individual animals in the later stages of the disease.

*M. avium* subsp. *paratuberculosis* is a slow-growing organism. Fecal culture on solid media requires 12 to 16 weeks for results. New liquid culture systems have reduced this time to as little as 5 weeks. PCR methods can detect the presence of *M. avium* subsp. *paratuberculosis* without its having to be grown. The test has the advantage that it takes less than 3 days and may not be affected by strain variations but has the disadvantages of higher cost and the potential of missing animals shedding only low quantities of bacteria.

Various serologic tests, including ELISA, agar-gel immunodiffusion (AGID), and complement fixation, detect antibody in the serum and can be used on a herdwide basis to screen for infection. Although less accurate than fecal culture, these tests are more rapid and less expensive. Serologic tests also work well to confirm clinical cases.

It is important to note that, as an accredited veterinarian, you should use only the USDA-licensed ELISA tests and USDA-approved laboratories.

In the dead animal, Johne’s disease may be diagnosed by culture and histopathology of the lower small intestine and associated lymph nodes.

The skin test takes advantage of the development of a delayed-type hypersensitivity (DTH) reaction to the intradermal injection of a mycobacterial extract, purified protein derivative (PPD). Intradermal skin testing has been and continues to be commonly used for the diagnosis of both bovine and human tuberculosis (Johnin test).

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<th>Stage II</th>
<th>Stages III, IV</th>
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<tr>
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<tr>
<td>Serology</td>
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<td>Maybe</td>
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Notes: PCR: polymerase chain reaction
IFN-γ: gamma interferon
References:

- https://en.wikipedia.org/wiki/Paratuberculosis
- https://johnes.org/johnes-disease-a-to-z/
- https://www.nap.edu/read/10625/chapter/5