More than a century ago, Johne and Frothingham discovered *Mycobacterium avium* subspecies *paratuberculosis* (MAP) in cattle, the causative agent of Johne’s Disease (Paratuberculosis), a chronic, debilitating intestinal infection. Johne’s Disease is found most often among domestic and wild ruminants but also has been reported in non-ruminants (1, 15). What’s more, the suggestion that MAP may play a role in human Crohn’s Disease has led to increased awareness in the medical community and the public (2, 13).

Johne’s Disease is a frustrating disease for both livestock producers and veterinarians and is associated with large economic losses in dairy cattle (14, 17). Losses are primarily due to decreased milk production and reproduction (5), and reduced salvage value of clinically affected animals. A national study of US dairies found that approx 22% of farms have at least 10% of the herd infected with MAP (12). In Michigan, recent estimates suggest that 54% of herds are test positive for Johne’s disease (8). The average loss ranges from $40 per cow (low clinical cull rate) to $227 per cow (high clinical cull rate).

In this report, we focus on new developments in diagnosis, vaccines, and control measures in the fight against Johne’s Disease. Through on-going national and international research programs, our ability to detect infected cattle is constantly improving and new vaccines are ready for testing.

### Signs and Transmission

Calves under 6 months of age are most susceptible to MAP infection. Although many animals in a herd may be infected, usually less than 5% of these will develop clinical disease, which is always fatal. Healthy, but MAP-infected animals (carriers) can actively shed the organism in their feces, thereby heavily contaminating the environment on the farm, without being recognized as infectious. The fact that MAP is able to survive in stagnant water, manure, and deep soil for up to a year makes the control of Johne’s Disease even more complicated (9, 10). Contaminated feed, milk and colostrum are thought to be the most important means of transmission (9, 10, 11).

### Diagnosis

Numerous diagnostic tools have been developed for MAP detection with several new tests on the horizon. Diagnosis of Johne’s Disease in individual suspect animals can be accomplished by fecal culture, serology, DNA probes, necropsy, and...
histology. Here, we focus on current methods used to diagnose healthy carrier animals and to determine true herd prevalence. Knowledge of true herd prevalence helps to decide which control measures should be instituted. It is important to note that there is no single, infallible test for Johne’s Disease and a combination of tests is often required.

**Serological tests.** Three tests to detect antibodies against MAP are available. The enzyme-linked immunosorbent assay (ELISA) is used for screening herds. Although relatively inexpensive (Table 1), ELISA sensitivity is correlated with stage of disease; cows in early stages of infection are often ELISA negative. The same is true for agar gel immunosorbent assay (AGID) (Rapid Johne’s test, ImmunoCell, Portland, ME). The third method, complement fixation test (CF), is not recommended for routine diagnostics, but is still required by some countries for export and import.

**What’s new.** The milk ELISA assay has been introduced as a rapid and easy screening tool (Antel BioSystems Inc., Lansing, MI). While this test is subject to the same drawbacks as other ELISA-based methods, it can be easily applied on a whole herd basis. When used in conjunction with other confirmatory tests such as polymerase chain reaction (PCR) assay (see below), the milk ELISA can provide an estimate of herd prevalence. In efforts to improve ELISA test sensitivity, scientists are exploring the use of new antigens and detection systems. Initial evaluations of these improvements show good promise and may dramatically improve ELISA assay sensitivity, while maintaining specificity. In contrast to detection of antibodies, the gamma interferon test measures responsiveness of blood lymphocytes to MAP antigens. Recent research suggests that this test may be more sensitive for detecting subclinical infections than ELISA-based tests, but samples must be handled more carefully and assayed within a few hours after collection. The gamma interferon test is not yet commercially available in the U.S.

**Identification of MAP.** Animals in early stages of infection typically do not have a detectable antibody response to MAP. Therefore, serologic tests are unable to provide evidence of infection. To compound matters, these animals might not excrete bacteria yet or excrete MAP only intermittently and in low numbers. In this case, bacterial culture of feces does not establish infection (Table 1). Bacterial culture of tissue is more sensitive (90±5%), but it requires a surgical procedure, which is in most cases not practical and not cost-effective.

**What’s new.** Polymerase chain reaction is becoming a widely used diagnostic tool in human and veterinary medicine. DNA probes have been developed which offer a means of detecting very low levels of MAP in diagnostic samples such as milk and feces. The PCR assay has also been used in conjunction with short-term liquid culture systems to increase sensitivity and verify that MAP is present in positive samples. PCR is commonly used in research to accurately assess the specificity and sensitivity of new diagnostic reagents, and to establish the most effective times for diagnostic testing in early phases of disease. While the PCR assay examines samples for MAP DNA, other new molecular based tests are looking for changes in host cell gene expression that could serve as indicators of infection. These studies are based on findings that gene expression profiles of bovine white blood cells from MAP-infected cattle are very different from those of healthy cows (4, 16).

**Vaccination**

An approved heat-killed MAP vaccine is available (Mycopar, Ft. Dodge Animal Health). However, vaccination only reduces clinical symptoms and does not prevent infection. Although vaccination is not considered part of control programs for Johne’s Disease in the U.S., there is ongoing research to develop safer and more efficacious vaccines to reduce the risk of infection within the herd.

**What’s new.** Recently, there have been promising approaches to develop DNA vaccines that might be safer, elicit more protective responses and would be less expensive (6). However, their ability to protect against MAP infection in cattle has not been tested yet. Recent developments in genetics have allowed scientists to construct defined and random mutants of MAP. Scientists are now targeting genes in MAP that have a role in survival and virulence. By deleting these genes, it is hoped that an effective modified-live vaccine might be developed that has little chance of reverting to a virulent strain. In addition, since genes that encode proteins recognized by the host immune system can now be selectively deleted, it should be possible to construct vaccine strains that are readily distinguishable from natural strains, allowing them to be used in control and eradication programs.

**Control and Herd Management**

Johne’s Disease is usually introduced into a herd when healthy but MAP-infected animals (carriers) are purchased by herd owners. Therefore, herds that are not infected should attempt to maintain a disease-free status by rearing their own heifer replacements. Otherwise test negative herds should serve as a source of low Johne’s Disease risk replacement animals. Collins et al. (3) have recently published a recommended test regimen for the detection of Johne’s Disease based on reason for testing (for example, herd status). Once MAP infection has been diagnosed in a herd, most control programs are aimed at breaking the cow to calf transmission of MAP, along with identification of infected animals for separation and culling from herds where needed (7, 18).

**What’s new.** A recent study from Dorshorst et al. (12) strengthened and updated a previous economic test-and-cull decision analysis model with current epidemiologic information. This novel ‘JD-Tree’ model incorporates costs and benefits of herd management changes, diagnostic testing, and different management actions based on test results to control Johne’s disease in commercial dairy herds. The model demonstrated that improving herd management practices to control
infection spread is often more cost-effective than routine testing; the recommendation was that not all herds should be routinely tested as part of a Johne’s Disease control program, but rather strict management controls put in place once the herd prevalence was known.

**Conclusion**

Johne’s Disease is considered to be one of the most serious diseases affecting dairy cattle it has an enormous economic impact on the dairy industry. Based on clinical reports, diagnostic records, historical reports and animal movements, the known prevalence of Johne’s Disease appears to be increasing worldwide. Although the causative agent of the disease, MAP, was discovered more than a century ago, there is no effective treatment or vaccine available. Therefore, control of Johne’s Disease still requires first and foremost effective herd biosecurity management protocols and judicious use of a combination of diagnostic tests. The novel consensus recommendations on diagnostic testing and the ‘JD-Tree’ model seem to be useful instructional tools helping farmers and veterinarians to make cost effective decisions about the control of Johne’s Disease in any individual herd. While the difficulties of working with a slow growing organism such as MAP have hampered research efforts, scientists are now making exciting progress and new vaccines and improved diagnostics are on the horizon. These new research efforts have been fueled by large well-funded programs on both sides of the Atlantic Ocean, Johne’s Disease Integrated Program (JDIP) in the U.S. and ParaTBTools in the European Union.

**References**


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1 Adapted from Collins et al. (3).
2 Cost/test is listed as a median for laboratory charges (30 laboratories offer the test).
3 I = bacterial culture from individual sample, P = bacterial culture from pooled samples [5 fecal samples/pool], n.r. = not reported.