



## An improved diagnostic test for Johne's disease ANH.01.09

**Project Title:** Enhanced Sensitivity of Detection of MAP in Bovine Feces by Integration of Bacteriophage-Based Capture with Loop-Mediated Isothermal Amplification

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**Background:** Effectively detecting subclinical Johne's disease would help reduce disease prevalence. More sensitive detection methods are required because most cattle with subclinical Johne's disease only shed the MAP bacterium in their feces intermittently and in low numbers. Using a modified version tuberculosis assay that included a PCR-based identification method for MAP, viable MAP cells have been detected in milk in just 48 hours. To be useful in beef cattle, the test must be modified to use manure samples instead of milk.

**Objective:** To improve MAP detection sensitivity without slowing test speed.

These researchers will immobilize a bacteriophage (virus) that targets MAP onto the surface of paramagnetic beads. When exposed to a MAP-infected manure sample, the phage will capture the MAP cells, infect them and propagate, producing 100 to 200 new phage particles. These new phage will be detected by a DNA assay for the MAP-specific bacteriophage. The new method is expected to detect MAP in bovine manure at level of 1 MAP cell per gram of manure, within 4 h.

**Implications:** Effective detection of subclinical MAP infected animals and herds would be of tremendous value in reducing the prevalence of bovine Johne's disease.

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