Part 1
Johne’s Disease Overview
A concise summary of the latest facts about Johne’s disease and recommended methods for diagnosis and control.

Prevalence - The slow epidemic

Dairy cattle
- Most experts agree that roughly 80% of U.S. dairy herds are infected with *M. paratuberculosis* (The 1996 USDA survey reported that 22% of U.S. dairy herds had at least a 10% within herd infection rate; herds with lower infection rates were not detectable by the survey methods).
- The infection is most common in the larger dairy herds that have rapidly expanded or regularly buy dairy replacements.
- The infection continues to spread among herds because few herds have instituted biosecurity programs to avoid purchase of infected cattle.

Beef cattle: cow-calf
- 8% of cow-calf operations in the U.S. test positive (USDA survey).
- Johne’s disease appears to be more common among registered seedstock herds (clinical impression of multiple experts).

Economic impact

Dairy cattle
- JD decreases milk production: the effect on production increases with progression of infection and is correlated with ELISA result levels.
- JD decreases lifetime production of cows due to premature culling.
- JD decreases slaughter value of the carcass.
- JD slows genetic improvement of herd due to involuntary culling.
- JD increases overall herd cull rate.
- The economic impact of JD on the herd increases each year as infection prevalence increases.
- JD decreases the value of seedstock when buyers learn about the *M. paratuberculosis* infection status of the herd.

Beef: cow-calf
- Limited reports on effects on growth or fertility
- Slows genetic improvement of herd due to involuntary culling
- Decreases value of seed-stock when buyers learn about Johne’s disease status
Pathogenesis

- Calves are most susceptible: resistance increases with age
- Adult cattle can be infected if exposed to large doses of *M. paratuberculosis*
- Infection starts in the ileum after ingestion of *M. paratuberculosis*
- Incubation period varies from 1 to 10 years: most cattle show signs at 2\(^{nd}\) or 3\(^{rd}\) lactation
- Infected cows excrete *M. paratuberculosis* in their manure, milk and colostrum
- Fecal contamination of drinking water (ponds) is a very efficient means of spread
- Fecal contamination of feed spreads the infection (same bucket for manure & feed)
- Poor farm hygiene increases infection transmission rate
- In the latter stages, the infection is disseminated beyond the GI tract

Diagnosis

- Clinical signs (diarrhea and weight loss) can resemble many other diseases
- Herd signs is often simply poor performance in spite of good nutrition
- The best test depends on the testing purpose and type of business – see table on the next page.
### Recommended test regimen for the detection of paratuberculosis in cattle on the basis of herd type and testing purpose (adapted from Collins et al. JAVMA, Dec 15, 2006)

<table>
<thead>
<tr>
<th>Testing purpose</th>
<th>Dairy</th>
<th>Seedstock</th>
<th>Cow-calf</th>
<th>Beef</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herd classification (infected or not infected)</td>
<td>Bacterial culture by ENV-HEY or ENV-LIQ</td>
<td>Bacterial culture by ENV-HEY or ENV-LIQ</td>
<td>Whole-herd testing, target testing, or bacterial culture by ENV-HEY or ENV-LIQ</td>
<td>Whole-herd testing, target testing, or bacterial culture by ENV-HEY or ENV-LIQ</td>
</tr>
<tr>
<td>Precise estimation of within herd prevalence</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
<td>NR</td>
</tr>
<tr>
<td>Control disease in herd with known infection, high prevalence (&gt;10% positive results on ELISA), and clinical disease, or owner is concerned</td>
<td>ELISA</td>
<td>Bacterial culture by IND-HEY or IND-LIQ</td>
<td>ELISA</td>
<td>Bacterial culture by IND-HEY or IND-LIQ</td>
</tr>
<tr>
<td>Surveillance (estimation of biological burden)</td>
<td>Bacterial culture by ENV-HEY or ENV-LIQ</td>
<td>NR</td>
<td>Confirmatory testing of clinically affected, suspect cattle</td>
<td>NR</td>
</tr>
<tr>
<td>Eradication (eliminate <em>M. paratuberculosis</em> infections from a herd)</td>
<td>Bacterial culture by POOL-HEY, POOL-LIQ, IND-HEY, or IND-LIQ</td>
<td>Bacterial culture by POOL-HEY, POOL-LIQ, IND-HEY, or IND-LIQ</td>
<td>Bacterial culture by IND-HEY or IND-LIQ</td>
<td>Bacterial culture by IND-HEY or IND-LIQ</td>
</tr>
<tr>
<td>Confirm a clinical diagnosis in herds with no prior confirmed cases of paratuberculosis</td>
<td>Necropsy, bacterial culture by IND-HEY, IND-LIQ, or IND-PCR</td>
<td>Evaluation of biopsy specimens, necropsy, bacterial culture by IND-HEY, IND-LIQ, or IND-PCR</td>
<td>Necropsy, bacterial culture by IND-HEY, IND-LIQ, or IND-PCR</td>
<td>Evaluation of biopsy specimens, necropsy, bacterial culture by IND-HEY, IND-LIQ, or IND-PCR</td>
</tr>
<tr>
<td>Confirm a clinical diagnosis in herds with a prior confirmed case of paratuberculosis</td>
<td>ELISA or bacterial culture by IND-HEY, IND-LIQ, or IND-PCR</td>
<td>Evaluation of biopsy or necropsy specimens or bacterial culture by IND-HEY, IND-LIQ, or IND-PCR</td>
<td>ELISA or bacterial culture by IND-HEY, IND-LIQ, or IND-PCR</td>
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</tr>
</tbody>
</table>

**IND-HEY** = Bacterial culture of fecal samples obtained from individual cattle; samples are cultured on solid HEY agar medium. **IND-LIQ** = Bacterial culture of fecal samples obtained from individual cattle; samples are cultured by use of any commercial liquid culture system. **POOL-HEY** = Bacterial culture of pooled fecal samples (5 samples/pool); pooled samples are cultured on solid HEY agar medium. **POOL-LIQ** = Bacterial culture of pooled fecal samples (5 samples/pool); pooled samples are cultured by use of any commercial liquid culture system. **ENV-HEY** = Bacterial culture of fecal samples collected in accordance with the Uniform Program Standards from areas in which cattle commingle; samples are cultured on HEY agar medium. **ENV-LIQ** = Bacterial culture of fecal samples collected in accordance with Program Standards from areas in which cattle commingle; samples are cultured by use of any commercial liquid culture system. **IND-PCR** = PCR assay of fecal samples obtained from individual cattle. **POOL-PCR** = PCR assay of pooled fecal samples. **ENV-PCR** = PCR assay of pooled fecal samples. **NR** = Not recommended.
Control

**Dairy herds** - the program I think is most effective and affordable
Owners MUST do BOTH parts

**Part #1: Calf management**
- Calves should be born in a clean maternity pen reserved exclusively for healthy ELISA-negative cows.
- Calves should be removed from cows in less than an hour from birth.
- Calves must be fed 4 quarts of high quality, hygienically collected, colostrum from a single ELISA-negative cow in less than 6 hours. This means a colostrum banking system must be established.
- Calves must be housed away from the adult herd - no access to adult herd manure.
- Calves must ONLY be fed pasteurized milk until weaning: NO WASTE MILK.
  - Option #1 = milk replacer
  - Option #2 = on-farm pasteurizer
- Calves must be fed water and feed free of manure contamination.

**Part #2: Regular herd testing**
- Test ALL cows one time during every lactation
  - Option #1 – early in lactation (15 - 45 DIM)
  - Option #2 – late in lactation (30 days before dryoff)
- Cull cows with strong-positive ELISA results at the end of their lactation
- Label and manage as infectious those cows with lower ELISA results (they might survive for another lactation but must be recognized as being an infection risk)
- ELISA-positive cows that are kept in the herd
  - Must calve in a separate pen
  - Must NOT donate colostrum
  - Should be culled if clinical signs of JD other problems develop

?? How about vaccination ??

Vaccination is only appropriate for the heavily infected herd with limited options or resources for JD control because vaccination:
- Does not stop infection, only decreases incidence of clinical disease.
- Interferes with TB testing.
- Requires regulatory paperwork from State Veterinarian (not allowed in many states).
- Causes unsightly lumps in cows that frequently abscess & drain.
- Is hazardous to use - ask vets who have been stuck with a needle while vaccinating.
- Prevents use of immunological tests like ELISA.
Biosecurity = Keeping Johne’s disease out of the herd

- Herds get infected only by buying infected cattle.
- Pre-purchase testing for Johne’s disease is today’s standard of veterinary practice.
- Testing the herd of origin is much more reliable than testing only the purchased cattle.
- Options below in order of decreasing risk of buying *M. paratuberculosis* infected animals (see Collins et al. JAVMA, Dec 15, 2006 for a more comprehensive chart).

<table>
<thead>
<tr>
<th>Options</th>
<th>Risk</th>
</tr>
</thead>
<tbody>
<tr>
<td>No testing</td>
<td>VERY risky; &gt;5% chance, for each purchased animal of being <em>M. paratuberculosis</em>-infected</td>
</tr>
<tr>
<td>ELISA test the individual animal before purchase - don’t buy anything from herds with cows positive by ELISA</td>
<td>Slightly less risky than not testing; more confidence in negative tests on older animals than heifers</td>
</tr>
<tr>
<td>Quarantine and test after purchase: ELISA + culture 2x @ 6 mo interval</td>
<td>Lowers risk and a sound policy for several infectious diseases of cattle</td>
</tr>
<tr>
<td>Partial test on herd of origin: ELISA on 30 2(^{nd}) lactation or older cows</td>
<td>Low risk of Johne’s disease in any animal from such herds but not 0% risk</td>
</tr>
<tr>
<td>Whole herd ELISA or fecal culture on the herd of origin</td>
<td>Very low risk of Johne’s disease if herd tests 100% ELISA or culture-negative</td>
</tr>
<tr>
<td>Buy only from test-negative status herds (level 2 or higher)</td>
<td>Lowest possible risk for purchase of <em>M. paratuberculosis</em>-infected herd replacements</td>
</tr>
</tbody>
</table>

**Laws & Ethics**

Johne’s disease is reportable in many states. If the laboratory does not report results directly to the State Veterinarian, then the veterinarian has a legal obligation to. The veterinarian’s license is at risk.

**Zoonosis Issue**

*M. paratuberculosis* either is or is not a zoonotic pathogen (infection transmissible from animals to humans). There is ample evidence to suggest it might be. However, the medical community has not yet labeled the cause of Johne’s disease a zoonotic agent. If it is NOT a zoonosis, then Johne’s disease is a common and pesky problem of modest economic importance in animal agriculture. One the other hand, if it IS a zoonosis, then it represents a serious threat both to human health and animal agriculture. Veterinarians must be cognizant of the latest information on this emerging issue.
A research team from the University of Wisconsin, School of Veterinary Medicine is conducting a field trial on Johne’s disease control in nine Wisconsin dairy herds ranging in size from 75 to 1,400 cows since January 2002. All herds in this so called “demonstration project” are on the same control program: standard herd *M. paratuberculosis* infection control management, ELISA testing every cow once during each lactation, culling cows that test strong-positive at the end of their lactation and labeling and managing the other ELISA-positives to maximize production income for the producer but minimize chances for *M. paratuberculosis* infection transmission.

### The specific management changes required in the program are:

**Maternity pen management**
- Separate, “clean”, pen for exclusively ELISA-negative cows
- Never allow sick cows in maternity pens; clean or for ELISA-positives

**Calf management**
- Remove calf from cow as soon as possible, ideally within 1 hour
- Feed 4 quarts of clean colostrum within 6 hours (3 qt for Jerseys)
- House calves well away from the adult cattle

**Colostrum management**
- Use only colostrum collected from ELISA-negative cows
- Take extra care for to assure hygienic colostrum collection
- Promptly feed or freeze colostrum

**Calf care & feeding**
- Feed only pasteurized milk until weaning either as milk replacer or as on-farm pasteurized milk
- Maintain good hygiene in calf rearing facilities and insure no manure contamination of feed and water

**Herd testing program:**
- Serum ELISA on all cows during every lactation
- Record ELISA numerical result or interpretation in the cow’s computer record
- Visibly label all ELISA-positive cows
- Manage cows based on ELISA result as follows:

<table>
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<tr>
<th>ELISA result*</th>
<th>Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>Negative</td>
<td>Calve cow in clean maternity pen and use colostrum</td>
</tr>
<tr>
<td>Low to medium-positive</td>
<td>Calve in segregated maternity pen; discard colostrum; consider not keeping heifer calf</td>
</tr>
<tr>
<td>Strong (high)-positive</td>
<td>Finish the cow’s lactation, do not breed back, cull at dry-off for slaughter only</td>
</tr>
</tbody>
</table>

*All ELISAs can and should be interpreted quantitatively*
Timing of testing:
Some herds use a pre-dryoff testing scheme where blood samples are collected from cows such that ELISA results are available before dryoff, specifically before use of any dry cow treatments. Pre-dryoff testing has the advantage of getting test results as close in time to calving as possible and at a time when culling decisions are normally made.

An alternative scheme is fresh cow testing (15-45 DIM). In this scheme, ELISA results are obtained before breeding. Cows that test ELISA strong-positive can be labeled DNB (do not breed). This effectively designates them as culls at the end of their lactation with the added advantage that when culled and not pregnant they are unlikely to be sold as dairy replacements thereby infected another dairy herd.

All herds are visited four times during the year by the project veterinarian, Dr. Vic Eggleston, assuring good program compliance. Herd DHI data is collected electronically from AgSource and processed by the Food Animal Production Medicine unit of the School of Veterinary Medicine. Fecal samples are collected and cultured by the BACTEC method but herd owners are not given the results to avoid culling bias.

The objective of the study is to reduce the within herd prevalence of *M. paratuberculosis* infections using only herd management and ELISA testing for serum antibodies to detect and manage infectious cows. The purpose is to demonstrate to veterinarians and producers that Johne’s disease control is both possible and affordable.

Results:
Overall within-herd apparent prevalence, as determined by either ELISA or fecal culture, has not yet changed in our demo herds but significant apparent prevalence reduction in 1st lactation heifers has been achieved. Over the past 12 months, heifers raised in herds after full implementation of the control program have entered the herds as milking cows and some have graduated to their 2nd lactation. The lower rate of ELISA-positive 1st lactation cows in the previous 12 months compared to before start of the program is statistically significant (P<0.001), providing solid evidence that the control program is succeeding (see adjacent figure). All 9 study herds observed a significant reduction in ELISA positive 1st lactation cows.
The same comparison of 1\textsuperscript{st} lactation cows based on fecal culture results provides independent confirmation that the Johne’s disease control program is effective (see adjacent figure). Since herd owners were not given the fecal culture results they were not making culling decisions based on that information. Among all 1\textsuperscript{st} lactation cows in the 9 study herds 17.6\% were fecal culture-positive at the start of the project while only 10.2\% were culture-positive in 12 months prior to April 1, 2007 (p<0.01). This also demonstrates that although the ELISA does not detect all of the cows shedding \textit{M. paratuberculosis} in their feces, and only the strong ELISA-positive cows are culled, use of the ELISA to detect the most infectious cows for culling or management is a useful adjunct to herd management changes to decrease the prevalence of infection.

In 2007 we are capturing data on 2\textsuperscript{nd} lactation cows as the program proceeds. As a steadily greater proportion of each herd is composed of cattle born after full implementation of the control program, we predict that a significant reduction of within herd \textit{M. paratuberculosis} infection prevalence as measured by ELISA and fecal culture will occur.

In addition to being effective, the control program is affordable; less than $10/cow/year added expense for ELISA results. Current studies are evaluating the accuracy of ELISAs on milk samples to support a Johne’s control program. If found sufficiently accurate, routine use of milk ELISAs could further reduce the costs of this type control program. Herd management changes are essentially considered best management practices and are easily justified in that they are effective at controlling multiple pathogens transmitted from cows to calves by the fecal-oral route.

Acknowledgements
This project has been made possible by financial and in-kind contributions from multiple sources. Most notable among these is the Wisconsin Milk Marketing Board which provided the first funding to launch the project. That money was matched by the USDA-APHIS-VS when the study herds joined the National Johne’s Disease Control Demonstration Project. In-kind support has been provided by the nine participating producers and the School of Veterinary Medicine.