

## Characteristics of milk ELISA results for Johne's disease in US dairy cows

Byrem, T. M.<sup>1\*</sup>, H. D. Norman<sup>2</sup> and J. R. Wright<sup>2</sup><sup>1</sup>Antel BioSystems, Lansing, MI 48910-3815<sup>2</sup>Animal Improvement Programs Laboratory, Agricultural Research Service, USDA, Beltsville, MD 20705-2350<http://antelbio.com>**INTRODUCTION**

- Johne's disease (JD) in cattle is caused by neonatal infection with *Mycobacterium avium* ssp. paratuberculosis and clinically, is characterized by granulomatous inflammation of the small intestine that fatally obstructs nutrient absorption and utilization.
- Although current tests for JD are considered ineffective in diagnosing early infection, most subclinical cases however, can be effectively detected with repeated testing over the course of an animal's productive lifespan.
- As a result, antibody-detection ELISA applied to milk samples routinely collected by Dairy Herd Improvement (DHI) has become a widely used tool to diagnose and control JD.

**OBJECTIVE**

- To determine relationships between data from JD milk ELISA and DHI dairy records for improved diagnosis and management.

**DATA & METHODS**

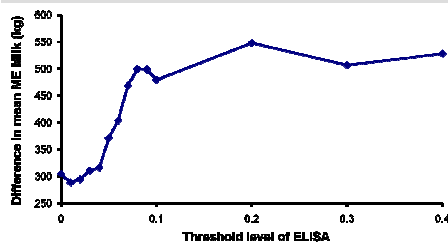
- DHI milk samples were commercially tested from 2002 to 2008 by antibody-capture ELISA (Parachek®, Prionics USA, La Vista, NE) with low diagnostic sensitivity (<50%) and high specificity (>99%).
- Testing strategies included:
  - Sporadic testing of clinically suspicious animals
  - Split-herd testing of animals at specific stages of lactation
  - Whole herd testing on single DHI test days
- JD ELISA results were available for 196,412 samples from 696 US dairy herds in 16 states.

**DATA & METHODS (cont.)**

- ELISA scores were matched with lactation and pedigree data from AIPL's national dairy database resulting in 101,849 total records. A subset of data (n=42,778) from select herds (n=25) represented repeated whole-herd testing.
- Several cutoff levels of ELISA scores were examined to define positive and negative JD status.
- Daughter-dam pairs were analyzed for vertical and environmental transmission of JD infection.
- SAS GLM was used to examine the effect of JD status on traits:
  - Milk
  - Fat, fat percent
  - Protein, protein percent
  - Somatic cell score (SCS)
  - Days open
  - Days in milk for lactation
- Effect of herd, year, and parity were absorbed in the model.

**RESULTS**

- Least squares means of difference in mature equivalent milk yield between positive and negative ELISA animals by cutoff level



- Effects on milk production were observed at an ELISA score cutoff level of 0.05. The current cutoff level for commercial JD diagnosis is 0.10.

**RESULTS (cont.)**

- Least square mean difference between positive and negative ELISA animals using a milk ELISA cutoff of 0.10.

Milk (kg)	-508*
Fat (kg)	-18.4*
Fat (%)	0.01
Protein (kg)	-15.1*
Protein (%)	0.00
Somatic cell score	0.1*
Days open	5.6*
Day in milk	-15.1*

- Differences in all traits between positive, negative and untested contemporaries were significant (P<0.0001)

- \*Significant difference between positive and negative cows (P<0.0001)

- Cows testing positive for JD by milk ELISA had reduced production and days in milk, and higher somatic cell scores and days open.

- Relationship of daughter-dam infection with JD (0.10 cutoff; n=16,001 pairs)

Dam ELISA	Daughter ELISA	
	Negative	Positive
Negative	94.7	5.3
Positive	90.8	9.2

- Milk ELISA test prevalence was 74% higher in daughters from test positive dams compared to daughters from test negative dams.

**RESULTS (cont.)**

- Distribution of JD positive ELISAs (0.10 cutoff)

Test Year	All Herds		Select Herds	
	Records	% JD positive	Records	% JD positive
2002	3,858	3.8	1,364	2.2
2003	8,287	4.1	2,501	2.2
2004	12,170	3.8	4,919	3.2
2005	14,614	4.0	6,129	4.1
2006	20,622	6.5	7,361	7.0
2007	25,715	6.5	10,380	6.6
2008	16,853	7.5	6,821	7.8
Parity				
1	45,954	3.8	19,464	3.5
2	26,823	6.7	9,760	6.8
3	15,002	8.1	5,283	9.2
4	7,873	7.7	2,766	8.5
5	3,728	7.2	1,279	7.7
6	1,617	5.7	548	6.9
≥7	1,122	6.7	375	5.9
Days In Milk				
1-60	12,498	4.7	4,044	4.0
61-120	8,573	3.9	2,526	2.5
121-180	7,965	3.5	2,466	2.3
181-240	11,257	4.5	3,640	4.0
241-300	23,220	5.2	9,081	5.6
301-360	18,536	6.6	8,684	6.3
≥361	20,070	8.4	9,034	8.3
All Groups	196,412	6.1	42,778	5.6

- JD test prevalence by milk ELISA has increased since 2002, is higher in cows after their first lactation and is lower during mid lactation.

**CONCLUSIONS**

- Negative associations between JD milk ELISA score and dairy performance records support a significant predictive value for infection.
- The diagnostic ability of milk ELISA for JD could be improved by integrating DHI dairy records and strategic testing.
- Daughters from infected dams are at higher risk of testing positive by milk ELISA.
- Despite organized efforts to control JD, test prevalence has increased over the last 6 years.