OPP Trial

Editor's Note: Minnesota's OPP Eradication Trial has received significant exposure over the past year, most recently via a related poster presented by Dr Devi Patnayak at the annual meeting of the United States Animal Health Association and American Association of Veterinary Laboratory Diagnosticians held in Kansas City this fall.

The poster tells the story of why the "Elitest" ELISA was brought to the U.S. by the MN Veterinary Diagnostic Laboratory. This new test plus recent OPPV genetic susceptibility and transmission research at USMARC form the basis of the eradication trial. Trial flocks are now completing their 2nd round of tests and early results are encouraging from those that have been able to keep young lambs segregated post-weaning. The authors of the poster have kindly granted permission for *The Shepherd* to reprint contents of the poster.

New Directions on Caprine Arthritis Encephalitis Virus/Ovine Progressive Pneumonia Virus Serology

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ABSTRACT

Caprine Arthritis Encephalitis virus (CAEV) and Maedi-Visna virus/Ovine Progressive Pneumonia virus (MVV/OPPV) are genetically and antigenically closely related. These viruses are transmitted from infected adults to younger animals via respiratory secretions and, to a lesser extent, from dam to kids/lambs via colostrum and milk. Historically the control strategies for these diseases have centered on serologically testing animals followed by culling or by segregating positive animals and orphaning newborns. Among tests available to identify positive animals, ELISA and AGID are the most commonly used serological tests. Various commercial ELISA test kits are available for the detection of antibodies to CAEV/MVV/OPPV. Three separate sheep flocks have experienced cross-reactivity to cELISA and AGID possibly due to recent use of chlamydia bacterin for abortion control. This presentation describes the use of "Elitest" ELISA for detection of antibodies to CAEV/MVV/OPPV. This test was used to confirm and eliminate this cross-reactivity and is now being used in an OPPV Eradication Trial in collaboration with the USDA, MN Board of Animal Health, MN Lamb and Wool Producers, and the MN Veterinary Diagnostic Laboratory and College of Veterinary Medicine.

INTRODUCTION

Maedi-visna virus (MVV) and caprine arthritis-encephalitis virus (CAEV) are two small ruminant lentiviruses (SRLVs) which cause inflammatory and debilitating diseases of economic significance in sheep and goats. In the USA, 36% of flocks and 24% of sheep tested have been found to be infected with MVV.

TRANSMISSION:

Old school: Ewe to lamb via colostrum and milk; control aimed at preventing lambs from maternal contact.

New school: Old infected ewes pass the virus to young replacement ewes via respiratory secretions and probably shared needles, ear taggers, drenching guns, etc.

CONTROL STRATEGY:

Old school: Weaning at birth and raising lambs/kids as orphans. Test entire flock annually, cull positives or keep negatives as separate flocks.

New school: Test adult animals to determine infection rate. Test replacement ewe lambs at 6-10 months and cull positives. Keep negative replacements away from adult flock forever. Test replacement flock annually with new replacement ewe lambs and cull positives.

SEROLOGY:

Due to lack of medical treatment and vaccines, eradication programs rely on periodic serologic testing, removal of seropositive animals and introduction of animals only from flocks documented to be SRLV test-negative flocks. Serological screening is done with enzyme linked immunosorbent assay (ELISA) and agar gel immunodiffusion (AGID) and Western blot is used for confirmation.

Several commercial ELISA kits are available for the detection of antibodies. In the USA, a commercial cELISA is widely used. We recently observed false positive reactions in cELISA and inconclusive results with AGID, possibly due to cross-reactivity with chlamydia bacterin for abortion control. The use of "Elitest " ELISA by Hyphen Biomed Lab (Saman et al., 1999) to eliminate this cross-reactivity is presented here.

MATERIALS & METHODS

Sheep flocks: Animals from three different flocks were tested before and after vaccination with Chlamydia bacterin.

Elitest ELISA and cELISA: These tests were performed according to manufacturers' instructions.

Agar gel immunodiffusion: Selected samples were tested using CAE/OPP Virus antibody test kit from Veterinary Diagnostic Technology.

RESULTS & DISCUSSION

Results for different serological testing conducted on individual flocks over a period of time are given below. These test were conducted at different times in different labs.

	DEC 2011 Chlamydia Vaccine	APRIL 2012 AGID 6 inconclusive	APRIL 2012 cELISA 5 positive	SEPT 2012 AGID 7 inconclusive	SEPT 2012 cELISA 7 positive	SEPT 2012 Elitest ELISA 9 negative	DEC 2012 Chlamydia Vaccine	JAN 2013 qPCR 7 negative	MAR 2013 cELISA 3 positive	MAR 2014 Elitest ELISA 3 negative
1	Yes	NEG	POS	NEG	POS	NEG	Yes	NEG	NEG	NA
2	Yes	INC	POS	INC	POS	NEG	NA	NA	NA	NA
3	Yes	INC	POS	INC	POS	NEG	Yes	NEG	POS	NEG
4	Yes	INC	NEG	INC	POS	NEG	NA	NA	NA	NA
5	Yes	NEG	NEG	INC	POS	NEG	Yes	NEG	NEG	NEG
6	Yes	INC	POS	INC	POS	NEG	Yes	NEG	POS	NEG
7	Yes	INC	NEG	INC	NEG	NEG	Yes	NEG	NA	NA
8	Yes	INC	POS	INC	POS	NEG	Yes	NEG	POS	NA
9	RAM	NEG	NEG	NEG	NEG	NEG	RAM	NEG	NA	NA
10	RAM	NEG	NEG	NT	NT	NT	RAM	NT	NEG	NT
11	RAM	NEG	NEG	NT	NT	NT	RAM	NT	NEG	NT

FLOCK "A"

NOTES:
- Prior to 2012, this flock had 15 years of documented OPP test-negative history.
- Of 25 animals tested with cELISA in April 2012, 5 were positive (20%).
- Ewe 2 had the highest positive readings, but necropsy detected no evidence of OPP.
- In addition to above tests, Western Blot failed to confirm OPP in "Flock A."

Flock B: Elitest ELISA could not be performed on these samples as the animals were culled.

	JULY 2012 Chlamydia Vaccine	AUGUST 2012 AGID 4 inconclusive	AUG 2012 cELISA 4 positive	SEPT 2012 AGID 5 inconclusive	NOV 2012 AGID 28 negative
1	Yes	INC	POS	INC	NA
2	Yes	INC	POS	INC	NA
3	Yes	NEG	NEG	INC	NEG
4	Yes	INC	POS	INC	NA
5	Yes	INC	POS	INC	NA
6	Yes	NEG	NEG	NEG	NA

FLOCK "B"

NOTES:
- Prior to 2012, this flock had 5 years of documented OPP test-negative history.
- August 2012 was a partial-flock test (6 of 25 animals); 4 cELISA positives (16%).
- November 2012 was a whole-flock test of 28 animals; 100% AGID test-negative.
- In addition to above tests, Western Blot failed to confirm OPP in "Flock B."

	OCT 2011 cELISA 12 negative	JULY 2012 cELISA 12 negative	MAY 2013 cELISA 16 negative	OCT 4, 2013 Chlamydia Vaccine	OCT 12, 2013 cELISA 16 positive	NOV 2013 cELISA 11 positive	DEC 2013 Elitest ELISA 0 positive	DEC 2013 cELISA 5 positive	MAY 2014 Elitest ELISA 0 positive	MAY 2014 cELISA 2 positive
1	NEG	NEG	NEG	Yes	POS	POS	NEG	POS	NEG	NEG
2	NEG	NEG	NEG	Yes	POS	POS	NEG	NEG	NEG	NT
3	NEG	NEG	NEG	Yes	POS	NEG	NEG	NT	NEG	NT
4	NEG	NEG	NEG	Yes	POS	POS	NEG	NEG	NEG	NT
5	NEG	NEG	NEG	Yes	POS	NEG	NEG	NT	NEG	NT
6	NEG	NEG	NEG	Yes	POS	POS	NEG	POS	NEG	POS
7	NEG	NEG	NEG	Yes	POS	POS	NEG	POS	NEG	NEG
8	NEG	NEG	NEG	Yes	POS	NEG	NEG	NT	NEG	NT
9	NEG	NEG	NEG	Yes	POS	POS	NEG	POS	NEG	POS
10	NEG	NEG	NEG	Yes	POS	NEG	NEG	NT	NEG	NT
11	NEG	NEG	NEG	Yes	POS	POS	NEG	NEG	NEG	NT
12	NEG	NEG	NEG	Yes	POS	NEG	NEG	NT	NEG	NT
13	NA	Lamb	NEG	Yes	POS	POS	NEG	NEG	NEG	NT
14	NA	Lamb	NEG	Yes	POS	POS	NEG	NEG	NEG	NT
15	NA	Lamb	NEG	Yes	POS	POS	NEG	POS	NEG	NEG
16	NA	Lamb	NEG	Yes	POS	POS	NEG	NEG	NEG	NT

FLOCK "C"

NOTES:
- Of 41 animals tested with cELISA in October 2013, 18 were positive (44%).
- MNVDL continues to follow this out-of-state flock, which first tested in 2011.

Interference with blue tongue virus (BTV) serotype 1 and 8 vaccination has been reported in routine diagnosis of SRLV in goats (Valas *et al.*, 2011). According to this report, when SRLV response in uninfected goats following BTV-1/8 vaccination was measured by different ELISAs, it was found that ELISAs based on SRLV antigen produced in eukaryotic systems resulted in false positive samples for SRLV. The ELISAs based on antigen prepared in prokaryotic systems did not detect any SRLV antibodies in the same samples. The authors hypothesized that false positive responses in these SRLV ELISAs could be induced by immune response directed against non-specific proteins derived from BTV vaccine, which was lacking in ELISAs based on antigen prepared in prokaryotic cells.

CONCLUSIONS

This preliminary study, comparing results from two different types of ELISA tests, illustrates interference with chlamydia bacterin injection in SRLV diagnosis in sheep.

However, more studies need to be performed in order to prove this hypothesis that a mechanism exists. i.e. cross-reactivity with chlamydia vaccination, similar to what has been observed with BTV vaccination.

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