Practical BLV eradication measures for commercial dairy farms

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Abstract
The presentation is intended to provide the dairy veterinarian with a process by which Bovine Leukemia Virus (BLV) can be eradicated from a dairy herd. The paper will describe both biosecurity measures to limit horizontal transfer of BLV within a herd as well as testing procedures to identify BLV positive animals and sort by their likelihood of infecting others in the herd. There will also be a discussion as to why BLV-free status may become more desirable in our modern society, and how the large animal veterinarian is well positioned to lead eradication efforts.

Key words: bovine leukosis, eradication, screening test, confirmatory test, biosecurity

Background
The purpose of this presentation is to lay out a practical scheme to eradicate bovine leukosis from a dairy herd. The paper will describe both biosecurity measures to help slow transmission of infectious animals to “clean” animals, as well as discussing a testing program, to isolate and eventually cull infectious animals. The practices we will be discussing have been applied to 2 dairy farms in northeast Wisconsin, Dairy Dreams Dairy with 3,000 cows, and Pagel’s Ponderosa Dairy with 5,400 head.

Both dairies have had very active embryo programs, whereby a significant number of animals in each herd serve as surrogate recipients for embryos owned by commercial bull studs. It is important for the recipients to be BLV ELISA-negative in order to meet biosecurity requirements for foreign markets. As a result, both herds have been doing intensive BLV ELISA testing for any animals that will be potentially used as recipients. Historically, we found approximately 20-30% of each herd, lact>=0, to be ELISA-positive. These positive animals would remain in the herd and receive AI breeding to make herd replacements. Although we didn’t see a great need early on to reduce or eliminate BLV from our herds, we did start to wonder how much more effort we would need to apply to simply be free of the virus.

In 2017 we were approached by Dr. Tasia Taxis of the Michigan State University (MSU) Department of Animal Science, to see whether we would be interested in having one of our herds participate in a trial she was doing with several herds in Michigan. The purpose of the study was to find practical biosecurity and testing protocols to allow herds to become BLV negative. We eagerly agreed to participate, and enrolled Dairy Dreams in the study. The program offered some consultation on biosecurity measures for blood borne pathogens, such as BLV. It also looked at several different laboratory diagnostic tests to identify BLV positive animals and assess their relative contagiousness to their herd mates. This information then allowed us to experiment with different ways of handling positive animals while at the same time protecting the clean portion of the herd from becoming infected. The entire project has now been completed and has been published in The Bovine Practitioner. The title of the article is “Reducing bovine leukemia prevalence on a large midwestern dairy farm by using lymphocyte counts, ELISA antibody testing, and proviral load.” In this paper, I will be describing what we learned from the study and how it has been put to practical use.

Biosecurity measures
Historically in the U.S., we have based herd BLV control on biosecurity measures that will reduce the spread of BLV from positive to negative animals. While important, we concluded that even aggressive biosecurity measures are not likely to eliminate BLV from a herd in any reasonable amount of time. A similar conclusion was reached by Ruggiero and Bartlett in 2019. Although biosecurity may help with horizontal transmission, we also found suspected cases of either vertical or colostral transmission as well. In fact, as the herd currently stands, with less than 1% PCR positive, almost all new positive animals we find are the offspring of a BLV positive dam that has already been removed from the herd. Another limitation of relying entirely on biosecurity measures is animals are only tested annually. That means that an animal may have an opportunity to become infected after testing negative and spread the virus before being retested in the subsequent lactation. I believe it would not make sense to undertake BLV eradication while ignoring biosecurity measures appropriate to prevent viral spread—it would be like bailing out a boat with a leaky bucket. Addressing both avenues of transmission is necessary. The measures we use to prevent the spread of this blood borne pathogen include rectal sleeve changes, single use hypodermic needles and an organized program to control biting insects. I will explain our application of each in detail.

Rectal sleeve changes
This procedure is relatively straightforward. Sleeves used for heifer pregnancy checking are changed between each animal. A pack of 50 sleeves is stored under the shoulder protector, and fresh lubricant is used for each sleeve. If an ultrasound is used, the sleeve previously used is inverted over the probe and a new sleeve is pulled on the arm. In the case of adult cows, since all animals are ELISA tested each year, fresh sleeves are not changed unless the previous animal checked was ELISA positive. While the individual sleeve changes in the heifers does slow down the speed of a herd check by an estimated 20 seconds per palpation, the fresh lubrication on each sleeve leads to less wear and tear on the palpator and palpatee alike.

Injection procedure
Early in our biosecurity protocol development, we started a program of needle changes between each injection. While manageable, it was tedious, time consuming and carried a significant risk of accidental needle sticks for the operator. We then switched to using the Pulse NeedleFree injection system. This is a portable system that uses compressed gas to carry the material to be injected through the skin, thus preventing any
cross contamination of infectious material from one animal to another. The system is very portable and works well with a light backpack carrier, which allows the operator enough freedom of motion to not interfere with rectal palpation. The compressed gas in the barn is a small tank of compressed CO$_2$. Because of the ease of using the Pulse system, and the minimal reaction from the animal being injected, we have now switched to giving all timed-AI injections, and all vaccines in the parlor, while the animals are being milked. Because the parlors have ample compressed air available, we use this to run the Pulse guns and have no need to use CO$_2$ bottles in the parlor. The only needles used on either farm at this time are for individual cows or calves receiving antibiotic treatments.

### Biting insect control

Biting flies have been a challenge for cattle and their caregivers since cattle were domesticated. Historically, our fly control measures were like most dairies. We would have a vague plan as to what we were going to do, and then make desultory attempts at fly control until flies got out of control. Then we would spend the rest of the summer in a doomed effort to get the situation under control with heavy applications of insecticide. As we started to see that these insects were thwarting our BLV control efforts, we took a more strategic approach. We consulted with a PhD entomologist who specializes in agricultural operations. As part of this process, we did a complete walk-through of our sites, accessed the critical control points and formulated a plan. In other words, we treated the fly problem the same way we are trained to approach herd mastitis, reproductive programs etc. In this plan, we establish dates when various products should be started, such as larvicide, rather than waiting for flies to be seen. We also have areas, such as used bedding piles, that need regularly scheduled treatments. Finally, we have plastic panels, treated with insecticide, and mounted in several areas that allow us to see when mature insects are arriving. For each of these measures, we have a product, dose, application method and applicator all set up.

We now have very low levels of insect pressure throughout the summer months. Also, due to the strategic rather than reactive process, we spend considerably less money on fly control than we used to. Most of the pharmaceutical companies that sell fly control products have these resources available. The key is to establish a plan and execute the plan as designed.

### Diagnostic testing program

While we feel that the biosecurity efforts are important, we have also come to believe, based on our experience in the MSU study, that biosecurity measure alone will take decades to eradicate BLV from a herd. To make reasonable headway, it is also necessary to have not only a comprehensive testing program, but also a committed management plan to deal with the positive animals that are identified. Failure to cull or segregate infectious animals, based on their likelihood of transmitting the virus, will doom the testing program to failure.

#### ELISA screening test

Our plan was to follow the sound epidemiological principles learned in veterinary school. That meant we had to identify both a screening test as well as a confirmatory test. The MSU study helped us immensely in that area. We followed the principle that the screening test needed to be quick, inexpensive and have a very high sensitivity. For this purpose, we relied upon the milk ELISA test available from our DHIA lab. This test met our criteria. Since it was a milk test, we could have samples taken on all fresh animals at the time of the DHIA test, thus reducing the labor resources we would need to utilize blood sampling. The lab fee was $6.50/sample, which we felt was reasonable. Since the assay is measuring antibody levels, we should find all animals that have been exposed to BLV antigens, whether they had become infected carriers or not. Once identified as ELISA positive, no further ELISA testing will be done for this animal while she remains in the herd. She will be recorded in DC305 as ELISA positive for infectious animals, based on their likelihood of transmitting the virus, will doom the testing program to failure.

#### Figure 1: Dairy herd BLV prevalence as measured by number of lactating cows ELISA-positive divided by total cows tested has decreased with time. The total number of cows tested is indicated with each year.

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#### Figure 2: Daily dreams proviral load by category with time

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life and will be subjected to the confirmatory test at the beginning of each subsequent lactation.

PCR confirmatory test

After screening all fresh cows, we needed a test that would allow us to both identify truly infected cows, those having BLV DNA in their lymphocytes, as well as assess the cow’s potential to infect herd mates. For this, we settled on a new assay being performed by Central Star Labs known as the SS1 (“Super Shedder 1”) PCR test that is designed to measure a cow’s proviral load. This is a quantitative PCR assay that detects not only the presence of the provirus, but also the relative levels in the blood stream, which strongly correlates with the animal’s likelihood of cross-infecting herd mates. Results are reported as a ratio of copies of BLV provirus to copies of an endogenous control gene representing the bovine DNA. This test is performed on whole blood, as the BLV provirus is found in the lymphocytes. The cost of the assay is $10.00/sample. It meets the requirement of a good confirmatory test in that it is highly specific and not prone to false positives. All ELISA-positive cows are subjected to the SS1 test.

Originally, all animals SS1 >1.0 were culled. At this level, cows are producing as much viral DNA in their lymphocytes as they are bovine DNA. We then established a second cut point of SS1=0.5-1.0. These animals were designated DNB, but were retained in the herd until their production dropped below the 90 lbs. Finally, cows with an SS1 value = .01-.49 were retained in the herd but bred with beef semen. Over the span of 20 months, we were able to steadily lower the action levels as the population of SS1 positive cows was steadily reduced. We are now at the point where all SS1 positive animals are culled immediately after each monthly round of testing. For the last several months that has been 2-4 head.

Using this approach, the management team can control the speed of eradication by controlling the population of replacement heifers being raised. If the replacement population is tight, these “luxury culls” will be limited. We chose to raise enough replacements to allow for more rapid eradication. The main consideration in taking this approach is the expense of testing. As the disease incidence is allowed to reduce, a dairy will find fewer and fewer positive animals each month. Consequently, if a month’s worth of BLV testing costs $2,500 and one positive cow is found, it will have cost $2,500 to find one animal. We wanted to shorten the number of years we would need to test, to get this expense behind us. Consequently, we needed to raise enough replacements to support the cleanup and maintain herd size. Basically, the herd team needs to decide how quickly they want to get through the process, and then decide if they want to “pull the Band-Aid off” quickly or slowly.

One incidental point that has become very clear to us is that vertical and/or colostral transmission of BLV is a significant route of transfer. Of the small number of first lactation SS1-positive animals we continue to find; in almost all cases their dam was previously found to be SS1 positive as well and is no longer in the herd. Since our long-established herd program uses the dam’s own colostrum for her calf, and no SS1-positive cows have delivered herd replacement calves since the start of the study, we have taken no further actions to address this route of potential infection.

Discussion

Up to this point, the discussion has been centered on how to eradicate BLV. Perhaps it would now be useful to ask if the result justifies the cost and time involved. This is an individual discussion to be had between the herd management and veterinary advisors. The literature contains many studies that demonstrate an association of BLV with lowered production, mastitis, lameness, etc. While I believe this to be true, despite all the extensive testing we did in the study, we did not personally see a strong association between SS1-positive cows and health conditions or production. I think it would be difficult to convince a client that it is worth the time and expense if the only consideration was milk production. The one health condition we saw obviously and clearly associated with BLV were cows condemned at the slaughter plant for malignant lymphoma. Once again, it would be hard to justify all the time and expense for the sole reason of avoiding these expensive culls.

Our original motivation, as mentioned previously, was to eliminate BLV from complicating our extensive commercial embryo program. To that end, we have been pleased by the rapid results. However, this won’t be an overriding factor in most commercial herds.

Perhaps some interest will come from producers and veterinarians that become more focused on producing “clean food”. Although there are papers that suggest an association between BLV virus and human mammary tumors, there are also many studies refuting that. Another issue that of potential concern is BLV is caused by a retro virus, the same category as human HIV, and that may be a stigma we want to avoid, if possible. It is hard to predict future markets and concerns, but to whatever degree this should become an issue for the dairy industry, I feel comfortable that we have found a path to relatively rapid eradication that would serve to help protect market access.

Finally, I think the most significant interest could arise from producers and veterinarians who want to be proactive from an animal welfare or standard of veterinary care point of view.
This is particularly true as the industry and profession strive to conduct business in a way that is acceptable to the evolving milk and cheese consuming public. If a veterinarian took the approach that there is not enough time to use alcohol wipes to dry treat cows, and as a result 1 or 2 recently dried cows develop toxic mastitis, he will likely be held accountable. If, however, a veterinarian cross-contaminated a clean cow with an infectious dose of BLV virus on a shared sleeve, and the cow was condemned for lymphoma years down the road, there is not likely to be the same type of outcry, due to the remoteness of the 2 events. However, in our modern society where only 1% of us farm, and 99% become more and more removed from the farm, we are increasingly judged by a population who is concerned about clean food and sound animal care. Furthermore, they have probably never met a large animal doctor, but would like to think that our standard of veterinary care compares reasonably to the standard of care they receive from their physician. Under that light, knowing that some very small but not insignificant number of our patients become contaminated with BLV virus and develop cancer, it would be hard to explain that we were too busy to change sleeves. As far as biosecurity with needles is concerned, we are still working our way through a global pandemic which has resulted in many millions of doses of vaccine being administered. If a school, nursing home or business is lined up for blanket vaccinations, the expectation will certainly be for individual needles to be used, regardless of the extra time and expense required. This scrutiny of animal care is not likely to go away, particularly as more and more non-animal-based foods become available.

As with any change in our industry, this will not happen overnight. All significant animal care changes I have experienced in almost 40 years as a dairy veterinarian occur gradually. They usually start with an innovative veterinarian and equally innovative producer working together to create change that makes sense to them, regardless of convention. For some practitioners, wishing to be in front of new standards of animal care, this may be a viable service to provide to the client that shares a desire to move the dairy industry forward. If a veterinarian finds they would like to move forward with BLV eradication program in such a circumstance, I hope this paper is of value in serving as a viable road map.

References