Salmonella Dublin: what you can do to help your herds

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Abstract

Salmonella Dublin (SD) is a serotype of *Salmonella* that is host-adapted to cattle and can lead to persistent carrier infections. It can cause serious disease outbreaks with high morbidity and mortality in youngstock, and abortions or morbidity and mortality in older cattle. There are also food safety concerns related to SD, a zoonotic pathogen that can cause serious human illness or death. A high morbidity and mortality outbreak has been associated with people consuming unpasteurized milk, and it is identified as one of the top three *Salmonella* serotypes found in beef products, notably ground beef. It also has the potential to infect and cause serious illness in cattle care workers exposed to infectious secretions of SD infected cattle. Veterinarians need to be fully educated on the clinical disease presentations of SD as well as the current diagnostic tests to identify and manage this disease. Strategies to keep SD out of individual herds need to be discussed and implemented. For those herds where it is already present, a plan to control the transmission of SD needs to be in place as well as a monitoring strategy to ensure its success.

Key words: cattle, dairy, *Salmonella* Dublin

Résumé

Salmonella Dublin (SD) est un sérotype de Salmonella qui est l'hôte adapté à bétail et peut conduire à des infections porteur persistant. Elle peut causer de graves flambées de maladies à forte morbidité et mortalité dans youngstock, et d'avortements ou de la morbidité et de la mortalité chez le bétail adulte. Il y a également des préoccupations en matière de sécurité alimentaire liés au DD, un pathogène zoonotiques qui peuvent causer des maladies humaines graves ou la mort. Un taux élevé de morbidité et de mortalité flambée a été associée avec de personnes qui consomment du lait non pasteurisé, et elle est identifiée comme l'un des trois principaux sérotypes de Salmonella trouvée dans les produits à base de viande bovine, notamment le boeuf haché. Il a également la possibilité d'infecter et de provoquer de graves maladies chez les bovins care Travailleurs exposés aux maladies infectieuses les sécrétions du DD des bovins infectés. Les vétérinaires doivent être pleinement renseignés sur la maladie clinique des présentations du DD ainsi que les tests diagnostiques actuels afin d'identifier et de gérer cette maladie. Des stratégies pour garder SD hors de troupeaux individuels doivent être discutées et mises en oeuvre. Pour les troupeaux où il est déjà présent, un plan visant à contrôler la transmission du DD doit être en place ainsi qu'une stratégie de suivi afin d'assurer son succès.

Introduction

Salmonella enterica ssp. *enterica* Dublin (SD) is a *Salmonella* serotype that is host adapted to cattle and has been known for years to cause serious disease in cattle.[1] It is also a serious human pathogen that has been associated with a significant disease outbreak in humans who have consumed raw milk containing SD.[2] In recent years it has become one of the top three *Salmonella* serotypes isolated from ground beef as reported by USDA FSIS.[14] Since approximately 2006 all of the SD that the Animal Health Diagnostic Center (AHDC) has isolated are resistant to the majority of antibiotics that can be legally used.[3] This means that prevention of SD versus the treatment of individual animals needs to be the focus of large animal veterinarians. In a study from the Netherlands it was determined that in over 50% of SD outbreaks the disease became persistent in a herd.[15] Furthermore, a modeling paper from Denmark estimates that in 60% of the situations there is within herd spread with the introduction of one SD infected springing heifer.[9]

Recent data from the 2014 NAHMS dairy study estimates 8% of dairy operations had *Salmonella* Dublin antibodies present in bulk-tank milk.[5] There looks to be a large difference, though, by region as the West region (CA, CO, ID, TX, WA) had 52.1% of bulk tanks SD antibody positive versus the East region [IA, IN, KY, MI, MN, MO, NY, OH, PA, VT, VA, WI] which had 2.8% of bulk tanks SD antibody positive.[5] A study done specifically in New York State (NYS) on greater than 95% of bulk tanks showed less than 1% of bulk tanks were SD antibody positive.[13] In a collaboration between the New York State Cattle Health Assurance Program (NYSCHAP) and the Animal Health Diagnostic Center (AHDC) at Cornell University, College of Veterinary Medicine, a significant education effort for large animal veterinarians focused on SD was undertaken over the last several years. This was initiated in part due to the increase in the number of SD cases that the AHDC was receiving in which the veterinarian did not have SD on the differential list. The following information is part of that education effort to ensure that veterinarians have the tools necessary to assist their herds in dealing with SD.

Clinical Presentation

The most common clinical presentation in the Northeast has been respiratory disease in calves ranging in age from one week to eight months.[6] The other common signs noted

in these calves with septicemia are high fevers and depression although sometimes practitioners have reported that a farm just finds dead calves. There also may be animals which present with one or more of the following signs: hot and swollen joints, bloody or watery diarrhea, and neurologic signs. Mortality rates on farms that the AHDC has worked with can be quite variable but in the worst scenarios have been over 90% in a group of affected calves. Some of the variability in mortality appears to be related to level of management of the facility with those farms that have the poorest hygiene, nutrition, and ventilation having the highest mortality rates. Practitioners have also reported that some of the calves that recover from SD are unthrifty, have scruffy hair coats, and grow poorly.

Since other types of *Salmonella* that present primarily as enteric disease have been much more common on Northeast dairies it has been a mindset change for practitioners to include SD on their differential list for respiratory cases. Educational outreach efforts have focused on providing practitioners with the necessary background information on SD so that it is not missed if it appears on their client's herds.

On necropsy the most common findings reported by practitioners to the AHDC have been heavy, wet lungs with diffuse changes throughout the entire lung field, a swollen liver with rounded edges and maybe a mottled appearance, and intestines with signs of inflammation.[6] Many practitioners also note fibrin throughout the peritoneal and pleural cavity.

In adult cattle, clinical disease has been much less common in cases reported to the AHDC but there have been cases of enteritis and abortions that were proven to be SD. In one study from Great Britain[4] abortion was the predominant clinical sign in adult cattle diagnosed with SD.

Diagnostics

Agent Tests

On a live sick animal the most reliable diagnostic test that the AHDC has found is blood culture. The blood sample needs to be collected aseptically and inoculated immediately into specialized blood culture media. Contact your diagnostic laboratory to obtain the appropriate media. For a basic protocol on blood culture technique see the following web link: https://ahdc.vet.cornell.edu/docs/Blood_Culture_Technique.pdf

Other diagnostic specimens on live, sick animals that are appropriate to submit for bacterial culture are feces, transtracheal wash fluid, and potentially nasal swabs.[6] For cattle that have aborted, the AHDC has cultured SD from vaginal swabs submitted in Amies transport media. The AHDC has found that the specific type of enrichment media used for SD versus other enteric *Salmonella* has a large influence on the recovery rate.[a] There have been notable cases where fecal culture was negative for SD but either blood culture was positive for SD ante mortem or tissue culture was positive for SD post mortem. Since the AHDC does not currently offer a SD specific PCR the author does not have information concerning the performance of this test although other US diagnostic labs do offer this option.

On post mortem specimens, SD can be quite reliably cultured from organs such as lung, spleen, lymph nodes, and intestines. The AHDC recommends a full set of fixed tissues as well for histopathology to further confirm the diagnosis. On an aborted fetus it is recommended to submit a standard set of fresh and fixed tissues to allow a full complete workup. Consult your diagnostic laboratory for specific directions or see the following web link for directions: https://ahdc.vet.cornell.edu/docs/Ruminant_Abortion_Kit_Complete_Paperwork.pdf.

Antibody Test

Since 2012, the AHDC has offered a commercial ELISA* which detects the presence of antibodies specifically to SD although there is a small possibility of cross reaction with *Salmonella* Typhimurium. The framework of this test was originally developed and tested in connection with the Danish Veterinary Institute and it has been shown to be useful in their national SD control program.[7] This ELISA is an antibody test that can be used on serum or milk from individual animals or a bulk tank milk sample. For the individual animal the estimation of sensitivity and specificity varies based on age and cut-off used but is estimated to be between 45-74% and 89-100%, respectively.[7] Although the results of this ELISA could be interpreted on the individual animal level, the strength of this test really is when it is interpreted at the herd level. In other words, although the results of individual animals are obtained with this test, it is more useful to consider the results of all the animals in the group tested and make herd level decisions rather than individual animal decisions. If repeated testing of individual animals over a longer period of time, for example four months to one year, are performed then there may be some individual animal decisions that would have more value.

For bulk tank milk samples it is recommended that repeated sampling be performed over time. A study from Denmark estimated that if four bulk tank samples were collected over a 5-12 month period and analyzed with the SD ELISA, the sensitivity would be 95% and the specificity would be 96% assuming a 15% national prevalence.[18] The current recommendation of the AHDC is to perform four bulk tank samples over the course of a year.

Determining Your Herd's SD Status

The first recommendation for your clients should be to determine the SD status of their herd which will then set the stage for the next step. If there are animals with clinical disease similar to what is outlined above then using the agent tests such as bacterial culture on the samples suggested above is the best strategy. If there has been previous undiagnosed disease that is suspicious for SD and individual animals have

recovered then the use of serology could be considered on these animals and their cohorts. One important point in the selection of animals to test is that the time to the maximal antibody titer for SD has been estimated in calves to be between six to eleven weeks.[7] If there has been no evidence of clinical disease that is suspicious for SD then most herds that the AHDC has worked with have either chosen to use the ELISA antibody test on repeated bulk tank milk samples or on a cohort of heifers between four to six months of age. Table 1 outlines the sensitivity of various testing methods to determine the status of your client's herds.[7]

Keep SD out of Your Herd

If a herd is determined to be at low risk for having SD then the farm should institute strict written biosecurity protocols specifically aimed at keeping it out. In several Danish studies, the largest risk factors for a herd to change from a test negative SD herd to a test positive herd was the number of other SD positive herds in the area, the number of purchased animals from a SD test positive herd, and herd size.[8,11] If the farm is not purchasing any animals or bringing heifers home from a heifer raiser that has commingled animals from other farms then the risk of bringing in SD is lower. There are still other areas to consider, though, and one of the primary areas to target is to not allow vehicles (such as rendering trucks, livestock trucks, etc) or visitors with manure contaminated tires or boots access to cattle or feed areas. See the following link for a more detailed discussion of other areas to consider: https://ahdc.vet.cornell.edu/Sects/NYSCHAP/docs/SalmonellaCCPs.pdf.

If the farm is purchasing animals or bringing springing heifers home that have been commingled with other animals then there is a need to have some awareness if SD could be brought into the home herd. The ideal situation would be to have confirmation of the SD status of the herd of origin of purchased animals or of the herds that heifers are commingled with. This could be achieved in the manner outlined above for determining the herd status. This may not be practical in some situations and therefore individual springing heifers may need to be tested with the SD ELISA to detect antibody positive animals. As outlined above, the goal with this type of testing would not necessarily be to interpret individual animal results but rather to get an assessment of the cohort of animals. This would provide stronger evidence that this group of animals potentially all were exposed to SD and therefore need to be handled differently as they move through the calving pens. It should be stated that a single antibody test at only one time point does not allow a distinction between an animal that was previously exposed and cleared the SD infection and a potential SD carrier animal that could shed in the future. It should also be stated that there is a small percentage of carrier animals that do not have positive antibody titers.[7] With the above assumptions in mind, there have been individual herds in the Northeast that have chosen to test purchased animals so that they have a better notion of the risk of bringing SD into their herd. A few have chosen to more closely monitor any SD positive animals and retest them at some time point later to determine if their antibody level remains elevated.

Isolation of any introduced cattle whether purchased or returning from off the farm is recommended to allow for the detection of any clinical illness prior to commingling with other cattle. This isolation is recommended for many different pathogens and should be performed although it should be noted that specifically for SD, a carrier animal may not show clinical illness and can shed SD well beyond the normal two to three week quarantine period.

For heifer raising facilities trying to keep SD out can be challenging especially if there are many source farms. It is the author's opinion, though, that a heifer raiser and their veterinarian should have a written SD plan in place with

Table 1. Adapted from Table 3.18. Herd sensitivity (HSe) for different herd testing procedures.

Herd testing procedure	HSe
Bulk-tank milk LPS ELISA at cut-off OD=0.4	38%
Culture of dung-pits	45%
Drinking water cultures	5%
Bulk-tank milk filter cultures	7%
Fecal culture of animals with current or earlier signs of salmonellosis	38%
Serology of all young stock	100%
Serology of all young stock between 4 to 6 months	91%
Serology of animals with current or previous signs of salmonellosis	80%
Combination of bulk-tank milk ELISA and serology of animals with current or previous signs of salmonellosis	91%
Combination of bulk-tank milk ELISA and serology of all young stock between 4 to 6 months of age	99%
Combination of bulk-tank milk ELISA in 4 samples collected over 5 to 12 months	95%

Veling J, Barkema HW, van der Schans J, van Zijderveld F, Verhoeff J. Herd-level diagnosis for *Salmonella enterica* subsp *enterica* Serovar Dublin infection in bovine dairy herds. *Prev Vet Med* 2002; 53:31-34. Warnick LD, Nielsen LR, Nielsen J, Greiner M. Simulation model estimates of test accuracy and predictive value for the Danish *Salmonella* surveillance program in dairy herds. *Prev Vet Med* 77:284-303, 2006.

each source farm. On the simplest level this could solely state that the risks of bringing heifers home infected with SD from the heifer raiser have been discussed with all parties. In the Northeast, a fairly frequent mode of introduction of SD to newly infected farms has been from heifers raised at a commingled heifer facility. In my opinion, this is not necessarily a fault of the management at the heifer raiser but most likely a result of one of the source farms introducing SD to the heifer raising facility. If the heifer raiser does not have an SD plan in place, though, and is not aware that it existed in their facility then it has been the experience of the AHDC that the source farms start to solely blame the heifer raiser.

Ideally as part of a heifer raiser's SD plan they would know the SD status of all the source farms and continue to monitor this over time such as by repeated antibody tests on bulk tank milk samples. The AHDC has also worked with heifer raisers that have implemented a monitoring strategy which involved performing antibody testing on a small percentage of incoming calves or heifers from each source farm. See the following web link to a NYSCHAP document that outlines in more details strategies for heifer raisers to deal with SD: https://ahdc.vet.cornell.edu/Sects/NYSCHAP/docs/Calf_HeiferRaiserSDRecommendations_12_2012.pdf.

Control in an SD-Positive Herd

For those herds that have definitively identified SD in their animals a detailed risk assessment needs to be performed. The NYSCHAP modified a risk assessment developed by the Danish group[10] and it is available at the following web link: https://ahdc.vet.cornell.edu/Sects/NYSCHAP/modules/salmonella/salmonellasection2.cfm

Click on link labeled "NYS Modified Risk Scores" which brings up a Microsoft Excel® sheet that can be completed in each category based on the history, farm visit, and walk through. It is the author's experience that there are numerous benefits to the herd veterinarian taking the time to walk through each part of the facility and make sure that all the questions of the risk assessment are answered accurately. Completing this document gives the veterinarian the opportunity to explore the small details that may have been overlooked but are crucial to making progress and at the same time reinforces to the farm which critical areas they need to manage on a daily basis. Having all the data in the spreadsheet allows for a comprehensive analysis and a graphic summary of the highest risk practices.

Solutions to correct the deficiencies noted in the risk assessment as well as systems to monitor success should be included in a herd specific written SD control plan. One of the most important areas to focus on is to close the infection routes that expose newborn and young calves to SD.[10] As is the case with other numerous other pathogens shed by adult animals, if calves are not exposed to SD from adult carrier animals then a population of animals that are free of SD develops. Over time this SD free population becomes a larger and larger percentage of the herd. This is still dependent as well on making sure there are no other routes of infection such as from older heifers which may be shedding, from manure contaminated feed that is offered to younger heifers, or from contaminated fomites. One large Danish study[12] showed good calving area management as one of the primary factors that prevented the exposure of calves to SD.

As with any good control program, having some way to document success and to monitor this over time is critical. For SD, one obvious way that farms monitor success is with the lack of morbidity and mortality that is associated with SD. This may not be a sensitive enough monitoring tool, though, for some herds that are experiencing low levels of the disease. This is another area where the use of the antibody ELISA test has proven valuable to both the Danish group[10] and herds that the AHDC has worked with. Testing cohorts of heifers in the 3-6 month age range with the ELISA test for the presence of SD antibodies allows farms to have reasonably timely feedback to gauge the success of their calf control measures. If all calves are negative for SD antibodies then it reinforces to the farm the success that they have been able to achieve and helps to motivate them to continue. If there are calves that are positive for SD antibodies then it points out that there are breaks in the protocol and that management needs to review, revise, and retrain employees on the protocols.

The use of the SD ELISA as a monitoring tool was demonstrated in a small study[17] performed in NYS. The dairy herd in this study was able to effectively prevent new calves from being exposed to SD when all of their calving pen management protocols were in place and followed by employees even when SD positive cows were being calved out. This was documented by performing the SD ELISA on all calves at 3-6 months of age and on all lactating cows four times per year. The situation changed, though, when this herd went through a large expansion which overwhelmed the system and caused a break in the calving pen protocols as documented by quarterly risk assessments by the herd veterinarians. Graph 1 illustrates the percent of SD positive calves over a nine month period. Note the change in the percent positive for SD in July 2014 which correlated to the start of the expansion.

Conclusion

Large animal veterinarians need to be concerned with SD not only because of the cattle illness caused by this pathogen but also due to its zoonotic potential, food safety risk, and multi-drug resistance. Herd veterinarians can play a large role in helping their herds to have a plan in place to keep SD out or control new infections in an endemically infected herd.

Endnotes

[a]McDonough PL. Personal communication, 2012

[b]PrioCHECK® Salmonella Ab bovine Dublin. Prionics AG, Switzerland

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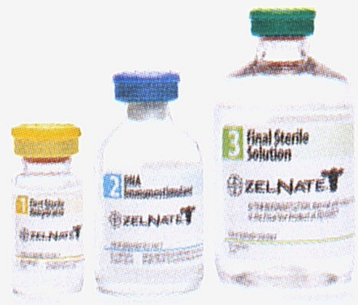

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