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Bovine Trichomoniasis

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INTRODUCTION

Trichomoniasis is a venereal disease of cattle that has caused significant economic losses in American cattle herds for many years. Its return to prominence in South Dakota in 2004-2005 is contrasted by the long-standing more endemic nature of the disease in other western states. The rate of diagnosis of trichomoniasis in the region has increased in recent years; whether this is due to an actual increase in numbers of affected herds or simply heightened awareness of the disease is not clear.[1]

Few detailed studies have been conducted to determine the actual prevalence of trichomoniasis in U.S. herds, but a 1990 survey of California beef herds revealed that 15.8% of herds had at least one affected bull, with a total of 4.0% of all bulls testing positive.[2] In a survey of Nevada samples in the mid-1980's, 26.7 - 44.1% of ranches that submitted samples had a positive bull.[3]

Economic losses in affected herds occur due to smaller and less uniform calf crops, costs of culling and subsequent replacements, and increased veterinary expense.[4] Models have put the drop in income from the presence of trichomoniasis in a herd at 22-37%.[4,5] In 2000, a group of Colorado producers figured the impact of trichomoniasis in their herd at $143.17 per cow.[6] Some estimates of the national loss due to this disease are as high as $650 million.[7]

Tritrichomonas foetus: THE ORGANISM

Tritrichomonas foetus is a single-celled protozoon that reproduces by binary fission (dividing itself). There are many different types of trichomonads (other protozoa in the same family of organisms). These include *Tritrichomonas vaginalis*, which causes venereal disease in humans, and several non-disease-causing species found in the gastrointestinal tract of domestic animals.[8]

T. foetus is pear-shaped and roughly the size of a head of a bovine sperm cell (Figure 1). It is characterized by three flagella (whip-like processes) at its front, and a wavy membrane over its entire length which ends in a posterior flagellum.[9] The three ("tri-") flagella ("-trich-," meaning "hair") give the organism its name. It is important to realize that Trichinella, which is a nematode (worm) associated with trichinellosis in swine is completely different from this organism.

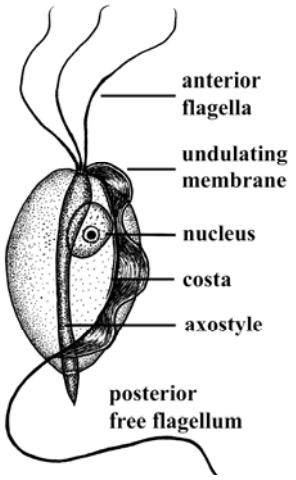

Figure 1. *Tritrichomonas foetus.*

This protozoon has an affinity for cattle, but has also been found in swine nasal and intestinal passages, and in the digestive system of cats. In both of these cases, the organisms are either considered non-pathogenic, or mildly pathogenic, such as in the cat, where it has been associated with chronic mild diarrhea.[10] *T. foetus* depends on the bull or cow to survive, and is rapidly killed by drying or high temperatures.[9] As such, survival in the environment outside the animal is very short-lived.

TRICHOMONIASIS IN THE BULL

Bulls infected by *T. foetus* are entirely without symptoms. Semen quality and sexual behavior are not affected. In bulls, the organism is only found on the penis and membranes inside the sheath. It localizes in the smegma, or secretions, of the penis, sheath, and end of the urethra.[11] *T. foetus* does not normally inhabit semen, but semen may become contaminated with organisms from the skin of the penis. Crypts, or microscopic folds within the skin surface of the penis and sheath, are sites for localization of the organism. Because these crypts become deeper as the bull ages, there is a definite association between age and infection: mature bulls are more apt to become infected and stay infected.[7] To further illustrate this fact, in the California prevalence study, the infection rate for bulls over three years old was 6.7%, while the rate for bulls three years old or younger was 2.0%.[2] Strong evidence exists that once a bull is infected with trichomoniasis, he is infected for life.[7]

TRICHOMONIASIS IN THE COW

As bulls infected with trichomoniasis are asymptomatic, the visible effects of the disease present themselves in the cows;[7] although, as is the case with bulls, cows do not appear ill while infected and overt signs of uterine or vaginal infection are not seen.

Infection of the cow takes place at breeding (see "Transmission" below). *T. foetus* organisms adhere to vaginal lining cells first with their posterior flagella, then with their body. The organism has adapted itself in several ways that enhance this attachment. Adherence to vaginal cells seems to be optimal at pHs of 6.0 - 7.5, which is the normal pH range of the bovine vagina. Also, *T. foetus* adheres better to highly keratinized (tougher) cells, which are present in greatest numbers during standing heat.[12]

After the protozoa attach to the lining cells of the vagina, they form colonies which spread to the uterus and oviducts (Fallopian tubes).[11] The uterus reacts to this colonization with an inflammatory response.

The time interval from initial infection to the maximum inflammatory response means that the cow or heifer likely will conceive to the breeding in which the infection was transmitted. Symptoms in the cow, therefore, depend on how quickly the inflammatory response takes place to the *Tritrichomonas* infection. For example, in a rapidly developing infection, inflammation may result in early death of the developing embryo. If this happens before day 18 after breeding, the cow may return to heat on her next 21-day cycle.[13] Usually this is not the case, as the inflammation due to trichomoniasis usually takes 50-60 days. This results in delayed (over 21 days) return to estrus, possibly with abortion of a small fetus

(which rarely is noticed on pasture), or formation of a pyometra, a pus-filled uterus. In some cases, infection may take an extended period of time to overtake the fetus and abortion may not occur until 7-8 months of gestation. This is relatively uncommon.[11]

Normally, infected cows will eventually mount an immune response and clear themselves of infection by two to four months, although there is a wide variation in this interval among infected cows.[7] This means that an infected female may lose her first conceptus, clear the infection, return to estrus, and conceive a pregnancy that goes to term.[11] The results of this scenario are a drawn-out calving period within the cow herd if bulls are left with the cows long enough.

The overall herd effects of trichomoniasis in cows can be summarized in this way:

1. Poor pregnancy rates (as much as 40% or more open cows at pregnancy-check) when bulls are pulled from pasture in a timely manner.

2. Spread-out calving season/late-calvers, especially when bulls are left in to breed for an extended time period. This may be seen in conjunction with an increased number of open cows; a majority of pregnancies do not correlate with the start of breeding season.[7]

3. Abortions are less commonly seen, as they are more apt to occur early in pregnancy.

4. Pyometras may occur in up to 5% of infected cows, and occur due to death of the fetus while the ovaries retain their corpora lutea.

5. Ultrasound abnormalities. Ultrasound examination may reveal evidence of dead fetuses or pyometras if cows are examined during the proper time window.

6. Returns to estrus on pasture. If careful observation takes place, an abnormal number of cows returning to estrus in the middle of the breeding period may be noted. Overworked, tired bulls may be an indirect indicator of this.

As previously mentioned, most cows mount a local immune response and clear the organism. This immunity means that subsequent breedings, even to an infected male, very well may result in a successful conception and pregnancy. This immunity is short-lived, however, and cows will be susceptible to infection in subsequent years.

Very rarely, an infected cow may carry a normal calf to term and not clear her infection, remaining infected, and a carrier to the next breeding season. The frequency with which this occurs is a source of frequent questions. Even a rare occurrence of this phenomenon may mean a source for carry-over from year to year, especially when large numbers of cows are involved.

The only published documentation of the prevalence of this type of long-term carrier is a 1987 case study of two California herds, one beef and one dairy.[14] A group of 280 cows

was examined from a herd of 3000 that had exposure to *T. foetus*-positive bulls. All 280 were pregnancy checked and cultured for *T. foetus*. Five of the 280 were found to be positive (1.7%) five months after exposure. Two of the five trichomoniasis positive cows were pregnant (five and seven months of gestation), or 0.7% of the exposed cows. The cow bred seven months had a normal calf and was still trich positive six weeks after calving. The other cow was not examined until five months post-calving, and she was trich negative then. So evidence exists that a cow was still trich positive six weeks after delivering her calf.

Further studies were not undertaken to determine how long the cow remained trich positive. It is unclear whether she remained positive long enough to be infective to a bull next breeding season once she returned to estrus post-calving. If one assumes the cow was infective into the next breeding season, this would represent a prevalence of 0.3% in this herd.

Also in the same case study, 366 cows in an infected dairy herd were examined. Thirty-five cows were found to be infected with *T. foetus*. Five (1.4% of the group) out of the 35 cows were pregnant, with gestational ages identified as 42, 85, 100, 100 and 185 days. Of the five, two cows delivered calves. One had premature stillborn twins and was still trich positive 41 and 63 days post-calving, but was negative 97 and 153 days post-calving. The second cow delivered a normal calf but was trich negative at 22 and 77 days post-calving.

Taken together, this represents two out of 646 cows exposed to trichomoniasis that remained positive post-calving, or 0.3%. Theoretically, in large herds this could play a role in "over-wintering' the disease when other control measures are taken. However, it is unknown whether, in a range situation, this prolonged infection would last over the interval between calving and breeding, or through the cow's estrus cycles when they resumed post-calving. So, while possible, the concept of an infected cow having a normal calf and becoming a carrier for next breeding season should be considered an exceedingly rare event.

Alternative causes of reproductive failure in beef herds to be considered in addition to trichomoniasis should include *Campylobacter fetus, venerealis* ("vibrio"), other venereal infectious agents, poor body condition in the cow herd, and sub fertile bulls. Of these, *C. fetus, venerealis* infection has nearly identical characteristics to trichomoniasis, with decreased pregnancy rates and spread-out calving seasons. However, pyometras post-breeding are not seen with "vibrio." Other infectious agents (*Histophilus somnus, Ureaplasma* spp., *Mycoplasma* spp., and IBR ["red nose"]) usually are much more sporadic in nature than either trichomoniasis or "vibrio." Pregnancy distribution in herds with poor body condition is shifted later, as these cows have a longer post-partum interval before they resume cycling. Bull infertility can be ruled out with breeding soundness exams of the bulls in question.[7]

TRANSMISSION OF TRICHOMONIASIS

T. foetus is transferred between infected animals in the course of breeding. It is estimated that anywhere from 30 to 90% of cows bred by an infected bull will become infected, as a relatively small number of organisms is required for effective transmission.[9]

Cow-to-cow transmission is only accomplished with the bull as an intermediary, as the organism does not survive long outside the body. Thusly, bulls become infected by breeding an infected cow once she resumes cycling after aborting.[15]

Bull-to-bull transmission has been theorized in a Canadian case study in which young virgin bulls were closely housed with older infected bulls. Presumably, younger bulls would be infected by mounting another animal in which an infected bull had just previously mounted.[16] This should be considered a rare event, but possible when "clean" and "dirty" bulls are penned in close confinement together.

It is technically possible for *T. foetus* to be mechanically transmitted in semen via its contact with infected skin of the penis and sheath, but procedures in place at AI centers virtually eliminate the probability of purchased semen acting as a means of trichomoniasis transmission.

Transmission of trichomoniasis into a previously uninfected herd, therefore, may occur when infected bulls are purchased, borrowed, or rented; when animals are co-mingled on common grazing tracts; or when cattle mix with neighboring herds through jumped or broken fences.[15]

DIAGNOSIS IN THE BULL

Diagnosing trichomoniasis in a herd is dependent upon having access to the right animals for testing. Since the bull is a chronic (lifetime) carrier, and cows clear the organism two to four months following infection, herd diagnosis usually is made by testing bulls.

Definitive diagnosis is made by observation of *T. foetus* in smegma samples (scrapings or washings). Culture techniques in which the organisms are encouraged to multiply are usually necessary for enough organisms to be present for identification.

It is recommended that bulls have at least two weeks of sexual rest before undergoing testing. Bulls are sampled in one of two ways. The most common method involves using a dry AI pipette attached to a syringe, and vigorously scraping the interior of the bull's sheath to obtain a smegma sample. The sample is then placed into a culture tube or pouch for incubation. Alternatively, a volume of saline or lactated ringers solution can be instilled into the sheath, the sheath opening held shut, and the sheath vigorously massaged before the residual fluid is collected for analysis. Similar test results are obtained with either method.[7]

The material obtained could be viewed directly under a microscope, but a much more sensitive method is to incubate the material in culture fluid for one to seven days and examine the fluid for the presence of the organism. Two different types of culture systems are commonly used: 1) InPouch TF™, which is a commercial pouch that the sample can be placed in, incubated in, and examined in; and 2) Diamond's media, which is placed into tubes into which the sample is also placed.

After incubation in either system, the culture fluid is checked under a microscope. *T. foetus* organisms, if present, are identified by their distinct features (described earlier: flagella and undulating membrane) and a characteristic rolling, jerky motion exhibited by the protozoa.

Studies have been performed comparing the sensitivity (ability to pick up positive bulls) of both the commercial pouch and Diamond's media. A recent study[17] comparing the two found the InPouch TF™ system to have an advantage over Diamond's media. Other experiments[18,19] demonstrate fairly equal results between the two. However, the pouch system has many other advantages over Diamond's media, including a long shelf life, the ability to transport the sample to the lab in the pouch rather than a separate transport media, and the ability to examine the pouch under the microscope directly.[7,17]

The finding of characteristic organisms in culture had long been considered to be 100% specific; in other words, if they were found, they were definitely *T. foetus*. However, in recent years, trichomonads, presumably contaminants of fecal origin, have been identified with new advanced molecular tools such as PCR. Such a case occurred several years ago in South Dakota when a supposedly virgin bull was found positive on culture. The organism was further typed molecularly and found not to be *T. foetus*, but a non-pathogenic contaminant. It is for this reason that all positive cultures at diagnostic labs are further characterized by PCR testing to assure the organism that grew was in fact *T. foetus*.

The sensitivity of a single sheath culture in an infected bull is estimated to be from 70-90%.[7] Therefore, in order to make a definitive diagnosis, it is recommended that bulls be sampled once a week for three weeks in a row. Serially testing bulls in this manner raises the sensitivity of the procedure to 99.8-99.9%. The consequences of a bull being called negative when in fact he is a carrier of trichomoniasis, necessitates the three weekly tests.

DIAGNOSIS IN THE COW

T. foetus organisms infecting cows may be demonstrated in mucus from the cow's vagina or cervix or from pyometra fluid. Samples are placed in culture fluid, incubated, and examined in the same manner as samples from the bull. In the case of pyometra, oftentimes that fluid is so teeming with organisms that it can be examined directly under the microscope for diagnosis.[7] Other avenues for diagnosis, such as testing vaginal mucus for antibodies to *T. foetus* are being explored, but none have proven practical or reliable yet.[20] Since cows clear the organism after infection, the sensitivity of testing cows is much lower than that for bulls, averaging between 58-75%.[8] It is recommended, therefore, that cows be tested as soon as possible after a problem is identified.

TREATMENT OF TRICHOMONIASIS

There is currently no approved, effective treatment for trichomoniasis in cows or bulls.

CONTROL AND MANAGEMENT OF TRICHOMONIASIS

Dealing with trichomoniasis can be thought of in two ways: 1) Managing the disease within an infected herd to minimize the biological (and thus economic) impact: "Biocontainment;" or, 2) Keeping the disease out of a non-infected herd: "Biosecurity."

Biocontainment: Management of the Infected Herd

Once diagnosed with trichomoniasis, a herd needs to implement measures that will reduce its impact next breeding season. These measures include:

1. Testing all herd bulls and removing positive bulls. Alternatively, producers may opt to sell all bulls and replace with virgin bulls. Implementing an AI program or increasing its scope is also to be considered.

2. Culling all open and late-calving cows. These are the cows most likely to be carriers of *T. foetus*. Cows destined to calve late are more at risk of being long-term carriers. If they are still infected after calving, then they will have less time to clear their infection before the bulls get put in (remember the cow still positive nine weeks after calving, above.)

    - Inherent in this step is timely pregnancy checking and possibly re-checking so infected cows are identified as soon as possible, and also, implementation of a short breeding season—90 days or less—is important.

    - Producers may wish to segregate cows based on gestation length when trich is diagnosed at pregnancy check time:

        a. Cows pregnant five or more months along are mostly out of the danger zone, although a very few may still abort.

        b. Cows pregnant less than five months should be pastured separately and watched closely. Their risk of abortion is greater than cows further along in gestation. Cull any of these cows that abort.

        c. Open cows with or without pyometras or other uterine abnormalities should be culled.[7]

3. Take proper biosecurity steps to ensure your hard work at managing the disease is not overtaken by an outside hazard:

    a. Ensure replacement animals are virgin bulls or tested clean.

    b. Maintain good fences.

4. Consider dividing herd into two: a clean herd and a "dirty" herd. This is only to be considered when facilities and personnel issues are optimal. The "clean herd" would only consist of virgin heifers and cows not exposed to infected bulls. The "dirty" herd would eventually be culled down by attrition (culling over time)

5. Vaccination. By itself, vaccination will not clear up a trichomoniasis problem. Currently, one killed whole-cell vaccine is available for use in the U.S. (Trich Guard™ or Trich Guard V5-L™, Fort Dodge Laboratories.) These products, when used according to label directions, show effectiveness in the female, but none whatsoever in the bull.[7] It is important to realize that the vaccines will not prevent transmission of *T. foetus* or infection with the organism, but it will limit duration of infection and result in more pregnancies being carried to term. Vaccine is best used in situations where bulls cannot be tested or removed, when desirable management practices cannot be employed, or in other high-risk situations.[4]

Biosecurity: Keeping Trichomoniasis Out of Your Herd

Keeping a herd free of trichomoniasis is dependent on managing potential sources of the disease:

1. Incoming cows should be virgin heifers from a reputable source. Current state regulations in South Dakota prohibit the sale of open cows for breeding. Purchase of bred cows or heifers should only be from reputable sources, preferably from areas in which trichomoniasis has not been found.

2. Bulls. Purchase only virgin bulls or bulls tested negative for three weekly tests.

3. Use AI and synchronization programs as much as possible to decrease the need for bulls.

4. Maintain good fences.

5. Avoid communal grazing situations if at all possible.

Biosecurity of individual herds has been aided by state regulations regarding bull testing and restrictions on movements of open cows. For example, rules implemented in South Dakota require three weekly trichomoniasis tests on all non-virgin bulls that are imported, sold within the state, leased, or borrowed. In addition, open cows may not be sold except for slaughter. Other states' regulations vary, including some states that mandate a test for all bulls within the state. In addition, Colorado has a voluntary herd certification program for trichomoniasis.

Trichomoniasis is a disease with a long history in the American West that has caused immeasurable economic loss to the beef industry over the years. It is only through understanding the disease and our responsibility and role in controlling it that we can greatly lessen the impact it has on future generations of cattle producers.

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