

Diagnosis, Prevention, and Control of Campylobacteriosis and Trichomoniasis

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Campylobacter infection in cattle otherwise known as bovine vibriosis is caused by *Campylobacter fetus fetus*, and Trichomoniasis is caused by a protozoan *Trichomonas fetus*.

Campylobacteriosis and Trichomoniasis are characterized by repeat breeding, delayed conception, and eventual development of convalescent immunity. Both the diseases are transmitted venerally and they are manifested by early embryonic loss or temporary infertility, and occasionally by abortions. Post-coital pyometra is occasionally seen with Trichomoniasis⁴.

The bull contracts both the diseases from infected females during breeding. The organisms grow in the crypts of the the penis and prepuce but do not enter the deeper tissues. A bull may be transiently infected or infected for years. More permanent infections are found in older bulls, probably because they have deeper penile and preputial crypts. Bulls develop no convalescent immunity from infection, since they can be reinfected after the organisms are removed by treatment. In beef herds, the bulls are not kept around very long, it is thought that carrier cows carry over the infection from one year to the next.

The bull is not affected directly by the infection, but in infected herds of susceptible cattle the burden of repeat breeding may inhibit his breeding capacity, resulting in temporary infertility or refusal to breed cattle in estrus.

Cows once infected usually overcome the infection or become immuned in a period of three to six months from infection.

Diagnosis of Campylobacteriosis and Trichomoniasis

History— Both the diseases are characterized by repeat breeding, delayed conception and eventual development of convalescent immunity. But sometimes this is not seen, since it requires all the bulls to be infected in the beginning of the breeding season, and that all of the females be susceptible.

Virgin-heifer test— An infected bull is bred to virgin heifers, then cervical mucus samples are collected for culturing. Cervical mucus samples are collected 12-19 days for *Trichomonas*, and between 18-30 days for *Campylobacter* after breeding.

Douche— The sheath is infused with approximately 200 ml of USP Saline or USP Ringer's lactate. The preputial opening is then occluded while the trapped fluid is massaged and then recollected. This fluid is centrifuged (2000 rpm for

10 minutes) and the sediment examined microscopically for *Trichomonas*. Fluid obtained from the prepuce is sent to the lab to culture for Campylobacteriosis and Trichomoniasis.

Swab— A teigland swab, or any guarded swab, is used to obtain a sample of smegma from the fornix of the bulls prepuce. The swab is rinsed in 5 to 10 ml of USP Saline or USP Ringer's, which is then centrifuged (2000 rpm for 10 minutes) and the sediment examined microscopically for *Trichomonas*. To isolate *Campylobacter* and *Trichomoniasis*, two different swabs could be sent to the lab to be cultured.²

Pipette— An AI pipette is introduced into the fornix of the bulls prepuce, and smegma is collected by scraping and aspirating. Vigorous scraping combined with alternating positive and negative pressure using a rubber bulb or syringe will aid in the collection of a suitable sample. The epithelial cells and smegma that accumulate in the pipette are mixed with 5 to 10 ml of USP saline or USP Ringer's lactate, centrifuged (2000 rpm for 10 minutes) and the sediment examined microscopically for *Trichomonas*.

The pipette method used in conjunction with culturing, for both *Campylobacteria* and *Trichomonas*, has been shown to be effective.²

FIGURE 1. An AI pipette with a syringe is introduced into the fornix of the bull's prepuce.

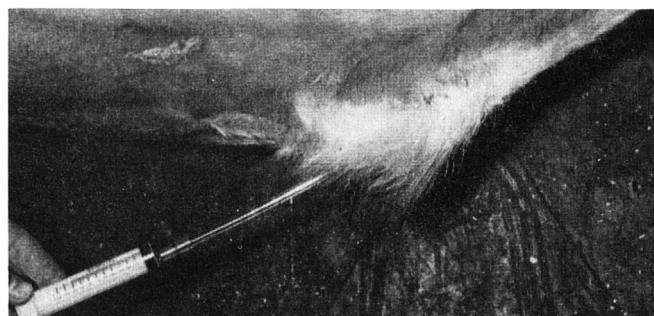


FIGURE 2. Smegma collected by scraping and aspirating using an AI pipette.



Culture— The sample collected through the pipette method is flushed into a sterile, chemical-free vial containing 2 cc of USP Saline or USP Lactated Ringer's solution, by aspirating the diluent in and out of the sampling pipette. It is important to use USP Saline or USP Lactated Ringer's Solution because other solutions may contain preservative which kill *C. fetus* and *T. fetus* organisms.²

Clark's transport medium is used for transport or shipment of samples to laboratories for culture of *C. fetus*.¹³ Clark's medium is ineffective unless it is microaerophilic environment is maintained; consequently, the suspended mucous samples should be injected through its rubber stopper.

Samples for culture for *Trichomonas* should not be shipped in Diamond's medium, but can be shipped in either USP Saline or USP lactated Ringer's solution provided they arrive at the laboratory and are transferred into Diamond's medium by 48 hours after collection.

Transport medium:

C. Fetus— Clark's medium

T. Fetus— USP Saline or USP Ringer's lactate.

Diamond's medium in the laboratory.

To examine for *T. fetus*, a small amount of fluid is aspirated from near the bottom of the incubated Diamond's medium and placed on a warm slide under cover slip, preferable phase contrast microscopy. If conventional microscopy is used, the light must be reduced to the lowest level with a condenser diaphragm. The characteristic jerky, undulating, erratic motility of the organism is seen at 100x and confirmed at higher magnification. The *Trichomonads* are examined under a microscope and identified over several days in the lab by their very irregular jerky, spiral motion of the fluid. A sample should be incubated for seven days before it is called negative. This method is 80 to 90 percent sensitive. (Ball et. al. 1984, Ben Duirant et. al. 1984). Bulls should be tested routinely for *Trichomoniasis* and *Campylobacteriosis* in herds with infertility problems, and in edemic areas, when they are evaluated for breeding soundness.

Bovine Campylobacteriosis

Bovine campylobacteriosis is a venereal disease of cattle caused by *Campylobacter fetus fetus* and is characterized by temporary infertility of the female.

Control

Control is based on three epidemiological facts; first, that transmission is venereal, secondly, the bulls remain permanently infected, and thirdly, that infected cows overcome infection, or become immune in a period of three to six months from service.³

Thus, if infected service is replaced by artificial insemination, 'self-cure' of the cows will occur. Exclusive use of artificial insemination with semen from non-infected bulls

is an efficient method of controlling the spread of vibriosis.

A question arises, "How long is it necessary to carry on an AI program in these herds?" It seems certain that in a majority of cows *C. fetus* will not survive a normal gestation, but Frank and Bryner (1953) recovered vibrios from a few cows as long as 196 days beyond the end of a pregnancy initiated by infected semen.¹¹ It would seem wise therefore to continue inseminating until every exposed cow has completed two normal pregnancies.³

Although this procedure can be utilized in most dairy herds it is useful in only a few beef cattle herds.

Prevention and Control Measures:

1. Exclusive use of AI. AI schemes prove highly successful and pose no problem in the case of the non-exposed females including the annual intake of clean heifers, these being effectively shielded from infection.
2. Natural breeding used on virgin heifers with a clean bull or a bull free of campylobacter infection. After the heifers have calved they may again be mated naturally to clean bulls.
3. Vaccination of all the female cattle where AI program is impractical. Vaccinate the female cattle 30-60 days before the breeding season. Annual vaccination is required to maintain maximal reproductive rates.
4. Vaccination of bulls with vaccines containing sufficient antigen was effective in decreasing the infertility observed in herds with vibriosis in Australia. A bull vaccination program should be approached with caution until additional research demonstrates its values.

The efficacy of a vaccine depends on four factors:

1. Adjuvant,
2. Amount of antigen,
3. Time of vaccination, and
4. Boostering *Adjuvant*.

Two general types of adjuvants are used. The most effective vaccine is a modified Freund's oil emulsion adjuvant. Water soluble alum and aluminum hydroxide precipitated vaccines do not provide a good immunity.

In the oil adjuvant vaccine, the antigen insert was adequate. Most aluminum hydroxide precipitated vaccines required two vaccinations to stimulate immunity in cows, and many of them are ineffective. Oil adjuvant vaccines provide significant protection for several months. However, the safest procedure is that of vaccinating approximately one month before the breeding season, and following the recommendation of the company.²

Bull Vaccination:

Researchers in the United States have used 5 cc of vibrin (Norden) for each vaccination dose, with reasonable amount of success. This is 2½ times the dosage recommended for cows. Two doses should be given three weeks to a month apart, just before the breeding season. Vibrin is the only

vaccine in the United States for which bull vaccination has been evaluated. Other vaccines should not be used for this purpose until dosage levels have been established.

Treatment

Cows usually overcome the infection or become immuned in a period of three to six months from infection.

Bulls

C. fetus is sensitive to streptomycin and it is evident from the work of Binns (1953), Basbech (1951), and Edgson and Scarnell (1955), that thorough application of streptomycin, chlortetracycline or chloramphenicol to the penis and prepuce of infected bulls is curative. Scarnell and Edgson first applied antibiotics to the extruded penis and prepuce under epidural anesthesia and then douched the preputial cavity with the same emulsion for four days.

Arthur (1953) suggested a simpler method in which the linings of the A.V. is smeared with chlortetracycline cream and the bull is then allowed to 'serve' into this several times daily for three consecutive days. This method is considered preferable because during penile erection, the crevices and folds of the prepuce and sheath are exposed and thus a maximum surface area is likely to be brought into contact with the antibiotic vehicle.¹⁰

The infection can also be effectively eliminated in bulls by antibiotic treatment. Dihydrostreptomycin is used in a dosage of 10 mg/lb of body weight infected subcutaneously, and 5 gm of a 50% solution of the same antibiotic applied locally to the penis and prepuce.³

Trichomoniasis

Trichomoniasis is a venereal disease of cattle, characterized primarily by early embryonic loss (infertility) and occasionally by abortions and pyometra.¹⁴ The etiological agent is a protozoan, *Trichomonas fetus*.¹

Cows and heifers which have been exposed to infected service fall into the following clinical groups:

1. Conceive and carry to term without clinical signs of infection developing.
2. Return to multiple services, but show no obvious signs of infection. Estrous periods may be regular or irregular.
3. Fail to conceive and develop an edematous condition of the endometrium with a mucopurulent discharge.
4. Conceive but abort at two to four months of gestation.
5. Develop pyometra with anestrus.

In most cases, initial service by an infected bull will result in the female contracting the disease. The protozoan localizes in the secretions of the vagina, uterus, and oviduct, initially does not interfere with the conception. Embryonic wastage may be due to the inflammatory response detectable in the uterus of heifers approximately 50 days post service by an infected bull. Once infected, females remain infertile for two to six months regardless of the method of service, i.e.,

use of non-infected bull or A.I. This infertility may be the result of several episodes of early embryonic loss or perhaps one episode of embryonic loss followed by a period of failure of conception or nidation due to an inflamed uterus. After a variable period of infertility, females become immune and will conceive and carry to term, even though bred by infected bulls. The duration of this immunity as well as the pathogenesis of reinfection varies with the individual cow.

Trichomoniasis is asymptomatic in the male. The protozoan localizes in the secretions of the epithelium lining the penis, prepuce, and anterior portion of the urethra. Some evidence indicates that young bulls do not become permanent carriers, but whereas the older ones do. However, it has been observed younger bulls fail to recover spontaneously.

Control:

Trichomoniasis is increasing in incidence throughout the United States and it is endemic in the Western, Southwestern, and Southeastern United States. In Texas, Florida, and Oklahoma approximately eight percent of the bulls sent to slaughter are infected.

In the Western United States, there is an epizootic of large proportions because of their vast open ranges. Mixing of cattle in these ranges and in pasture often result in trichomoniasis becoming widely disseminated. No vaccine is available for trichomoniasis, so it is more difficult to control than campylobacteriosis.²

Proven control methods generally fall into five broad categories:

1. Depopulation of infected animals is often of limited value because of economic considerations but should be considered when the disease is not wide spread or relatively few animals are involved.
2. AI is also effective and often used on a percentage of the affected cows. However, total change from natural service to AI is often impractical because of managerial or economic factors or both.
3. The herd can be separated into two groups, one containing exposed cows and the other unexposed cows. This method is potentially effective, but is generally not practical because of the difficulty and potential hazards involved in maintaining an infected herd in isolation.
4. Cows bred to suspect or infected bulls, or both, can be sexually rested before rebreeding. Following successful calving, cows are not bred until at least 90 days have elapsed following calving.
5. A method combining sexual rest and isolation is also reported to be effective. Following successful calving, a 90 day postpartum interval with no less than two normal estrous periods, cows then in the infected group are moved into the clean group.
6. A method of selective culling combined with the use of young bulls is also effective under field conditions. Cows failing either to conceive or to calve before the next

breeding season are culled.

Breeding is limited to bulls younger than four years of age that have been tested for trichomoniasis prior to use. This procedure is repeated for several years with the progress of the herd monitored by testing the bulls for trichomoniasis after each breeding season.¹

Treatment:

Dimetridazole 50 mg/kg of body weight daily for five days orally. Bulls often either do not like the drug in their ration or tend to go off feed for several days following the initial dosage. Rumen stasis is frequently observed during or following treatment of bulls on high concentrate rations. This can generally be prevented by feeding a diet high in roughage for several days before and during treatment.

Dimetridazole is also effective when given intravenously in dosages ranging from 10 mg/kg of body weight for five days to 50-100 mg/kg of bodyweight in a single intravenous injection. Dimetridazole is soluble only in relatively low pH solutions, and in fact the vehicle reportedly used was 10 to 20 percent sulphuric acid.

Response to injection of these acidic solutions varied among bulls with respiratory difficulty, ataxia, recumbency for a few minutes, etc. All these effects were transitory with no serious complications.

Ipronidazole 30 gm/bull and 15 gm/cow in a single dose is effective. The drug dissolved well in water, however it is easily oxidized, so it should not be compounded in metal containers or left standing more than a day or two, or exposed to excessive sunlight before it is used. Bulls should be pretreated with broad spectrum antibiotic for three or four days before treatment with ipronidazole to kill a micrococcus that is sometimes present in the prepuce. This bacteria inactivates ipronidazole making it an ineffective treatment. Three treatments with ipronidazole 30 G in 60 ml of sterile water given IM on the first day, and 15 G in 30 ml of sterile water given IM on the second and third day was highly effective.¹²

Metronidazole is reported to be effective when given intravenously; dosages ranging from 75 mg/kg of body weight for three successive injections at 12 hour intervals and to 10 mg/kg of body weight given once daily for 20 days.²

Prevention:

Trichomoniasis and campylobacter infection can be prevented by adequate screening, testing, and isolation procedures for all additions to an established herd. Prior to use, bulls unless virgin, should be tested by culture method at least three times at weekly intervals. Female additions should be cultured prior to entering the herd, unless coming from familiar herds or they are virgins.

Open cows following breeding by AI or natural service should be isolated from all bulls until calving and a 90 day postpartum period has elapsed and another culture examination performed.

Pregnant females should be isolated from bulls until 90 days postpartum and another culture exam performed.

Bulls should be tested prior to use on clean cows.

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