# Diagnosis of Ovine Abortion – Getting the Most Out of Your Diagnostic Laboratory

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# Introduction

When samples for ovine abortion workup are submitted to the veterinary diagnostic laboratory at North Dakota State University, there are four infectious agents that are primary target of our investigation. They are Toxoplasma gondii, Campylobacter fetus or jejuni, Chlamydophila psittaci, and Salmonella arizonae (as well as other Salmonella serotypes). When a sheep fetus or tissues from a sheep fetus are processed, several tests are run which will help us determine if any of these pathogens is responsible for the abortion. Ideally, the entire fetus and placenta should be submitted to the laboratory, and the pathologist of record is able to select tissues and samples for analysis. However, if the post mortem examination is done on the farm or ranch, the tissues to submit for optimal results include the brain. heart, lung, a piece of the diaphragm (the thin wall of muscle between the thoracic and abdominal cavities), liver, kidney, spleen, stomach contents (collect in a syringe), and heart sack or chest fluid (collect in a syringe). When possible, include the placenta, and a serum sample from the dam. Remember, most of these organisms can cause significant disease in humans, therefore, always handle sheep fetuses, tissues, and placentas with rubber or latex gloves.

#### Toxoplasma gondii

*T. gondii* is a parasite, a one-celled organism belonging to a group known as protozoans. In order for the organism to successfully survive in nature, it must spend part of its life cycle inside a member of the cat family,

domestic or wild. The cat excretes the infective form of the parasite, the oocyst, in its feces which is then ingested in contaminated feed by a susceptible ewe. This is why it's advisable to restrict, or, if possible, eliminate the access of cats to sheep feed locations. The classic feature (lesion) associated with T. gondii abortion is inflammation and tissue damage (necrosis) of the cotyledons on the placenta. In some cases, they may become calcified. This change can be observed with the naked eye (grossly) if the placenta is washed, and one of the suspect cotyledons suspended in a salt solution. The surface of the cotyledon unwinds and exposes the necrosis/ calcification which looks white and feels gritty. However, this will give a tentative diagnosis only, and it is necessary to submit fresh and formalinfixed placenta to the lab for confirmation under the microscope. T. gondii infection will also cause damage in the fetal brain, so it is important to submit brain for suspect Toxoplasma cases. Your veterinarian can help with the collection of needed tissues. Special staining procedures are available (immunohistochemistry) which allow the actual organism to be highlighted in infected tissue. A pathologist will view the stained sample under the microscope and identify the parasite. DNA from the parasite can be detected by a method known as the polymerase chain reaction (PCR). This procedure is not typically offered by all laboratories, but it is a tool than can help with diagnosis. Finally, it is possible to detect antibody to T. gondii in samples of fetal fluid, either from the chest or heart sack, and fetal blood. The presence of antibody to the organism in the fetus indicates exposure to the

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parasite, and identifies it as the cause of abortion. So, in a typical Toxoplasma abortion, observation of necrosis in the placenta and brain, followed by special staining of the tissue ought to identify *Toxoplasma* gondii as the cause. Polymerase chain reaction and serology are available as needed for confirmation. One final note: please remember that T. gondii is a human pathogen, and can cause severe disease in immunocompromised individuals. This group of people would include the elderly, those with chronic disease, cancer chemotherapy patients, organ transplant patients, anyone on therapy to suppress the immune system, AIDS patients, and the very young. In addition, it is capable of causing abortion in humans, therefore pregnant women should exercise care around lambing facilities, and limit or eliminate exposure to cat fecal matter (litter boxes, gardens, playgrounds, etc.).

## Campylobacter fetus/jejuni

Campylobacter is a bacteria which causes abortion in sheep. The classic gross lesions associated with Campy*lobacter* abortion are large, circular, white spots on and in the liver. Unfortunately, these areas of liver damage do not occur in all cases, and cannot be relied on for diagnosis. In addition, there may be microscopic evidence of fetal pneumonia, and inflammation in the placenta. The bacteria are best isolated from abomasal contents, so it is important to submit abomasal fluid along with tissue if the entire fetus cannot be submitted to the laboratory. It is possible to look at a wet mount of abomasal contents and actually see the comma-shaped Campylobacter bacteria. Premises become infected when the bacteria are shed in the fecal matter of a carrier ewe. This contaminates feed and water which are ingested by susceptible ewes. The bacteria gains access to the bloodstream and travels to the placenta where it causes tissue damage and abortion. This organism is not considered a cause of disease in humans.

### Chlamydophila abortus

C. abortus is the cause of enzootic abortion in sheep. The older name for this organism was *Chlamvdia psittaci*. but has been changes to the current designation. Typical changes associated with Chlamydophila abortion include inflammation of the placenta, brain, liver, spleen, lymph nodes and blood vessels. In some cases, the bacteria can be identified by special staining methods. Serological tests are available as well. Ewes become exposed by ingesting material contaminated with the bacteria, and it is usually the younger ewes which are affected. This is because older animals will develop immunity over time, and have some natural protection from the organism. When examining aborted fetuses, hemorrhages may be seen in muscle and subcutaneous tissue. There is often fluid accumulation in the thoracic and abdominal cavities. The placenta may be inflamed to the point of appearing leathery. A normal placenta is very thin and clear; in fact, it should be possible to see through normal placental tissue. The bacteria can be isolated using special culture techniques, but the disease is usually identified through observation of microscopic changes, serology or PCR. C. abortus has been reported to cause abortion in humans, therefore anyone working with aborted lambs or aborting ewes should observe good hygiene to limit exposure to potential pathogens. This would include the use of rubber or latex gloves, thorough handwashing, and reduction or elimination of exposure of pregnant women to sheep abortions.

#### Salmonella arizonae

*S. arizonae* is a bacterium which causes sporadic abortion in sheep. In fact, several types of *Salmonella* can cause abortion. Quite often, the ewe will show some signs of illness (fever, diarrhea) as well. Ewes usually recover, but can die of uterine infections. The best method of diagnosis is through culture of fetal tissues

collected at necropsy. Placenta, fetal tissues and vaginal discharge would all be appropriate samples for culture. The Salmonella organism is ingested by a susceptible ewe and then by means of the bloodstream infects the pregnant uterus. Microscopic changes in fetal tissues are related to widespread damage caused by the bacteria in blood. The diagnostic laboratory is able to use enrichment techniques to select for the Salmonella bacteria in suspect samples. Stains are then forwarded to the National Veterinary Services Laboratory in Ames. Iowa for serotype identification. Additional diagnostic techniques are available, but culture and histopathology remain the best method of diagnosis for Salmonella abortion. As was the case with Toxoplasma and Chlamydophila, Salmonella can cause disease in humans, and so observing proper hygiene is important.

# Implications

The aforementioned agents are considered the primary causes of infectious abortion in sheep. While they are not the exclusive causes, these four are part of any routine ovine abortion workup in the upper Midwest. While the clinical pattern of abortion may help focus the investigation, there is enough variation in how abortion outbreaks appear to warrant a search for all of these pathogens. Selection of a full set of tissues as mentioned in the introduction is critical, as well as submission of placenta and serum from the dam where possible. The laboratory can take this material and generate results which will identify or rule out the problem. If none of these agents are identified, the producer can use the "negative" diagnosis to look at other possible problem areas such as nutrition, environment, or genetics. Due to the potential for human disease, it is important that individuals at risk reduce or eliminate exposure to aborting sheep and aborted fetuses.