OVINE CHLAMYDIAL ABORTION IN ALBERTA

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Introduction

Ovine chlamydial abortion (enzootic abortion of ewes, ovine viral abortion) was first reported in North America in 1958 by Young *et al* (17) who described the infection in Montana sheep. Parker (9) characterized the organism isolated from that outbreak. Subsequently the prevalence and distribution of the disease in Idaho sheep were reported by Frank *et al* (2) and later, the infection was described in California and in Oregon sheep by McKercher *et al* (6). Meinershagen *et al* (8) reproduced the condition successfully in 11 of 19 pregnant yearling ewes inoculated with tissues from aborted chlamydiae-infected ovine fetuses examined by these investigators. A similar chlamydial agent has also been associated with abortion in cattle (13) and in goats (4) in North America.

Prior to this report, firm evidence of the occurrence of ovine chlamydial abortion as a flock problem in Canada has been lacking. The following report documents some of the gross histopathological, cultural, and serological features of the disease in three areas of Alberta in 1972 to 1974.

History

Flock 1 – This southern Alberta flock consisted of 420 ewes of mixed breeding. The only new additions during the past year were three Suffolk rams bought at auction in the fall of 1971. In late February, 1972 when the problem began, approximately 310 ewes had lambed normally. At this time, and without premonitory signs, approximately 25 of the remaining 110 animals aborted. Despite occasional vaginal discharge following abortion, most ewes remained clinically healthy and expelled the placenta normally, frequently with the lamb. No age related abortion rates were observed in the ewes.

Flock 2 – This central Alberta flock was composed of 700 ewes of mixed breeding. A portion of this flock had been purchased in 1972 from a southern Alberta breeder. The few abortions observed in the flock in 1972 were attributed by the owner to rough handling at shearing time and no post mortem examinations were performed. In 1973, approximately two weeks before term, 18 lambs were aborted; several of these were submitted for laboratory examination.

Flock 3 – This northern Alberta flock consisted of 100 ewes from three sources as follows:

Lot A consisted of 40 ewe lambs purchased in the fall of 1973 from a breeder in central Alberta. These animals were bred after purchase. Although lambing was not due to begin until mid-April 1974, ten abortions occurred from mid-March to the end of April. In addition, five premature weak lambs were born.

Lot B consisted of 52 two to three year old bred ewes purchased in the fall of 1973 from a breeder in central Alberta. One abortion occurred in this group.

Lot C consisted of eight pregnant ewe lambs purchased in the early spring of 1974 from a breeder in northern Alberta. No abortions occurred in this group.

Flock 4 – This flock, also located in northern Alberta, consisted of 214 ewes of mixed breeding from three sources as follows:

Lot A consisted of 200 ewe lambs purchased in the fall of 1971 from a breeder in southern Alberta.

Lot B consisted of six ewes purchased in the fall of 1971 from another southern Alberta breeder.

Lot C consisted of eight ewe lambs purchased in the fall of 1972 from a breeder in central Saskatchewan. These lambs were transferred to an intermediate location in central Alberta for three weeks before being moved to their final destination in northern Alberta where they were bred in early December 1972. The ewes in this group began to lamb in May 1973, after those in Lots A and B had lambed. Subsequently, there were three abortions in late pregnancy and three weak lambs delivered to ewes in Lot C. In the spring of 1974, more than half of

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the lamb crop from Lots A and B were lost as the result of abortions, weak lambs which died soon after birth, and deaths of normal lambs because of maternal neglect and/or exposure, ten to 14 days *post partum*.

Materials and Methods

Complete post mortem examinations were conducted on all lambs submitted to the laboratories. For bacteriological examinations, tissues including brain, liver, lung and spleen, as well as abomasal content and when possible, placenta, were inoculated onto 5% bovine blood, MacConkey's and Brilliant Green agar, incubated both aerobically and in an atmosphere of 10% CO_2 at 37°C for up to seven days.

For chlamydial isolation, small portions of fetal and placental tissues from all flocks were ground in 7 ml of nutrient broth containing 1.5 g of dihydrostreptomycin and centrifuged at 10,000 rpm for 20 minutes. The supernatant fluid was inoculated in 0.2 ml volumes into the yolk sacs of seven day embryonated chicken eggs which were then incubated for an additional seven days. Harvested yolks and allantoic fluid from embryos dead primarily on days four to six postinoculation, were passaged serially to a maximum of six passages. Tissues from Flock 1 were inoculated into ovine kidney tissue cultures and passaged twice.

Similar ground tissue and placental suspensions from Flock 1 were inoculated into prebled guinea pigs. At 21 days postinoculation, serum from these animals was examined using the complement-fixation (CF) test and a chlamydial group antigen (1).

Acute and convalescent phase blood samples from aborting ewes in Flocks 1 and 2 were drawn by jugular venipuncture at two to three week intervals into plain Vacutainer[1] tubes. Several weeks following abortion, single blood samples were obtained from seven randomly selected ewes from Flock 3. Resulting serum was examined by the CF test (1).

Tissues for histopathology were fixed in 10% neutral buffered formalin, processed routinely, embedded in paraffin and cut at 6 μ. Stains included hematoxylin and eosin (H & E) and azure eosin.

Results

Gross Pathology

Most lambs were aborted near term in a fresh state, but several lambs in each flock were markedly autolytic. In some cases live, weak lambs were born. At necropsy it was common to observe partially inflated lungs in animals found dead. Gross lesions included excess serous fluid in the thoracic cavity, gross enlargement of the liver with the presence of fibrin tags, and splenic enlargement. Many lambs did not present grossly detectable lesions at post mortem. Frequently the placenta was autolytic and unsuitable for examination. In other cases, however, cotyledons were necrotic or hemorrhagic; lesions of the intercotyledonary area included edema, necrosis and thickening and sometimes, the presence of a reddish fluid.

Histopathology

Histologically similar lesions were seen in most aborted lambs and in placentas from each of the flocks examined. The salient features of these lesions are summarized:

In the liver, reactions were most marked near portal triads, around central veins and along sinusoids. The reaction consisted of large accumulations of several cell types, including reticuloendothelial (RE) cells (Figures 1 and 2), lymphocytes, neutrophils and a sprinkling of eosinophils. Most livers had continuing hemopoietic activity.

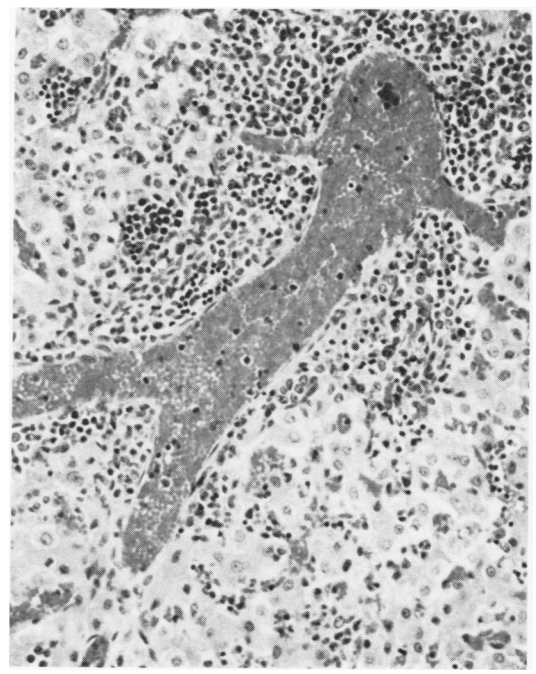

Figure 1. Liver showing characteristic RE cell activity adjacent to a vein. Cells with dense nuclei are probably erythropoietic cells. H. & E. ×175.

[1]Becton, Dickinson & Co., Canada Ltd., Mississauga, Ontario.

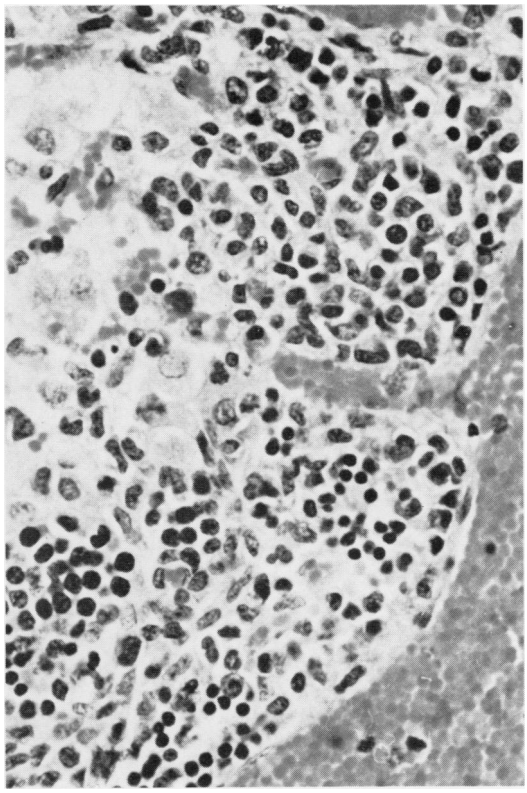

FIGURE 2. Higher magnification of the previous figure. H & E. ×450 (approximately).

In the spleen, there was marked activity of RE cells, particularly in follicular germinal centers where cell populations were almost exclusively of this type. The red pulp contained a monotonous population of RE cells, interspersed with some lymphocytes, neutrophils and occasional eosinophils. Cellular reactions were diffuse in the red pulp but tended to concentrate along the course of the penicilliary and sheathed arteries and in some cases, encroached upon arteries of the trabeculae.

Reaction in the lungs was restricted largely to perivascular areas and consisted mainly of accumulations of neutrophils as well as discrete aggregations of RE-like and hemopoietic cells.

In lymph nodes, reactions were very similar to those seen in the spleen with much RE cell activity in the germinal centers of lymphoid follicles. In some cases there was complete blurring of germinal centers by proliferating and infiltrating cells; the latter were chiefly neutrophils present in the sinuses.

Foci of RE and hemopoietic cells were observed at various levels of the renal cortex and medulla, with no apparent predilection for either site.

In some adrenal glands, granulomatous reactions, in addition to infiltrations of neutrophils and eosinophils, were observed in the zona glomerulosa, with limited extension into the zona fasciculata.

In the heart, infiltrations of mononuclear cells, chiefly plasmacytes and some neutrophils, were seen scattered throughout the myocardium.

Acute placentitis with necrosis and denuding of the chorion with the presence of numerous intracytoplasmic initial bodies and inclusions in swollen chorionic epithelial cells were features of the placental lesions. In Flocks 2, 3 and 4 organisms resembling chlamydiae were readily appreciated in placentas stained with azure eosin. Arterial thromboses were seen frequently in the chorioallantois. Intense neutrophilic perivasculitis was common in some placentas. Numerous macrophages, a few lymphocytes, scattered eosinophils and hemorrhages were seen in most placentas examined.

Bacteriology

Routine cultural examinations of various tissues and abomasal contents from Flocks 1 to 3, failed to demonstrate significant pathogenic bacteria. However, *Listeria monocytogenes* was isolated from two newborn lambs in Flock 4. Chlamydiae, identified serologically as members of the *C. psittaci* group, were recovered from Flock 4 only, in embryonated chicken eggs by two independent laboratories[2,3]. No cytopathogenic effects were observed after two passages in tissue cultures inoculated with tissue suspensions from Flock 1.

Serology

Serum samples from aborting ewes in Flocks 1 and 2 demonstrated positive titres, and in some instances, a twofold or greater increase in titre in the convalescent phase samples (Table I). Serological testing of Flock 3 several weeks following abortions did not demonstrate positive titres. Guinea pigs inoculated with fetal tissue and placental suspensions from Flock 1 were serologically negative when examined by the CF test at the end of 21 days.

DISCUSSION

In the past, ovine abortion epizootics in Alberta have been associated mainly with *Vibrio fetus*. Findings of this report therefore, add a new dimension to diseases causing abor-

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TABLE I

COMPLEMENT-FIXATION TITRES OF CHLAMYDIAE-INFECTED SHEEP

	Animal #	Acute	Convalescent
Flock #1	170	>1:160	>1:160
	172	1:80	1:80
	174	1:160	1:80
	315	>1:160	1:160
	316	1:160	1:160
	318	>1:160	1:160
	495	1:80	>1:160
	497	neg.	1:80
Flock #2	—	1:80	—
	9	1:160	>1:160
	16	1:160	>1:160
	23	1:160	1:80

tion in sheep in Alberta and in Canada, as ovine chlamydial abortion has not been reported previously as a flock problem in Canada. In 1961, Bannister *et al* (1) reported the isolation of chlamydiae from an aborted ovine fetus in Alberta, but because the aborting ewe was part of a flock which had been in contact with known chlamydiae-infected calves, evidence suggested that cross-infection had occurred.

The most striking and characteristic histological lesions in aborted lambs were foci of RE cells which were especially prominent in the liver but were observed also in other organs examined. Chlamydial initial bodies and inclusions in chorionic epithelial cells were best appreciated by the use of azure eosin stain. Gross and histological lesions in aborted lambs were similar to those described by Studdert and Kennedy (14); with minor variation, placental lesions were similar to those described by Stamp *et al* (11).

In chlamydial abortion of sheep, transmission of the organism to susceptible animals is believed to occur at the time of abortion or the birth of weak lambs (16). Further, it has been suggested by investigators in the U.K. that ewe lambs born into a chlamydiae-infected flock and subsequently removed from further exposure at weaning, may abort in their first pregnancy (5). Epizootiological investigations in North America however, were unable to confirm this observation (10, 15). Despite such differing opinions, the following summary appears to be valid (3):

1) Chlamydiae infect susceptible animals through the alimentary and respiratory tracts.
2) Transmission of chlamydiae occurs at abortion or parturition, with massive shedding of organisms in uterine discharges and in placenta.
3) Transmission of chlamydiae to ewes which are 30 to 120 days pregnant may result in placentitis, fetal injury and abortion, or the birth of weak lambs.
4) Transmission of chlamydiae to ewes in the last month of gestation does not usually result in abortion in the current pregnancy.
5) Transmission of chlamydiae to nonpregnant ewes, to ewes in the last month of gestation, and to lambs, may result in latent infections in these animals and subsequent abortion in the next pregnancy.
6) Recovered ewes are resistant to re-infection (3, 11).

Some or all of these observations appear to be important in the histories for Flocks 2 to 4, although information on specific sources of the infection in each flock is speculative. In retrospect, it is tempting to suggest that the purchase of three rams introduced the infection to Flock 1. Although it has been suggested that venereal transmission of chlamydiae may have occurred in a caprine abortion epizootic (4), a similar role for the ram has not been well explored (12). Experimentally, venereal transmission of chlamydiae in cattle was unsuccessful (7).

It is probable that some of the 40 ewe lambs introduced into Flock 3, and most of the eight ewe lambs introduced into Flock 4, were infected prior to purchase. Ewe lambs in the latter flock may have been exposed to aborting chlamydiae-infected ewes, either in their flock of origin in central Saskatchewan or at their intermediate location in central Alberta. However, fall lambing did not occur in resident ewes during this three week period, suggesting that exposure had occurred in the flock of origin. Further evidence suggested that the ewe lambs which aborted or produced weak lambs in Flocks 2 and 4 in 1972 and 1973, respectively, shed chlamydiae at that time and thus transmitted the infection to many susceptible, resident ewes. These ewes, in turn, aborted or produced weak lambs in the following lambing season. Approximately 50% of the numerous losses of lambs which occurred in Lots A and B of Flock 4 were estimated to have been associated with chlamydial infection. The remaining losses, aside from those associated with *L. monocytogenes*, were attributed to inadequate maternal care and to exposure to extremely cold climatic conditions.

Chlamydiae were isolated from aborted lambs in Flock 4 only; except for two isolations of *L. monocytogenes* from Flock 4, no other recognized bacterial pathogens were demonstrated in aborted or weak lambs from any flock.

Ewes in Flocks 1 and 2 demonstrated both high positive serological responses and in some cases, significant increases in titre in convalescent phase serum, when tested against a chlamydial group antigen (Table I). Titres of 1:16 or greater are considered to be significant (3). The negative serological findings in Flock 3 were similar to limited results obtained by McKercher *et al* (6) who, in studying experimental and natural ovine chlamydial abortions, observed that CF titres were inconsistent or had disappeared completely, particularly in sheep tested three or more months after exposure.

These serological observations, together with similar characteristic histopathological findings in aborted and weak lambs and in placentas from each flock, and the isolation of chlamydiae from aborted lambs in one flock, provided evidence which suggested that infection by chlamydiae had caused a significant percentage of the losses incurred by each flock.

The occurrence of chlamydial abortions in southern, central and northern Alberta suggested that this infection may be more widespread in sheep in Alberta, and perhaps in other provinces, than is presently recognized.

Summary

A diagnosis of chlamydial abortion (enzootic abortion of ewes, ovine viral abortion) was made in four flocks of sheep in Alberta in 1972 to 1974. Losses were not excessive except in one flock. The diagnosis was based on a composite of history, gross and histological lesions in fetal and placental tissues from all flocks, serology in two flocks, and on the isolation of chlamydiae from the tissues of aborted lambs in one flock. This is the first published report of ovine chlamydial abortion as a flock problem in Canada.

Résumé

Les auteurs ont posé un diagnostic d'avortement à Chlamydia (avortement enzootique des brebis, avortement viral ovin) dans quatre troupeaux de moutons de l'Alberta, de 1972 à 1974. Des pertes sérieuses ne se produisirent que dans un troupeau. Les critères du diagnostic comprenaient: l'anamnèse, les lésions macroscopiques et histologiques observées dans les avortons et les placentas reçus de tous les troupeaux, la sérologie de deux troupeaux, ainsi que l'isolement de Chlamydiae à partir des tissus d'avortons provenant de l'un des troupeaux. Il s'agit de la première publication sur l'avortement ovin à Chlamydia, comme problème de troupeau, au Canada.

Acknowledgments

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LETTER TO THE EDITOR

FUTURE OF THE VETERINARY PROFESSION IN CANADA

DEAR SIR:

The letter from Dr. Rouse, although not entirely accurate, has raised some valid points which have also been expressed in Council and Executive Committee discussions. The position of the CVMA is that we do not know with reasonable certainty and cannot at this stage make off-the-cuff decisions regarding the future of the profession. Like anybody else we have strong views but many of these views are impressions only, and cannot be factually supported.

It has become increasingly obvious to the CVMA Council that the profession is going to be faced with many major decisions in the next decade regarding its role and its contribution to society. Veterinary medicine has matured considerably over the past two decades and Council felt that what was required was an in-depth study of all facets of veterinary medicine. This should include amongst others a study of our manpower resources, our traditional and future roles, specialization, effective use of animal health technicians as well as proper utilization and incentives for regulatory veterinarians. Consideration must be given to possible changes in veterinary education to relate to the changing role of veterinary medicine in a changing society.

With this in view Council set up a committee of enquiry into the veterinary profession in July 1974, under the chairmanship of Dr. J. Archibald. It became patently obvious when this committee got off the ground that an enquiry into the profession by the profession alone was subject to severe criticism as to its credibility, and could be accused of producing a biased report based on self interest. Politicians with some justification would tend to dismiss the whole exercise. Approaches were made to the Federal Government for an independent enquiry into the veterinary profession. The response from government has been cool, but we have reason to believe it may be more attentive now, and this route is being actively explored again. Our own committee in the meantime is still active in gathering information for the CVMA brief. If government response is still negative to an independent enquiry then at least we will have background information from which to argue.

The Government in the United Kingdom has recently published the Swann Committee Report, the result of their own enquiry into the veterinary profession. Those who have had an opportunity to read this report must be impressed with its inestimable value to society and to government in making logical decisions. It is equally obvious that such an enquiry, undertaken by a professional society alone is simply not feasible.

When we contemplate the massive expenditure of government funds required to enlarge schools and build new ones, it is, in our opinion, not unreasonable to ensure that the decisions are based on supportable current evidence. As an example, the reasons for recruitment problems into the regulatory field and the widespread professional dissatisfaction within that field should be fully aired and objectively examined. Perhaps then there will be some reasonable hope that enduring solutions will be forthcoming.

The CVMA does not encourage or oppose the expansion of Canadian veterinary education, but we would be less than responsible if we endorsed proposals without a thorough examination of all the relevant data, which no one to this time has been able to produce.

R. G. STEVENSON
President
Canadian Veterinary Medical Association