THE CANADIAN VETERINARY JOURNAL

LA REVUE VÉTÉRINAIRE CANADIENNE

Volume 20 **April-avril 1979** **No. 4**

Bovine Granular Vulvitis Associated with Ureaplasma Infection

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SUMMARY

A granular vulvitis syndrome associated with ureaplasma infection was first recognized in Ontario dairy herds in 1972.

The acute form of the disease was characterized by a purulent vulvar discharge, an inflamed hyperemic vulvar mucosa and varying degrees of granularity. In the chronic form, there was an absence of a purulent discharge and a gradual decline in the severity of the hyperemia and granularity. Epithelial inclusion cysts were observed in the vulvar epithelium of approximately 10% of affected cows.

A seasonal variation in the incidence of the disease was observed. Herd morbidities during the summer months reached a low of 37% and increased to 75% during the winter months with constant housing.

When widespread in herds, the acute form of the disease had a significant effect on fertility. In four herds examined, first service conceptions dropped on average by 27%.

The chronic form of the disease had a less detrimental effect on fertility with first service conceptions being reduced on average by 13%.

Intrauterine infusions of a tetracycline 24 hours postbreeding were found to be of value in improving conception rates in acutely affected herds.

RÉSUMÉ

Vulvite granuleuse bovine attribuable à une infection par ureaplasma

En 1972, les auteurs diagnostiquèrent, pour la première fois en Ontario, un syndrome de vulvite granuleuse qui sévissait dans des troupeaux de bovins laitiers.

La forme aiguë de la maladie se caractérisait par un écoulement vulvaire purulent, ainsi que par une inflammation hyperémique et un aspect granuleux plus ou moins marqué de la muqueuse vulvaire. Environ 10% des vaches malades présentaient des kystes d'inclusion, au sein de l'épithélium de la vulve.

La maladie manifesta une incidence saisonnière. La morbidité se situait aux environs de 37%, en été: elle atteignit toutefois 75%, au cours de la stabulation hivernale.

Lorsque la forme aiguë de la maladie atteignit des proportions élevées au sein d'un troupeau, elle en affecta la fertilité. C'est ainsi que dans quatre troupeaux atteints, le taux de conception consécutif à une première saillie diminua de 27%. Cette baisse s'avéra moins drastique et ne fut que de 13%, lors de la forme chronique de la maladie.

L'infusion intra-utérine d'une tétracycline, 24 heures après la saillie, se révéla efficace en améliorant le taux de conception chez les vaches des troupeaux aux prises avec la forme aiguë de la maladie.

INTRODUCTION AND LITERATURE REVIEW

Granular vulvitis (granular venereal disease, nodular venereal disease) was first described in Switzerland by Isseponi in 1887 (cited by Williams) (15). Since that time there has been controversy as to the etiology, clinical severity and possible effects on fertility (1, 3, 5, 10, 15).

Numerous etiologies, both infectious and noninfectious, were proposed prior to 1958. Bacteria, most notably Streptococci, were regarded by many as the most probable cause (5). Others however, noted very early that the disease "did not behave like an ordinary bacterial infection" (3, 16). Afshar (1) was the first to note an association between *Mycoplasma (M. bovigenitalium)* infection and granular vulvitis. The disease was reproduced following the application of the organism to lightly scraped vulvovaginal epithelium. However, the isolation of the organism from naturally affected cows was infrequent and made any cause-effect incrimination doubtful.

Ruhnke *et al* (11) also isolated *M. bovi-*

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genitalium infrequently from natural cases of granular vulvitis. There was a much stronger association with ureaplasma infection and the natural disease. These findings tended to rule out *M. bovigenitalium* as a primary cause of the disease and formed the first direct link between ureaplasma infection and bovine genital disease.

Ureaplasmas had been found previously to be common inhabitants of the normal bovine vagina (7, 9, 12) and were not associated with disease (7, 9). However, the fact that both virulent and avirulent strains of bovine ureaplasma have been demonstrated (4), may account for the variable findings (11).

The purpose of this paper is to describe a newly recognized granular vulvitis syndrome of Ontario dairy cattle which has been shown to be associated with ureaplasma infection (11).

MATERIALS AND METHODS

Cows from 16 dairy herds in southern Ontario were studied during 1976. The majority of the cows were of the Holstein-Friesian breed. In four of the herds, reproductive records were available for the period 1972-76 and were used to monitor the effect of the disease on fertility as well as response to treatment.

The techniques used in the microbiological studies have been reported (11).

RESULTS

Clinical Findings

The clinical syndrome was found to have both acute and chronic forms, easily distinguished by the presence or absence of a purulent vulvar discharge (Table I).

The acute form appeared initially in individual cows three to six days postbreeding. Other cows in the herd soon became infected with direct contact appearing to facilitate transmission. In two cases the farm dog was suspected of spreading the disease by licking vulvas. The main presenting sign was the sudden onset of a sticky purulent vulvar discharge. The volume of the discharge varied from a small amount observed on the tail or vulvar hairs to a larger volume a few days later which appeared to pool in the vagina before emptying behind the recumbent cow in 60 to 100 ml amounts.

The vulvar epithelium during the acute stage was inflamed, sensitive and hyperemic. Small 1 to 2 mm raised granules were evident, usually most prominent around the clitoris. Purulent material was often observed in the ventral commissure. In severe cases the granularity extended dorsally along the lateral walls of the vulva and occasionally involved the dorsal commissure. Coalescence of the granules produced raised ridges resulting in a corrugated and pebbled vulvar mucosa. The granularity did not appear to extend cranially to involve the vaginal epithelium.

The vulvar discharge persisted for three to ten days before the disease progressed to a chronic form (Figure 1). In many cases the acute form would reappear at subsequent heats.

TABLE I
CRITERIA FOR DIVIDING BOVINE GRANULAR VULVITIS INTO ACUTE AND CHRONIC FORMS

Signs	Acute	Chronic
Hyperemia	+	+ or -
Granularity	+	+
Purulent Discharge	+	-
Epithelial Cysts	+ or -	+ or -

FIGURE 1. Chronic granular vulvitis showing the degree of granularity commonly observed immediately following the acute form of the disease.

The chronic form was characterized by an absence of a purulent discharge and a gradual decline in the severity of both the hyperemia and granularity. Occasionally an excessive discharge of clear mucus was observed which made heat detection difficult for many owners. The granularity gradually disappeared over the next few weeks and the vulvar epithelium returned to normal within six weeks to three months. Reinfection however was common. The disease became endemic within many herds and numerous reinfections were observed over a four year period. The clinical signs were generally less severe during reinfection, with the acute stage being short or apparently absent.

A characteristic finding observed in approximately 10% of affected animals was the presence of

TABLE II

RESULTS OF A MICROBIOLOGICAL STUDY OF 81 COWS FROM 16 DAIRY HERDS AFFECTED WITH GRANULAR VULVITIS (11)

Classification	Number Cultured[a]	Percent Positive						
		Ureaplasma[b]	M. *bovigenitalium*	Strep. (non-faecal)	Strep. (faecal)	*E. coli*	*C. pyogenes*	*Hemophilus*[c] *somnus*
No vulvitis	34	23.5	0	55.8	23.5	23.5	0	0
Mild moderate vulvitis	27	74	7.7	51.9	11.5	40.7	7.7	0
Acute vulvitis	20	100	20	45	5	20	15	25

[a]Results are from a single sampling from each cow

[b]Six cows with vulvitis had ureaplasma but no bacterial growth

[c]Other bacteria isolated were *Erwinia* — 2, *Geotrichum* — 1, Beta-hemolytic streptococcus — 1, *P. aeruginosa* — 1, *P. multocida* — 1, *Mima polymorpha* — 1, *Klebsiella* — 1. No viruses were isolated.

discrete raised white nodules 2 to 5 mm in diameter and usually arranged in rows or clustered on the dorsolateral wall of the vulva or in the dorsal commissure (Figure 2). The nodules did not appear to form around the clitoris but were observed on two occasions in small numbers on the lateral vaginal wall and outer cervical ring. A creamy white exudate could be expressed from the nodules during the acute stage. The contents tended to become inspissated later in the chronic form.

Disease Incidence

The clinical incidence of the disease in 16 herds was monitored closely for a seven month period (Table III). A definite seasonal variation was observed. The disease incidence varied from a low of 37% in midsummer to a high of 75% in midwinter. The averages for the two seasons were 44.6% and 61.1% respectively.

FIGURE 2. Epithelial inclusion cysts clustered on the lateral vulvar wall of a cow recovering from acute granular vulvitis.

TABLE III

SEASONAL INCIDENCE OF BOVINE GRANULAR VULVITIS (BASED ON 2784 OBSERVATIONS)

Season	In All Cows Examined	In Repeat Breeder Cows
June — August (Summer)	44.6%	69.1%
September — December (Winter Stabling)	61.1%	75.0%
Average	54.5%	69.8%

In 353 cows examined because of a failure to conceive after one or more breedings, the average seasonal incidence was slightly higher: 69% during the summer and 75% during the winter.

Effect on Fertility

The reproductive records of four herds for the four year period 1972-76 were examined (Table IV). With the exception of one farm where natural service was temporarily used, all herds utilized artificial insemination.

The acute form of the disease had a severe effect on fertility. In one 35 cow herd, first service conception rates dropped by 48% (from 69% to 21%) with five cows being subsequently culled due to persistent infertility. A second 300 cow herd experienced a 30% drop (64% to 34%) in the first service conception rate over a two month period immediately following the appearance of the disease. On the average in all herds, first service conceptions during the acute form were reduced by 27%.

In one 30 cow herd the first service conception rate during the winter months was 25%. During the

TABLE IV

CONCEPTION RATES ASSOCIATED WITH BOVINE GRANULAR VULVITIS

Herd Status	Number of Records Examined	1st Service Conceptions	+	2nd Service Conceptions	Services Per Conception
Before disease appeared	494	55.9%		74.9%	1.83
During acute form	230	28.8%		51.4%	2.71
During intensive treatment	51	51.7%		73.7%	1.81
During chronic form with sporadic treatment	343	42.2%		69.4%	2.16

following summer months the owner introduced natural service with a young bull and first service conceptions improved to 65%. By fall, conception rates were again declining and natural service was discontinued. First service conceptions over the next eight months again plummetted to 17%.

There was no apparent increase in the incidence of abortions in the herds studied over the four year period.

Response to Treatment

Many conventional douches, suppositories and uterine infusions were used initially in an attempt to control the disease and improve conception rates.

Products contining nitrofurazone, chloramphenicol, chlorhexidine, 0.2% Lugol's iodine or acriflavine solutions were judged to be of no value. When it was determined that ureaplasmas were associated with the disease, it was found that intrauterine infusions with 1 g tetracycline suspension[1], 24 hours postbreeding resulted in a significant improvement in conception rates.

In herds where conceptions were greatly reduced, all affected cows were treated 24 hours after artificial insemination. Conception rates with this treatment returned to the level observed prior to the appearance of the disease (Table IV). Later with the chronic form of the disease, treatment was more sporadic and limited to only repeat breeders and cows with a purulent discharge. Conception rates with this form of therapy remained 10-12% lower than the level observed prior to the appearance of the disease.

Histopathological Findings

The granular lesions were due to diffuse and focal accumulations of lymphoid cells within and beneath the squamous epithelium. There were germinal centres in many of the focal lesions.

The nodular lesions were epithelial inclusion cysts similar to those commonly found in the skin of dogs (Figure 3). As in dogs, they were lined by squamous epithelium and some of the largest cysts contained keratin squames. Occasionally, there was mucous metaplasia of the squamous epithelium.

FIGURE 3. Epithelial inclusion cysts associated with vulvar epithelium.

The cysts seemed to develop in two ways. Either an isolated subepithelial nodule continued to grow and became cystic (Figure 4), or the opening to an invagination of surface epithelium was blocked by detritus (Figure 5) and then closed by overgrowth of that surface epithelium. The isolated subepithelial nodules were commonly found in areas showing diffuse lymphocyte infiltration and marked epithelial proliferation.

Microbiological Findings

The results of a microbiological survey of affected cows have been reported (11) (Table II).

Ureaplasmas were isolated from 100% of acute cases, 74% of chronic cases and 23.5% of apparently normal herd mates. *Mycoplasma bovigenitalium* and *Hemophilus somnus* were also isolated in lower numbers from more diseased cows than from normal herd mates.

[1]Reverin Suspension, Hoechst Pharmaceuticals, Montreal, Quebec.

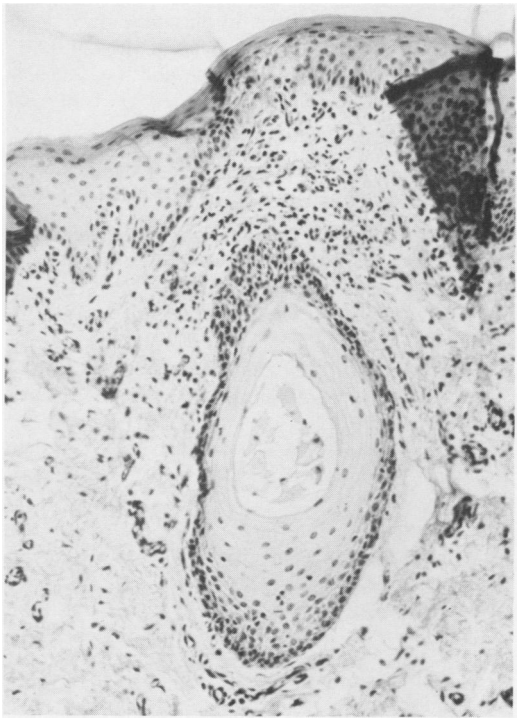

FIGURE 4. Subepithelial nodule showing early cyst formation with accumulation of keratin.

FIGURE 5. Invagination of inflamed epithelium showing accumulation of detritus. Note mucous lining of adjacent cysts.

No bacteria, mycoplasma or chlamydia were isolated from the contents of four epithelial cysts.

In one herd, granular vulvitis appeared in cows one to two weeks after recovery from an outbreak of infectious pustular vulvovaginitis (I.P.V.). Ureaplasmas were isolated from six of six cows with acute vulvitis and *M. bovigenitalium* from four of six.

DISCUSSION

The incidence and clinical signs observed in these outbreaks appear similar to previous reports (1, 2, 3, 5, 15). The only notable exception was the formation of intraepithelial inclusion cysts in approximately 10% of affected cattle.

The cysts have been described previously in two herd outbreaks of "concomitant granular vulvitis, palate lesions and respiratory illness in Connecticut dairy cattle" (14). The etiology of the condition was not determined but the outbreaks were observed prior to the recognition of bovine ureaplasma infections. What significance or part the cysts have in the disease has not been determined. It seems likely that they are formed as a result of proliferation of the inflamed epithelium.

The influence of granular vulvitis on fertility has been as controversial as the proposed etiologies. The disease is believed by some to have no effect (8, 10) while others have reported a significant lowering of fertility (2, 3, 15). The apparent variability may be due in part to a failure to adequately correlate severity and duration of infection with the resulting infertility. From our findings, the differentiation of the disease into both acute and chronic forms is very important for the ease of isolating ureaplasma and also for determining the subsequent effect on fertility. There was little doubt that the acute form of the disease had a severe effect on conceptions while the chronic form was much less detrimental. It would also appear that there may be some differences with natural service versus artificial insemination, as the use of a bull in one herd resulted in a significant although temporary improvement in conception rates.

In the only previous study of the influence of the disease on fertility following artificial insemination, it was found that severely affected cows had a 10% reduction in 60 day nonreturns, and mildly affected 3% when compared to controls (13). Severe cases were reported to have many nodules plus inflammation. It is not clear whether a purulent discharge was present in all or any severe cases. Our findings of much lower conception rates with acute cases may be due to the fact that our "acute" classification was much more restrictive. Many of our chronic cases also had numerous nodules and inflammation (hyperemia) and the resulting 13.5% reduction in conceptions compares closely to the previous findings.

Granular vulvitis has been observed to occur following I.P.V. infection (6) and a similar

observation was made in one herd in this study. However, all cows cultured in this herd were also positive for ureaplasma infection. As well viruses were not recovered from any of the cases of granular vulvitis studied in herds free of clinical I.P.V. (11). These findings would indicate that although I.P.V. may act as a predisposing lesion, the virus should not be incriminated as a primary etiological agent in granular vulvitis.

Numerous treatment procedures have been advocated to control the disease. The most common type of therapy has been in the form of various douches usually containing either irritant drugs or antibiotics. Autogenous bacterins have also been reported to be of value (3, 5). The tremendous variability in past treatments has prompted some to question the value of any form of therapy (10).

In affected herds in this study, utilizing artificial insemination, the treatment procedure was directed toward reducing the effect of possible mechanical transmission of infection from the vulva to the uterus. Uterine infusions with a tetracycline, 24 hours after service appeared to be effective in improving conception rates. Further studies are needed to determine the best method of eliminating the vulvar infection. This would facilitate control of the infection in dairy herds and also would avoid transmission as would be expected to occur with natural service.

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CANADIAN VETERINARY MEDICAL ASSOCIATION

1979 Annual Meeting

Notice of Meeting

In conformity with by-law 33 of the Canadian Veterinary Medical Association notice is hereby given that the 1979 Annual Meeting of the Association will be held on Saturday, July 7 in Toronto, Ontario at the Harbour Castle Hilton. The meeting will convene at 3:30 p.m.

D.A. LANDRY, D.M.V.
Secretary Treasurer

Ottawa, Ontario, April 1, 1979

ASSOCIATION CANADIENNE DES VÉTÉRINAIRES

Assemblée annuelle 1979

Avis de convocation

Conformément au règlement 33 de l'Association canadienne des vétérinaires, avis est par la présente donné que l'Assemblée annuelle de l'Association en l'an 1979 sera tenue samedi le 7 juillet à Toronto, Ontario à l'hôtel Harbour Castle Hilton. La réunion débutera à 3:30 p.m.

D.A. LANDRY, D.M.V.
Secrétaire trésorier

Ottawa, Ontario, le 1 avril 1979