Farm Practice

Clinical aspects of the bovine virus diarrhoea / mucosal disease complex in cattle

by Joe Brownlie

CATTLE infected with bovine virus diarrhoea (BVD) virus can present a variety of clinical signs, both enteric and respiratory. It was this variety that led originally to the description of two separate diseases, bovine virus diarrhoea and mucosal disease of cattle. Both diseases are now thought to be caused by the same virus.

Furthermore, as the virus is widespread throughout the national herd there is every opportunity for infection. Most adult cattle have antibodies to BVD virus by the age of two years. However, there now appears to be a significant number of 'closed' herds with no detectable antibody to the virus; these represent particularly susceptible groups of cattle.

Bovine virus diarrhoea virus

BVD virus is an RNA virus which, with border disease virus and hog cholera virus, forms a group which are classified as pestiviruses in the Togaviridae. The three viruses cross-react antigenically and have other biological and structural similarities.

Although isolates of BVD virus from different clinical cases of disease may be serologically distinguishable, the most obvious difference is their separation into one of two forms by

their growth characteristics in cell culture: they may be non-cytopathic (Fig 1) or cytopathic (Fig 2). This distinction has only recently been shown to be important in the aetiology of mucosal disease.

Aetiology of mucosal disease

An explanation for the aetiology of mucosal disease has recently been put forward which proposes that mucosal disease develops following an early in utero infection with non-cytopathic BVD virus resulting in the birth of a calf with a persistent viraemia. If this calf is later superinfected with a cytopathic BVD virus, mucosal disease can develop (Fig 3).

It is only those animals that are persistently infected with non-cytopathic BVD virus which will later develop mucosal disease.

Aspects of infection with BVD virus

Acute infection

BVD disease refers to the acute infection of seronegative cattle with BVD virus. The original description of explosive
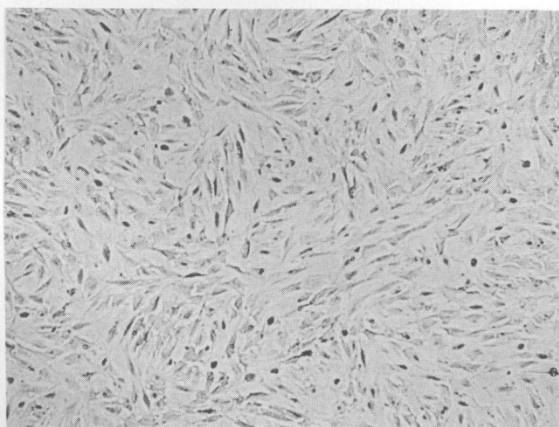

Fig 1: Monolayer of calf testis cells infected with non-cytopathic BVD virus (courtesy of M. C. Clarke, IRAD, Compton)
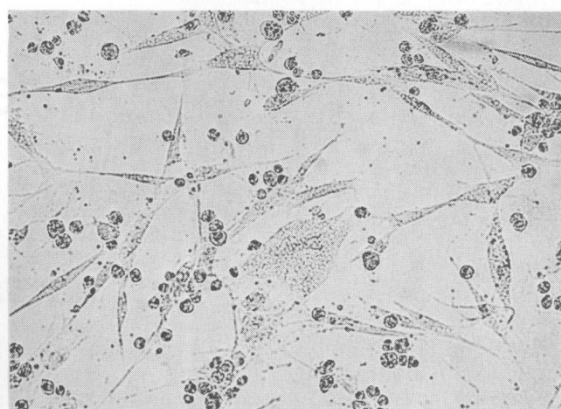

Fig 2: Monolayer of calf testis cells infected with cytopathic BVD virus. Note rounded and vacuolated cells with cell depletion (courtesy of M. C. Clarke, IRAD, Compton)

COW

CALF

Seronegative cow

in early pregnancy,
120 days

BVDV(nc)

Infected with BVD virus
(non-cytopathic)

subclinical disease,
transient viraemia

Viraemic

Antibody response

Fetus not
immunocompetent;
no antibody

Parturition

colostral antibody

Persistently
viraemic
at birth and
for life

Colostral
antibodies –
short duration

6–24 months

Superinfection with
BVD virus (cytopathic)

BVDV(c)

Mucosal
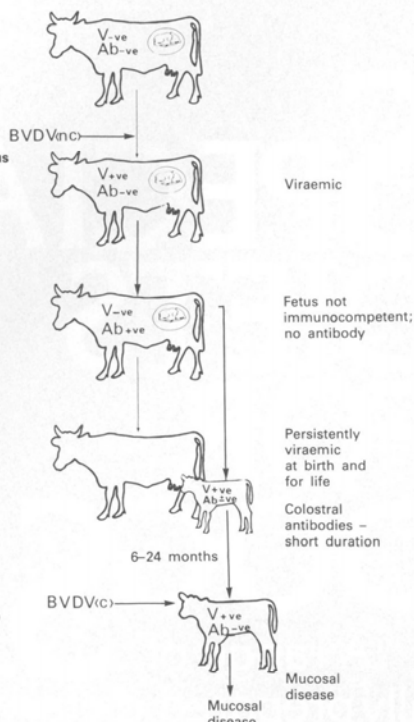disease

Mucosal
disease

Fig 3: Hypothesis for the aetiology of mucosal disease. V – BVD virus; Ab – antibody to BVD virus; nc – non-cytopathic; c – cytopathic

outbreaks of diarrhoea caused by the virus is now rarely observed and most infections may be subclinical. Experimental infection of calves produces a similarly mild disease with a transient leucopenia and some animals may show a rise in temperature. In both natural and experimental infections antibodies are produced in two to three weeks and the animal appears immune to later challenge.

In utero infection

Crucial to the development of mucosal disease is the in utero infection of the fetus with non-cytopathic BVD virus. There is no evidence from either field or experimental work that persistent viraemia occurs with cytopathic BVD virus.

Infection with non-cytopathic BVD virus must be before 120 days of gestation for the virus to persist. The immune system is not functionally developed before that stage so the virus can become widely established in the fetal tissue. Subsequently although the immune system is competent it recognises the virus as 'self' and a state of immune tolerance develops. This tolerance allows the non-cytopathic virus to persist into neonatal life and also the unchecked replication of superinfecting cytopathic BVD virus.

Abortions and stillbirths

A feature of BVD virus infection in pregnant cattle is the frequency of abortions and stillbirths. Retrospective inquiries following a mucosal disease outbreak will often reveal a series of abortions and births of dead or weak calves. Sometimes there may have been an infertility problem with increased return to service caused by early embryonic loss.

Congenital abnormalities

Fetopathology caused by BVD virus infection has been well documented. Typical is the intrauterine growth retardation observed in various organs, eg, thymus, and also the pathology caused to the central nervous system. Clinically, affected calves are born weak, ataxic and often with visual disorders. They may have antibody to BVD virus and no virus.

Mixed infections with BVD virus and other pathogens

BVD virus is considered to be an immunosuppressive agent which infects the cells of the immune system, resulting in leucopenia. There is a reduction in the defence mechanisms and as a result the host is more susceptible to infection with other pathogens. Such mixed infections have been reported from both field and experimental work and may represent an important sequel of acute BVD virus infection.

For example, a combined experimental infection with BVD virus and *Pasteurella haemolytica* results in a severe fibrino-purulent bronchopneumonia with the area of pneumonic lesions increasing to 40 to 75 per cent compared to 5 to 15 per cent with pasteurella alone. This synergism has also been observed with parainfluenza, infectious bovine rhinotracheitis and respiratory syncytial viruses.

Mucosal disease

Clinical signs

Mucosal disease usually occurs in six- to 24-month-old cattle and is invariably fatal. The cattle possess no specific antibodies to the infecting virus even though it has persisted in the blood during their lifetime.

The first clinical sign is usually anorexia. Close inspection may reveal erosions either on the oral mucosa particularly at the gingival margin (Fig 4), on the tongue (Fig 5), the external nares and in the buccal and nasal cavities. These lesions are present in about 75 per cent of cases. In some animals, there is desquamation of the muzzle with extensive crusting and even purulent exudate. There may also be nasal discharge (Fig 4).

Lesions can be seen around the coronet and on the interdigital surface, often with redness and swelling. The animal is disinclined to walk and soon becomes recumbent. There is often profuse diarrhoea and invariably death. Death can be so sudden that it may be the first clinical sign, but normally it follows three to 10 days from the onset of symptoms.

Post mortem findings

Much useful information can be gained from a detailed post mortem examination of suspected cases of mucosal disease. The full extent of oral, lingual and buccal erosions should be observed. A common finding in the buccal mucosa is that the small erosions have coalesced into larger areas of necrosis and sloughed epithelium.

Similar erosions may be seen in the oesophagus. Ruminal lesions, if present, are areas of congestion and oedema along the ruminal pillars. Ulceration is rare. The large ruminal papillae can be reduced in size.

The abomasum and small intestine provide the most reliable sites for inspection but immediate autopsy is important as post mortem changes in the gut are rapid and gaseous extension can often mask the enteric erosions.

The abomasum usually shows several (five to 50) small discoid erosions, about 5 mm diameter, with surrounding hyperaemia in the mucosa. Submucosal petechial haemor-

Fig 4: Muzzle of calf with mucosal disease. Note the desquamation and crusting. At the gingival margin there is hyperaemia and mucosal erosions

Fig 5: Tongue of a calf with mucosal disease showing complete loss of epithelium on the apex

rhages are a common finding, particularly in the pylorus (Fig 6). Occasionally, the erosions can be larger and ulcerated.

The small intestine, if opened throughout its length to expose the antimesenteric surface, will reveal oval erosions (2 cm to 5 cm long) that overlie the lymphatic tissue in Peyer's patches. The erosions may vary from two to three to 30 to 40. Towards the terminal ileum the erosions can become extensive and may be up to 10 to 20 cm in length. The exposed submucosal surface of the erosions can vary from the chronic lesion, with food adhering (Fig 7), to the acutely congested one often with interluminal haemorrhage.

In the large bowel, there may be congestion of the mucosa which gives a thickening to the mucosal folds and a striped appearance. There may also occasionally be petechial haemorrhages and small erosions along the folds. The contents are dark, watery and often foul smelling.

Field outbreaks

A number of mucosal disease outbreaks have been extensively investigated and several salient features have become recognised.

(1) The initial investigation is usually prompted following the rapid deterioration or perhaps death of an animal in the six to 24-month-old age group.

(2) The clinical investigation, together with post mortem findings, guides the practitioner to consider a diagnosis of mucosal disease.

(3) Examination of blood samples from the remaining animals in the group reveals a number that have little or no antibody to BVD virus and some that are also viraemic.

It is therefore essential to examine all in-contact animals for the presence of both virus and antibody. There are four possible categories for these animals (Fig 8).

Within these four combinations, it is only animals in category 4b and occasional animals in category 3b that will subsequently develop mucosal disease. The early identification of these animals is all important.

Sequential study of an outbreak

Most of the larger outbreaks of mucosal disease that have been studied have shown that, at some time, new animals have been introduced into a 'closed' unit or herd of susceptible cattle, often about 18 months previously.

Fig 6: Post mortem appearance of the abomasum with erosions surrounded by congestion and petechial haemorrhage

Fig 7: Mucosal disease. Post mortem appearance of the small intestine. Some erosions may appear chronic, even with food adhering to the surface

The newly introduced animal can bring in the virus either as an acute infection or as a persistent one. The virus is then transmitted by animal-to-animal contact. If seronegative heifers in early pregnancy are present, there is a basis for in utero infection, with the subsequent production of persistently viraemic calves and later an outbreak of mucosal disease. Obviously should the introduced animal be persistently
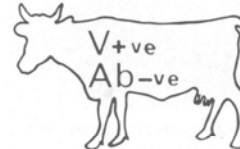
Category	Animal	Status	Result of exposure to BVD virus	Final antibody status
1	V - ve Ab - ve	no previous exposure to BVD virus	transient mild infection	+ve
2	V - ve Ab + ve	previous exposure to BVD virus from 120 days gestation onwards	immune	+ve
3	V + ve Ab + ve	(a) acute viraemia presently sero-converting	will become immune	+ve
		(b) persistently viraemic; occasionally these animals may have low levels of antibody	may later succumb to mucosal disease	±ve
4	V + ve Ab - ve	(a) acute viraemia	will become immune	+ve
		(b) persistently infected with BVD virus	may later succumb to mucosal disease	-ve

Fig 8: Combinations of BVD virus and antibody in cattle and their significance

infected, there is far greater chance for transmission of virus to susceptible cattle. At present there is little evidence for BVD virus becoming latent following acute infection and at a later date being able to recrudesce.

Once a case of mucosal disease has been diagnosed, the presence of cytopathic virus can be assumed. The transmission of cytopathic BVD virus is rapid and, once super-infected, persistently infected cattle rapidly lose condition and invariably die.

An analysis of all the effects that can follow from BVD virus infection reveals the extent and true cost of an outbreak (Fig 9).

By the time the practitioner is called, the earlier damage caused by virus infection has already occurred, but has not been associated with BVD virus.

Control measures

To prevent an outbreak

In the UK there is no effective vaccine and caution will be needed before live virus vaccines are introduced. Reports from Europe and the USA have shown that live vaccines which originate from cytopathic BVD virus can precipitate mucosal disease.

The best means of control at present is to screen newly introduced stock for the presence of virus and antibody. This is particularly important for new animals that will be in contact with pregnant cattle, ie, a new bull or young heifer.

During an outbreak

The remaining animals in the susceptible group should be bled and examined for BVD virus and antibody. Any animal found to be persistently viraemic should be either isolated or,
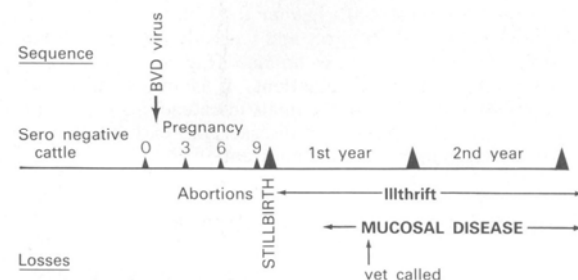
Fig 9: Possible sequence of events following introduction of BVD virus into a susceptible group of cattle
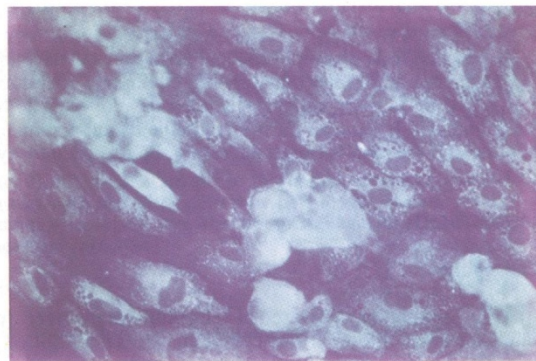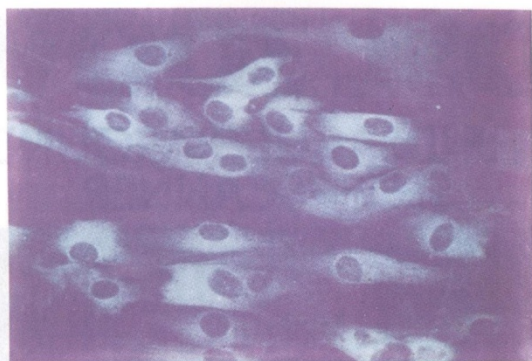
Fig 10: Cell culture monolayers infected with (left) non-cytopathic and (right) cytopathic BVD virus, stained with specific BVD virus fluorescein-coupled antibody

as experience now indicates, sent for immediate slaughter.

Isolation to prevent contact with cytopathic virus can occasionally work, but by the time the laboratory diagnosis can be made, there has been general transmission of cytopathic virus throughout the remaining cattle. Cattle can quickly lose condition following superinfection and there is little remaining carcase value.

Differential diagnosis

Bovine virus diarrhoea

A primary BVD virus infection is usually mild and rarely diagnosed except by serology.

Mucosal disease

There are three major features of mucosal disease that may be present: mucosal erosions, diarrhoea and death. The differential diagnosis should be as described below.

Foot-and-mouth disease

All cloven-hooved animals are susceptible to foot-and-mouth disease which is characterised by pyrexia, anorexia and excessive salivation. Tongue and buccal erosions are consistently present and are preceded by the formation and rupture of vesicles containing straw-coloured fluid.

This contrasts with mucosal disease where the erosions arise not from vesicles but directly from necrosis of erythematous areas in the mucosa.

With foot-and-mouth disease the morbidity can be 100 per cent whereas mortality is less than 5 per cent in adults, although it may rise to 50 per cent in younger animals. With mucosal disease the morbidity of a herd is lower and generally restricted to the six to 24-month age groups, but mortality may approach 100 per cent.

Malignant catarrhal fever

Malignant catarrhal fever is another disease characterised by gastroenteritis and an erosive stomatitis. There is usually bilateral corneal opacity and general lymph node enlargement. The serosa of the abomasum and large intestine is hyperaemic, oedematous and often with haemorrhage into the rumen.

The small intestine shows less damage and the discrete ulceration of Peyer's patches, seen in mucosal disease, is not present. Sporadic cases of malignant catarrhal fever are seen

in the UK and diagnosis depends on histological examination of tissue and cerebrospinal fluid.

Salmonellosis

Salmonellosis is often considered in the differential diagnosis. Outbreaks of acute diarrhoea with some deaths are features common to salmonellosis and mucosal disease. There are rarely oral or intestinal erosions with salmonellosis and often younger calves are more severely affected.

There is frequently a marked spleen, liver and lymph node enlargement. Diagnosis depends on isolation of bacteria from faeces or post mortem tissue samples.

Rinderpest

Rinderpest is another disease where vesicles precede erosions on the tongue, oral mucosa, teats and coronet. There is more severe lymph node and intestinal oedema than in mucosal disease but similar Peyer's patch erosions are seen. The morbidity and mortality are both high.

Ibaraki disease

This disease, not yet reported in the UK, is caused by an orbivirus and has been classified as epizootic haemorrhagic disease virus, type V. It presents an erosive stomatitis, abomasitis and often diarrhoea.

There is damage to the oesophageal musculature often resulting in megaoesophagus with the retention of imbibed fluids. These are regurgitated as copious clear mucus fluid through the mouth and sometimes nose. This 'deglutative disorder' is particularly characteristic of Ibaraki disease and not seen in mucosal disease. The erosions of Peyer's patches are rarely presented. It is dependent on insect transmission and therefore seasonal.

Laboratory techniques for diagnosis of BVD virus

Detection of antibody

Serum neutralisation

Serum neutralisation depends on the ability of antibodies in the serum to neutralise BVD virus and thereby prevent infection of cell culture. The test usually takes four to seven days to obtain a result but is dependent on cell culture facilities and an experienced observer.

ELISA

An enzyme-linked immunosorbent assay (ELISA) technique for BVD virus antibodies that depends on binding of antibody to specific BVD virus antigen has been developed. The test takes one day. It requires purified ingredients but is simple to operate and the results can, if necessary, be recorded by eye.

Detection of virus

Cell culture

BVD virus can be cultivated in cell culture monolayers (eg, calf testis or calf kidney). The cytopathic virus is identified by changes in the monolayers such as vacuolation of cell cytoplasm, rounding of cells and their subsequent lysis. Non-cytopathic virus produces no such changes. Both viruses can be visualised by fluorescein-coupled antibody (Fig 10). Primary identification of these viruses may be made in about seven days.

Enzyme staining

Virus grown on cell culture monolayers in microtitre assay plates or small petri dishes can be identified by enzyme-linked antibody. This assay may take only three to four days.

Deer and sheep

Deer and sheep can be infected by BVD virus. Therefore possible transmission to or from both species should be considered during any outbreak of mucosal disease of cattle.

Conclusion

Our understanding of this disease, first described in 1946, has advanced recently and more may be revealed in the next five years, so this report should be regarded only as an up-to-date reference. New technology has improved diagnosis of the disease eg, ELISA techniques, molecular virology and detailed immunology. There is greater confidence in predicting the course of a disease and in ascribing a correct viral and antibody status to an animal.

It may be suggested that a test for persistent viraemia should be considered in any health inspection of cattle by the practitioner. What is becoming clear is that outbreaks of clinical disease can be the cause of severe loss and that the cattle practitioner needs to be aware of all its aspects.

Further reading

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