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In this issue :

ANIMAL HEALTH UPDATES

Animal Health Group

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Disease - Bovine Viral Diarrhoea and its Control in farms

Bovine viral diarrhoea is a viral disease of cattle and other ruminants.

Major financial losses occur due to poor reproductive performance and the birth of calves with persistent virus infection.

While elimination of infection from the herd is optimal, vaccination is highly effective and produces substantial cost benefits.

Etiology

BVD is caused by the bovine viral diarrhoea virus (BVDV) within virus family Flaviviridae. In India both BVDV -1 and BVDV -2 occur. BVDV -1 occurs predominantly whereas BVDV-2 occurs sporadically. BVDV-1 has been identified in cattle, buffaloes, sheep, goats and yaks while BVDV- 2 has been detected in cattle, sheep and goats. BVDV-1b strains are predominantly prevalent in cattle. BVDV-1c has been identified in Himalayan yaks.

There are two bio-types designated as non-cytopathic (NCP) and cytopathic (CP) depending on their effect on tissue culture cells. The NCP type is common and most important as they can cross the placenta, invades the foetus and establishes persistent infection in the foetus. The CP biotype is usually associated with only mucosal disease in animals already persistently infected with the non-cytopathic biotype. There is evidence that the CP biotype appears by mutation from the NCP biotype within persistently Infected (PI) animals.

Epidemiology

BVD have been recorded in most countries where cattle are raised. The prevalence of the infection is high but incidence of clinical mucosal disease is low. Mucosal disease cases have not yet recorded in India.

Following natural infection of sero-negative immuno competent cattle with most of the strains of BVDV which do not cause severe disease, there is a transient viremia, serum neutralizing antibodies develop within 2-3 weeks, peak at 8-10 weeks, and remain detectable for many months.

Prevalence of infection

Young cattle which are persistently infected with a NCP biotype are major source of infection in a herd. The mean prevalence of PI animals in herds is about 1-2 %.

There is serological evidence of BVDV infection within a range of 14-38% in cattle and 13 to 31% in buffaloes in different states of India. During a survey in 14 states (1999-2004), an overall prevalence of 30 % with 32 % in cattle and 23 % in buffaloes was recorded.

The high percentage of animals which are sero positive in the cattle population or in herds which have experienced the disease is due to the presence of PI animals in the herd.

Morbidity and case fatality rates

The incidence of mucosal disease in a herd is usually less than 5% of the animals up to 2 years of age.

In outbreaks of per-acute BVD in immuno-competent non PI animals, morbidity may reach 40% with mortality up to 20%.

Methods of transmission

The major source of infection is the PI animal. Acutely infected non PI animals also can transmit infection.

Direct transmission

Discharges from the respiratory tract of an infected cow, either PI or systematically immune and aborted foetuses.

- Trans-placental transmission
- PI sheep and goats also transmit infection to cattle.

Indirect transmission

- Blood transfusions, nose tongs, rectal gloves, contaminated needles, vaccines, biologicals and even biting flies can transmit BVD experimentally.
- PI and acutely infected bulls can transmit the virus through semen.

Transmission through vaccines

Where live vaccines against BVDV are in use, vaccination of animals in early pregnancy may result in birth of persistently infected animals. Transmission is also possible via live vaccines contaminated with the BVDV from foetal serum used for growing cell cultures.

Clinical findings

Acute infections

Acute infections of cattle occur particularly in young animals, and may be clinically inapparent or associated with diarrhoea. Affected animals may be predisposed to secondary infections, for example those leading to shipping disease, perhaps due to an immunosuppressive effect of the virus.

Bulls may suffer a temporary depression of fertility and can show transient shedding of virus in the semen.

Cows may also suffer from infertility. A serological response is the most certain means of diagnosing a previous infection.

The clinical picture is generally one of high morbidity and low mortality, though more severe disease is sometimes seen. In particular, outbreaks of a severe form of acute

disease with haemorrhagic lesions, thrombocytopenia and high mortality have been reported sporadically from some countries and infection with Type 2 viruses in particular has been demonstrated to cause altered platelet function.

Other acute outbreaks may show fever, pneumonia, diarrhoea and sudden death in any age group, with haemorrhagic signs.

Congenital infections

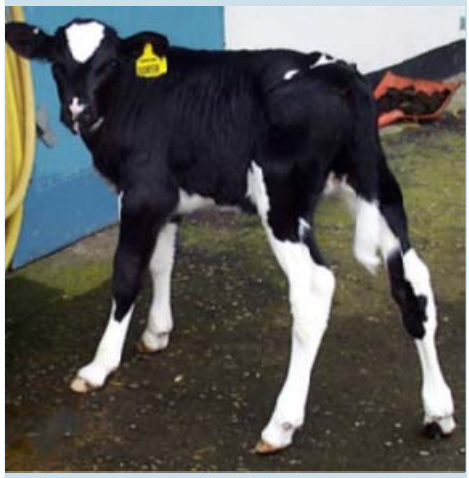
If NCP virus infects the foetus, it may result in abortion, stillbirth, teratogenic effects or a congenital infection that persists in the neonatal calf.

Confirmation that an abortion is caused by BVDV is often difficult to establish, but virus may be isolated from foetal tissue in some cases, or viral antigen or genome may be demonstrated. Although congenital infection with BVDV often leads to abortion, it is not always recognised in the field.

Infection during the first third of the gestation period can result in early embryonic death or mummification. Aborted foetuses may have subcutaneous oedema and copious pleural and peritoneal effusions.

There may also be congenital abnormalities that result in growth retardation and in selective central nervous system (CNS) defects, such as cerebellar hypoplasia and dysmyelination, and eye defects, such as cataracts and retinal atrophy. Sometimes there are skeletal defects, the most advanced of which is arthrogryposis.

Stillborn calves has been reported to be sequel to congenital infection before 150 days of gestation and the calves usually appear to be fully developed at parturition, but fail to survive. However, it has been reported, that in many cases, BVD virus cannot be isolated from these animals and they are PCR negative. If infection occurs after day 150 of pregnancy, the immune system of the foetus will be developed and infection of the foetus will usually result in an antibody response and the birth of a normal calf.



Wide stance is one of the birth defects of the nervous system due to BVD infection
Source : www. nadis.org.uk

Persistent infection

When infections of the foetus occur **before approximately 110 days** of gestation and before immunocompetence, the calf may be born with a persistent infection.

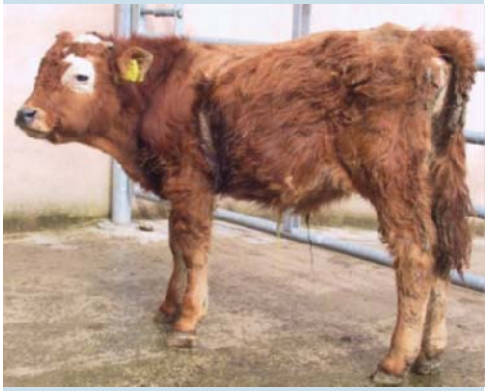
Identification of these animals is readily made by detection of NCP BVDV virus in blood.

Furthermore, animals with a persistent infection will also lack specific antibody, but diagnosis in the young calf, up to approximately 3 months of age, may be confused by the presence of maternal antibody to BVDV. Maternal antibody may also interfere with virus isolation.

In older animals with persistent viraemia infection, low levels of antibody may be present due to their ability to seroconvert to strains of BVDV (including vaccines) 'heterologous' (antigenically different) from the persisting virus. To confirm a diagnosis of persistent infection, animals should be retested after an interval of at least 8 weeks.

There are no pathognomonic lesions in the viraemic calf. The clinical signs vary from the apparently normal healthy animal to the weak, unthrifty calf that has difficulty in standing and sucking.

These latter calves can show CNS defects, such as muscular tremors, incoordination and blindness. They often die within days of birth, thus contributing to the 'weak calf syndrome'. Approximately 1–2% of cattle within a population are persistently infected, with many viraemic animals surviving to sexual maturity and retained for breeding. Calves born to these infected dams are always persistently viraemic, and are often weak at birth and fail to thrive. Persistently viraemic animals are a continual source of infective virus to other cattle, and thus their rapid identification and removal from the herd are required.



A **PI** calf with chronic pneumonia & ringworm infection
Source : www. www. nadis.org.uk

Bulls that are persistently infected usually have poor quality, highly infective semen and, as a result, reduced fertility.

Female cattle used as embryo recipients should always test negative for BVD viraemia before first use. Donor cows that are persistently infected with BVDV also represent a potential source of infection, as oocysts without an intact zona pellucida are shown to be susceptible to infection *in vitro*. Embryos may also become contaminated following acute infection of the donor. Biological materials used for *in-vitro* fertilisation techniques (bovine serum, bovine cell cultures) have a high risk of contamination and should be screened for BVDV. Recent incidents of apparent introduction of virus via such techniques have highlighted this risk.

Mucosal disease

It is well established that PI animals may later succumb to mucosal disease, however, cases are rare. This syndrome has been shown to be associated with the presence of the cytopathogenic biotype, which can arise either through super-infection, recombination between non-cytopathogenic biotypes, or mutation of the persistent biotype.

A **PI** calf (right) compared to similarly-aged normal calf (left)
Source : www. www. nadis.org.uk

Consequently, confirmatory diagnosis of mucosal disease should include the isolation of cytopathogenic virus from affected cattle. This biotype may sometimes be isolated from blood, but it can be recovered more consistently from a variety of other tissues.

Mucosal disease is invariably fatal. Its onset may be so rapid that the first signs seen are dead or moribund animals. However, it is more common for animals to become anorexic over a period of several days, to be disinclined to move and to show signs of

abdominal pain. They can develop a profuse diarrhoea and rapidly lose body condition. Erosions can often be seen in the mouth, particularly along the gingival margin.

Lacrimation and excessive salivation occur. Generally, cases of mucosal disease are sporadic and rare.

Post-mortem examination reveals erosions in the mucosa at various sites along the gastrointestinal tract.

The most noticeable are those overlying the lymphoid Peyer's patches in the small intestine and in the ileocaecal lymph nodes. Severe acute BVD infection can be clinically similar to mucosal disease and confusion can arise, particularly when a number of animals are so affected. Seroconversion among recovered animals is indicative of acute infection, whereas two antigen or virus positive results on samples from an affected animal, taken 8 weeks apart, is diagnostic of mucosal disease. Generally, animals with mucosal disease are antibody negative, though low levels of antibody can sometimes be detected.

Laboratory tests

Agent Identification

Persistently viraemic healthy animals resulting from congenital infection can be readily identified by isolation of noncytopathogenic virus in cell cultures from blood or serum. It is necessary to use an immune-labelling method to detect the growth of virus in the cultures.

Alternative methods based on direct detection of viral antigen or viral RNA in leukocytes are also available. Persi

stence of virus should be confirmed by resampling after an interval of at least 8 weeks. These animals will usually have no or low levels of antibodies to BVDV.

Virus testing can be done via the blood or, particularly in calves < 12 weeks of age, skin (usually a plug of tissue from the ear). Skin testing is useful in younger calves because detection of the virus is not impaired by the presence of antibodies from the colostrum which may be present in the blood.

Viraemia in acute cases is transient and can be difficult to detect. In fatal cases of haemorrhagic disease, virus can be isolated from tissues post-mortem. Confirmation of mucosal disease can be made by isolation of the cytopathogenic biotype of BVDV, particularly from intestinal tissues. Noncytopathogenic virus may also be detected, especially in blood.

ELISA for antigen detection

Several methods for the enzyme-linked immunosorbent assay (ELISA) for antigen detection have been published and a number of commercial kits are available. The best of the method gives sensitivity similar to virus isolation, and may be preferred in those rare cases where persistent infection is combined with seropositivity. Due to transient viraemia, the antigen ELISA appears to be less useful for virus detection in acute BVD infections.

Immunohistochemistry

Enzyme-labelled methods are useful to detect BVDV antigen in tissue sections, particularly where suitable MAbs are available. For persistently infected cattle almost any tissue can be used, but particularly good success has been found with lymph nodes, thyroid gland, skin, brain, abomasum and placenta. Skin biopsies, such as ear-notch samples, have shown to be useful for *in-vivo* diagnosis of persistent BVD infection.

Nucleic acid detection

The RT-PCR method can be adapted to the detection of BVD viral RNA for diagnostic purposes. This may have a special value where low-level virus contamination is suspected. The RT-PCR technique is also sensitive enough to enable the detection of persistently infected lactating cows in a herd of up to 100 animals or more, by testing the somatic cells within bulk milk. A positive result indicates that at least one such animal is present in the milking herd.

Low head carriage & unsteady feet are some birth defects of the nervous system due to BVD infection
Source : www. www. nadis.org.uk

Serological tests

Acute infection with BVDV is best confirmed by demonstrating seroconversion using sequential paired samples from several animals in the group. The testing of paired (acute and convalescent samples) should be done a minimum of 21 days apart and samples should be tested side by side. The enzyme-linked immunosorbent assay for antibody and the virus neutralisation test are the most widely used.

In India, testing facilities for BVD are available with High Security Animal Disease Laboratory (HSADL), Bhopal and NDDB R&D, Hyderabad.

Treatment

Acute BVD

Treatment of any concurrent infections if present.

Persistent infection

Such cattle have often been treated several times for digestive and respiratory infections. PI animals should be disposed of immediately as they act as a source of BVD infection.

Prevention and control

The three main components of BVD control in a herd are:

1. Prevention of introduction of infection into the herd.
2. Identification and elimination of PI animals.
3. Vaccination of breeding animals before breeding.

The following steps should be adopted in the control plan for BVD:

a. Incoming cattle should be isolated, with no fence line contact or runoff going to the current herd.
b. All incoming cattle should be tested on arrival for persistently infected (PI) BVD animals. A retest after 60 days is advisable.
c. All incoming cattle should remain in isolation a minimum of 60 days and until all test results are received.
d. Only semen from negative bulls should be used for artificial insemination (AI).
e. Colostrum from outside sources should not be used. It could be a source of BVD, Johne's disease or other pathogens.
f. Personnel, vehicles and equipment — including, syringes, intra venous tubes etc should not be used on any other farm unless properly disinfected.

Vaccination

Inactivated vaccines

BVD vaccines have been based on a cytopathogenic strain of the virus and fall into two classes: **modified live virus** or **inactivated vaccines.**

Although live virus vaccines are available in some countries, they are used under careful veterinary control because a cytopathogenic strain may precipitate mucosal disease by superinfection of persistently viraemic animals, while in pregnant cattle, a noncytopathogenic component of the vaccine may cross the placenta and infect the foetus.

Inactivated vaccines are more widely used and comprises two doses 3-4 weeks apart before first service followed by booster

(Continued on page 4)

OIE - Significant animal diseases reported to OIE during Apr – Jun'14

Sl.No	Disease Outbreak	Countries reporting
1	Anthrax	Botswana, Croatia, Germany
2	FMD	Botswana (pending), China(O), Russia (O), Tunisia (pending), Zimbabwe (SAT 1)
3	BSE	Brazil, Romania
4	Porcine epidemic diarrhoea	Cananda, Colombia, Dominican Republic, Japan, Mexico
5	Blue Tongue	Greece
6	Lumpy skin Disease	Iran
7	Q fever	South Korea
8	Middle East Respiratory Syndrome (MERS)-Corona virus (MERS Cov)	Kuwait, Oman
9	African Swine Fever	Latvia, Poland
10	Porcine reproductive/ respiratory syndrome	Mongolia
11	African horse sickness	Mozambique
12	Rabbit haemorrhagic disease	Norway
13	Classical swine fever	Russia
14	Swine novel enteric corona virus disesae	USA
15	Vesicular stomatitis	USA
16	Rabies	Uruguay
17	New castle disease	Algeria, Sweden
18	Low pathogenic avian influenza	China, Taipei, USA
19	Highly pathogenic avian influenza	Taipei, Japan, North Korea

RNA revolution - The future of medicine

A series of discoveries in late 20th century revealed several previously unknown forms of RNA that play active, regulatory roles in cells. These latest insights are allowing scientists to create a new world of medications against bacteria, viruses, cancer and other chronic conditions.

Clustered Regulatory Interspaced Short Palindrome Repeats (CRISPR) is a RNA strand that complements the exact genetic sequence in the DNA to be modified. This is then attached to a protein that cuts the DNA. The combined RNA protein complex searches out the targeted DNA and permanently disrupts it, small pieces of corrective DNA also can be added at the same location in a separate process.

Small interfering RNA (siRNA) complements a section of the m-RNA that is to be disrupted. The siRNA is taken up by a complex of proteins that cut the singled-out mRNA at the spot indicated by siRNA.

MicroRNAs does not have to be a perfect match for the mRNA being targeted. A small number of microRNAs can temporarily alter production of many proteins.

Source : Scientific American, April 2014.

(Continued from page 3)

vaccination at 12 months' intervals. If all breeding females are vaccinated then this will control disease by preventing BVD infection of the developing foetus during pregnancy and production of PI calves.

No vaccines are presently available in India.

BVD eradication

BVD eradication is possible following whole herd blood testing and elimination of all PI carrier animals. If eradication is to be achieved then strict herd biosecurity measures must be maintained to prevent re-introduction of virus infection as the herd will soon become naïve and therefore fully susceptible to infection.

Welfare implications

Cattle with mucosal disease must be culled immediately upon diagnosis. Calves born with eye and brain defects, due to virus infection during their development, should also be culled.

References

i. http://www.oie.int/fileadmin/Home/eng/Health_standards/tahm/2.04.08_BVD.pdf
ii. http://www.aphis.usda.gov/animal_health/emergingissues/downloads/bvdinfosheet.pdf
iii. http://www.nadis.org.uk/bulletins/control-of-bovine-virus-diarrhoea-(bvd).aspx
iv. *Rev. sci. tech. Off. int. Epiz.*, 1990, 9 (1), 75-93.
v. http://hsadl.nic.in/BVD.htm
vi. http://www.merckmanuals.com/vet/digestive_system/intestinal_diseases_in_ruminants/intestinal_diseases_in_cattle.html
vii. Veterinary Medicine-A textbook of the diseases of cattle, sheep, pigs, goats and horses : Otto M. Radostitis, C.C. Gay, D C Blood, K W Hinchcliff , 9th edition

It takes a Prion to remember

The protein family notorious for causing diseases like scrapie and mad cow appears to play an important role in healthy cells. Animals studies reveal that they make and use prions in their nervous system as a part of an essential function: stabilizing the synapses involved with forming long-term memory. These are not infectious, but on a molecular level chain up exactly as their disease causing brethren!

Source : Scientific American, May 2014.

Biosensors for germ detection

Machines that can quickly identify virtually any bacterium, virus or fungus are becoming a reality. These sophisticated biosensors that employ a combination of biological, physical and mathematical tools that can identify any of the more than 1000 pathogens that cause human illness.

The lab technician draws blood from a patient– though most genetic material is of the person , it would also contain genetic material of the pathogen causing the illness.

Carefully chosen primers (snippets of nucleic acids) are then added. The primers seek out foreign genetic material and multiple copies of these targeted sections are then generated using Polymerase Chain Reaction (PCR).

A mass spectrometer is used to weigh the amplified material and based on complex mathematical formulae, deduce the total number of each code found in unknown sequences.

Matching the calculated number of code letters with those in a database of specific viruses, bacteria or fungi uncovers the pathogen's identity.

Source : Scientific American, June 2014.

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