Bovine Viral Diarrhoea (BVD)
Summary

Introduction

1. This note provides a brief summary of the Disease and Product analysis prepared by a DISCONTOOLS group of experts on bovine viral diarrhoea (BVD). They reviewed the current knowledge on the disease, considered the existing disease control tools, identified current gaps in the availability and quality of the control tools and finally determined the research necessary to develop new or improved tools. Full details are available on the web site at http://www.discontools.eu/.

Disease profile

2. Bovine viral diarrhoea (BVD) is a pestivirus infection affecting cattle and other ruminant species. Clinical presentation may vary from subclinical to severe and can include enteric, respiratory or reproductive disease. Infection *in utero* can result in embryonic death or congenital defects including persistent infection, the latter only occurring with the non-cytopathic biotype. Persistent infection results in lifelong replication and shedding of the virus. The persistently infected (PI) animal is a major vector for viral dissemination. Persistently infected animals are also at risk of developing mucosal disease (MD) if they are superinfected with an antigenically related cytopathic virus. The immunosuppression that accompanies acute infections predisposes animals to secondary infections. In addition, synergy may occur between the pestivirus and secondary pathogens that results in increased virulence.

3. BVD is caused by genetically and antigenically distinct viruses belonging to three different species of the *Pestivirus* genus within the *Flavivirus* family, bovine viral diarrhoea virus 1 (BVDV-1, Pestivirus A), BVDV-2 (Pestivirus B) and HoBi-like viruses (Pestivirus H). These viruses are single-stranded, positive-sense enveloped RNA viruses. Multiple subgenotypes have been identified within each viral species. Viruses in each of the three species may exist as one of two biotypes, cytopathic and non-cytopathic, based on their activity in cell cultures. Only viruses from the non-cytopathic biotype can establish persistent infections. The three species may be differentiated from each other and from other pestiviruses by genetic analysis (most frequently based on comparison of 5'UTR, N^{pro} and/or E2 partial sequences), monoclonal antibodies directed against the E2 and E^{rns} major glycoproteins and comparative neutralization using polyclonal sera. Molecular methods are available that enable virus typing directly from blood samples. Overall, BVDV-1 viruses are more common worldwide, however prevalence of the three species vary by geographic location. The non-cytopathic biotype predominates in ruminant populations. In most cases acute infections with the three species are clinically indistinguishable.

4. Cattle of all ages are susceptible to infection. Clinical signs range from subclinical to fatal (MD and haemorrhagic syndrome). Viral strains can vary in virulence with some strains reproducibly causing haemorrhagic syndrome following acute infection. However, frequently acute infections result in transient clinical disease with unspecific signs (respiratory tract, intestinal tract, fever, leukopenia).The virus spreads mainly by contact between cattle or via indirect contact, but vertical transmission plays the major role in its epidemiology and pathogenesis. Infections of the bovine foetus may result in abortions, stillbirths, teratogenic effects or persistent infection of the neonatal calf. PI (viraemic) animals may be born as weak, unthrifty calves or as normal appearing calves without overt clinical pathology. Antibody positive pregnant cattle ("trojan" dams) carrying persistently infected calves are an important means by which infection may be introduced to herds. There is some degree of cross-protection between species. Cross-reactivity in also observed with other pestivial species such as classical swine fever virus (CSFV, Pestivirus C) and border disease virus (BDV, Pestivirus D).

Risk

1. There are no known risks to human health. In the absence of systematic control programs, disease results in major economic losses for the cattle industry worldwide. Reproductive effects figure prominently (abortions, deformed offspring, PI animals, conception failure) as does the contribution of infection to bovine respiratory disease complex (BRDC). PI animals are the most serious risk for any herd as they are the main source of infection and their role on the epidemiology of the disease cannot

be overstated. However, significant financial losses also occur due to acute infections through a general impairment of health and its consequences on productivity in endemically infected herds (chiefly due to the immunosuppressive effect of the virus).

Diagnostics

2. Current diagnostics are focused on detection of BVDV-1 and BVDV-2 infection/exposure primarily in cattle. Surveillance for HoBi-like virus or infection/exposure in domestic species other than cattle and for free ranging wildlife populations is not regularly performed.
3. For virus detection, isolation is the gold standard but for routine screening it is more common to use antigen capture ELISAs or real-time RT-PCR. Many real-time RT-PCR kits are commercially available for testing blood, tissues (including ear notch samples) and individual or bulk milk samples. The RT-PCR is recognized as the most sensitive method for virus detection. Antigen capture ELISAs are commercially available for detection of virus in blood and tissues, including ear notch samples. Repeated testing within a specified time frame allows persistently and transiently infected animals to be differentiated. The industry standard for determination of persistent infection is two positive tests taken at an interval of 3 weeks or more. This is based on the assumption that acute infections are cleared within three weeks. However, it has been observed, in a minority of cases in neonatal calves and young stock that transiently infected young animals may test positive for longer than two weeks. Freedom from virus in a population rest on systematic surveillance.
4. Routinely, blocking ELISAs (NS3, E^{rns}) or indirect ELISAs are used for detection of antibodies in serum, plasma and milk (individual or bulk). At the individual level, seroconversion may be demonstrated via ELISA or serum neutralisation test (SNT) using paired serum samples taken at least 21 days apart (but this is rarely done). At the herd level, antibody detection in bulk milk or spot samples are more commonly used as the basis for surveillance regarding the likely presence or absence of persistently infected animals in a herd. Serosurveillance cannot differentiate between natural exposure and vaccination. Although antibody levels tend to be higher after natural exposure or repeated exposure to a persistently infected cohort, this is not a reliable differential. Further, because there is cross-reactivity between pestiviruses, the differentiation of antibodies to BVDV-1 and BVDV-2 from those arising following infection with other viruses such as BDV is of great importance in areas where cattle and sheep are kept together. At present, in the absence of suitable ELISA tests, it is only the time-consuming and costly cross-SNT that can differentiate between antibodies to the different pestivirus species.
5. Opportunities for future test developments are linked to BVDV eradication/control programmes. A ban on vaccination after BVDV eradication may provide an opportunity to develop antibody detection tests with higher sensitivity that can be applied to bulk milk testing to detect early circulation of virus and associated seroconversion. Alternatively, protection of BVDV free herds by vaccination creates the need for the development of DIVA systems (marker vaccine plus corresponding antibody detection test). On-site tests might be helpful to create awareness for BVDV and support activities leading to eradication/control programmes. Highly sensitive but robust pathogen detection systems are needed for each different diagnostic specimen (ear notch, blood, semen, milk etc.), which are suitable for sample pooling. Test systems which allow multiplexing of samples in one test run may facilitate running different programmes at the same time thereby saving costs.
6. In the later phases of eradication spot testing protocols may be employed (testing of a few young animals for the presence of antibodies to indirectly indicate exposure).

Vaccines

7. Current vaccines are licensed for the control of BVDV-1 and/or BVDV-2 in domestic cattle. Vaccines may have a varying claim of efficacy against two key aspects of the disease, one for the prevention/reduction of acute clinical disease and one for the prevention/reduction of persistent infections. Vaccine strains are grown to high titre on conventional bovine kidney cell lines although it would be valuable to look for alternative production cell lines. Modified live virus (MLV) and inactivated vaccines containing BVDV-1 and 2 species are available globally although efficacy, duration of immunity and cross protection are still issues. BVD control programmes with improved biosecurity (which may or may not include vaccination) and elimination of PI animals, are underway in a number of countries and regions. Vaccines containing the Hobi-like virus species are not

commercially available and there are no control programs addressing the eradication of HoBi-like viruses.

8. The prevention of persistently infected (PI) cattle by protecting the foetus is a major goal of control by vaccination. While vaccines reduce the number of PI cattle born, the complete prevention of foetal infection requires the establishment of near sterilizing immunity at the herd level. To date this cannot be reliably achieved, due to variation in the immune response of cattle within a herd, antigenic differences between strains and vaccination protocols. The evaluation of the efficacy and cost-efficiency of different vaccines and vaccination strategies under field conditions is missing (in particular, one that also incorporates user compliance). DIVA vaccines and accompanying assays are currently not available.

Pharmaceuticals

9. No pharmaceutical therapy is available commercially. Prophylactic treatments may be used and antibiotics may be used to treat secondary infections. Prototype vaccines are being developed and tested for related human viruses as well as for CSFV. Other possibilities include siRNA, new antivirals or breeding resistant animals.

Knowledge

10. Knowledge gaps that affect vaccinology, epidemiology and control include the following.
    a. The role of innate and cellular immunity in the suppression of the immune response is not fully understood.
    b. The virus's ability to cross the placenta and cause reproductive disorders is a major mechanism of pathogenicity and the action of neutralising antibodies versus cell-mediated immunity in foetal protection in preventing this is not well understood.
    c. The mechanism(s) of immune suppression and mechanism(s) associated with pathogen synergy that result in increased virulence of other pathogens in the presence of BVDV needs further investigation.
    d. The factors determining strain virulence are undefined.
    e. Mechanisms of adaptation of viruses to different hosts (including domestic species outside of cattle and free ranging wildlife species) are largely unknown.
    f. Estimates of the impact of endemically infected cattle populations on the economics of animal production (particularly as it applies to different farming practices), antimicrobial usage, and the general health and wellbeing of the domestic and wildlife population are lacking.
    g. Risk factors for reintroduction of BVDV-1 and/or BVDV-2 or introduction of HoBi-like viruses into BVD-free cattle populations are not fully defined.

Conclusions

11. A number of European countries have experience with systematic large scale BVDV control, at the country or regional levels, aimed at eradication of BVDV-1 and BVDV-2 from domestic cattle populations. Details of the requirements for approval of a programme of eradication, and recognition of freedom, are provided in Delegated Regulation 2020/689 under EU regulation 2016/429 ("the Animal Health Law"). Despite different pre-conditions in terms of initial prevalence, herd density, regulatory support etc., these have all proven to be successful in effectively reducing or eliminating the infection. These programs are based on systematic strategies to detect and remove persistently infected cattle combined with biosecurity protocols. The EU Regulation 2020/689 permits vaccination while eradication programmes are underway. However, routine vaccination is prohibited once eradication has been achieved. Non-systematic vaccination strategies have been widely used in many settings but so far with no proof of sustainable decrease in disease prevalence or impact. The main obstacles to implementing national or regional systematic control programmes can be found in the attitudes and priorities of influential individuals/groups within the industry, academia and authorities rather than the availability of suitable tools for the control of BVD.