Swine vesicular disease surveillance and eradication activities in Italy

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Summary
Swine vesicular disease (SVD) was first observed in Italy in 1966, and was initially diagnosed as foot and mouth disease (FMD). The causative agent of SVD was classified as an Enterovirus within the family Picornaviridae. It was included in the list of diseases notifiable to the World Organisation for Animal Health (OIE) because of the similarity of its lesions to those produced by FMD; however SVD is often mild in nature and may infect pigs subclinically.

During the last decade SVD has been persistently reported in Italy, and surveillance and eradication activities are in place. The central and northern parts of Italy have been designated SVD free since 1997, while the southern regions have not achieved disease-free status. However, occasional outbreaks of SVD have occurred in central and northern Italy and have been eradicated using rigorous control measures.
Most recent SVD outbreaks in Italy have been subclinical; SVD can rarely be diagnosed now on the basis of clinical signs and it is necessary to use laboratory diagnosis.
This paper examines the epidemiology of SVD in Italy, and considers the measures adopted in Europe for SVD control on the basis of current knowledge of the disease.

Keywords
Eradication – Italy – Surveillance – Swine vesicular disease.

Introduction
Swine vesicular disease (SVD) was first observed in Italy in 1966, when it was identified clinically as foot and mouth disease (FMD) (16). Despite the production of similar clinical signs, the biological, physical and chemical properties of the virus showed that it differed from FMD, vesicular stomatitis and vesicular exanthema viruses. Swine vesicular disease virus (SVDV) was classified as an Enterovirus within the family Picornaviridae. Phylogenetic studies suggest that it evolved as a genetic sub-lineage of the human pathogen coxsackievirus B5 between 1945 and 1965 (20). Although all current SVDV isolates are of a single serotype, four antigenic variants, distinguishable by the use of monoclonal antibodies, have been reported since its first appearance (8). The most recent group consists of viruses isolated in European Union (EU) countries since 1992. This variant, which has maintained a stable antigenic profile, has been circulating in Italy for more than 10 years, and has sporadically been detected in other EU countries and in Taipei China (5).

Swine vesicular disease virus is extremely resistant in the environment and to normal disinfectants; the virus is noted for its persistence over a pH range from 2.5 to 12. The extraordinary stability in the environment of SVDV is a feature of major importance in the epidemiology and control of the disease (15).
Swine vesicular disease virus causes a vesicular disease in pigs, with clinical signs that resemble those of foot and mouth disease, but it does not affect ruminants. Direct contact of susceptible animals with infected pigs or contaminated materials, premises or transport vehicles is usually necessary for disease transmission. The infection can also be transmitted via the feeding of meat from pigs slaughtered during the viraemic period or offal from infected pigs.

Unlike FMD, swine vesicular disease has a limited tendency to diffuse, even within infected premises. Spread from one pen to another may not occur in the absence of movement of infected pigs or contaminated material. This is why SVD is regarded as a ‘pen disease’ rather than a ‘farm disease’ (15).

The viraemic period of SVD lasts for two to three days. After virus has been cleared from the blood and most internal organs, it can still replicate in the gastrointestinal tract and be shed in the faeces for a period of days to weeks.

The disease is characterised by the appearance of vesicular lesions on the limbs, snout, lips and tongue of pigs; the severe form is usually only seen when animals are housed on a concrete floor in humid conditions (9). However, SVD is often mild in nature and may infect pig herds subclinically. Stress caused by mixing and transporting animals may play an important role in increasing virus shedding, the severity of clinical signs and the likelihood of further spread of disease.

Although the disease is frequently mild in nature it was included in the list of diseases notifiable to the OIE because of the similarity of its lesions to those produced by FMD. If pigs show suspicious clinical signs, rapid differentiation between FMD and SVD is necessary. Very sensitive and specific tests are available for SVD diagnosis and for its differentiation from FMD.

In the majority of recent outbreaks in Italy, the course has been subclinical; in such cases, SVD can rarely be diagnosed on the basis of clinical signs and it is necessary to resort to laboratory investigation (4).

During the last decade, SVD has been persistently reported in Italy and for this reason surveillance and eradication activities are currently in place.

Swine vesicular disease control measures

Swine vesicular disease control measures are currently established in the EU by Council Directive 92/119/EEC, Annex II. These measures take into consideration the fact that the disease is included in the OIE list of notifiable diseases (10).

Similar to the legislation on FMD control, in the case of confirmation of SVD, Directive 92/119/EEC provides for a rigorous stamping-out policy, together with the establishment of protection and surveillance zones in which movements of pigs are restricted or banned, and cleansing and disinfection of infected premises and vehicles.

Diagnostic procedures for swine vesicular disease are laid down at Community level in Commission Decision 2000/428/EC. This establishes diagnostic procedures, sampling methods and criteria for the evaluation of the results of laboratory tests for the confirmation and differential diagnosis of swine vesicular disease (11). The Decision does not lay down requirements for routine surveillance, which have been established for Italy by Commission Decision 2005/779/EC ‘concerning animal health protection measures against swine vesicular disease in Italy’ (13).

In 1973, Italy placed SVD on its list of notifiable diseases. A surveillance plan has been implemented since 1995, the aim of which is to achieve eradication by means of an SVD health certification scheme in each Italian region. The plan is updated annually according to the epidemiological situation and has been approved by the EU Commission. The main activities of the plan are:

- a random serological check of all breeding farms
- a virological check of faecal samples collected from dealers’ premises (virus monitoring on dealers’ premises was introduced in 1998)
- random serological monitoring of imported pigs (introduced in 2001).

Extraordinary control measures have been adopted at the regional level in cases where there is a serious risk of disease spread (for example Campania, Calabria) or when the disease has occurred in an area with a high density of pigs (e.g. Lombardy in 1999 and 2002) (18, 19).

Declaration of an SVD outbreak follows the rule laid down in Directive 92/119 (10), which establishes that SVD is confirmed:

a) on holdings in which SVDV is detected either in pigs or in the environment;
b) when the holding contains pigs that are seropositive for SVD, provided those pigs or others on the holding show lesions characteristic of SVD;
c) when the holding contains pigs that show clinical signs of disease or are seropositive, provided there is a direct epidemiological connection with a confirmed outbreak;
d) on any holding where seropositive pigs are detected. In this case, before confirming the presence of the disease, further investigations are undertaken, in particular resampling and retesting with an interval of at least 28 days between successive samples. If subsequent investigations show no evidence of the disease, but the pigs are still seropositive, the competent authority ensures that the pigs tested are slaughtered.

As soon as the presence of the disease is suspected the competent authority carries out an epidemiological investigation to determine the possible origin of the disease on the holding. The origin of the disease must be traced and the spread of infection stopped.

In order to collect and analyse the data gathered during the surveillance activities for SVD in Italy, a web-based information system for the management of the national surveillance plan has been implemented (3).

Laboratory diagnosis and virus characterisation

Due to the characteristic subclinical course of the disease, clinical inspection is frequently inconclusive; for a surveillance plan aimed at SVD eradication it is therefore necessary to resort to laboratory diagnosis.

In Italy laboratory diagnosis is carried out according to the rules laid down in EC Decision 2000/428 (11) and in the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (17).

Antibody detection

The OIE reference test, which is a competitive enzyme-linked immunosorbent assay (ELISA) based on the monoclonal antibody 5B7, is used as a screening test (7, 17). Serological screening is carried out at regional level using kits supplied by the National Reference Laboratory (NRL) in Brescia. The performance of the screening test in the regional laboratories is monitored yearly through ring tests organised by the NRL. Confirmatory testing of doubtful and positive samples is performed at the NRL using the prescribed virus neutralisation test. All positive sera are also submitted to an isotype-specific ELISA to identify the class of anti-SVDV immunoglobulins, in particular IgM and IgG. The presence of IgM, alone or together with IgG, is evidence of recent infection and indicative of virus shedding, while detection of IgG alone suggests an older infection (7).

Virus detection

Virological tests are carried out at the NRL. For virological testing of faecal samples (for surveillance and in suspected outbreaks) the recommended virus isolation (VI) test was used until 2002. Since then, virus isolation has been replaced by a reverse transcriptase polymerase chain reaction (RT-PCR). The RT-PCR is based on preliminary immunocapture of SVDV by a monoclonal antibody coated on to an ELISA plate; this strategy prevents false-negatives that may occur due to the presence of inhibitors of the reaction in faecal samples.

The immune-PCR was extensively validated on a large number of field samples, and showed advantages in terms of sensitivity, rapidity (1–2 days vs 1 week) and simplicity compared with the ‘confirmatory’ VI test (14). The VI test is performed on IBRS-2 or equivalent cell lines and often requires two to four subcultures. The cell lines used are susceptible to SVD virus as well as to other enteroviruses potentially present in porcine faeces. Samples containing other viruses may give a false-negative result for SVDV when growth of the other enteroviruses exceeds or affects the growth of SVDV. In our experience, the prevalence of enteroviruses other than SVDV in faeces samples submitted for the VI assay corresponds to 20% to 25% of the samples examined. Moreover, due to inadequate preservation and transportation of samples, the SVD virus present in faecal samples may no longer be viable.

Due to these limitations, a proportion of faeces samples that are positive by immune-PCR score negative when examined by the reference VI assay. The specificity and higher diagnostic sensitivity of the immune-PCR are supported by evidence that, whenever conditions have enabled us to perform follow-up testing, all samples positive on immune-PCR, even when the VI test failed to confirm the positive result, originated from seropositive herds or farms epidemiologically connected with previously confirmed outbreaks. In these cases, the positive immune-PCR results were used to declare an outbreak, because the criteria established in EU Commission Decision 2000/428/EC were fulfilled (11). Follow-up testing is seldom feasible on dealers’ premises, due to the conditions of management and husbandry. Taking into account the role such premises play in the spread of disease, restrictive criteria are applied to farms of this type. Any positive immune-PCR result from a dealership is considered to represent an outbreak.

Characterisation of swine vesicular disease virus isolates

The antigenic profile of SVDV isolates collected from outbreaks was studied at the Italian Reference Laboratory using panels of monoclonal antibodies that identify six
characterised antigenic sites (6), while genomic analyses were conducted at the EU Reference Laboratory by comparison of VP1-coding sequences.

The pattern of reactivity with monoclonal antibodies provided evidence that all the isolates belonged to the fourth and most recent antigenic group, which was first detected in the EU in 1992 (8) and is still circulating (6). Antigenic drift was observed in one antigenic site, which was subject to frequent variability; other minor antigenic mutations were extremely sporadic and did not become stabilised in the field, leading to the conclusion that the antigenic profile of SVDV isolates collected over approximately 15 years (1992 to 2005) has been stable.

The results of nucleotide sequencing confirmed that the isolates are clustered in a unique genomic lineage, which is known to be correlated to the fourth antigenic group (8), although progressive genomic divergence is apparent from the older (isolated in 1992 and 1993) to the more recent isolates. An exception to this tendency concerns two geographically related isolates from 2004, which clustered in a distinguishable genomic sub-lineage together with viruses isolated in Portugal at the end of 2003 and the beginning of 2004. This cluster appears to be most closely related to older isolates, and in particular to one strain isolated in Portugal in 1995 (N. Knowles and D. Paton, unpublished data). One explanation could be that SVDV has persisted unchanged in parts of Europe since the 1990s and has later resurfaced in Portugal, from where it may have spread to Italy. Alternatively, this cluster could represent a reintroduction from outside the European Union, where subclinical infection may be missed in the absence of active surveillance.

Surveillance and eradication activities in Italy: results

The results of the main activities of the surveillance plan in Italy are reported in Figures 1 and 2; the results correspond to a 10-year period for breeding farms and a period of 8 years for dealers’ premises. During the 10-year period the criteria for control of the disease on farms have changed slightly.

The central and northern parts of Italy are designated SVD free and have maintained this health status since 1997, while Campania, Calabria and Sicilia have never attained SVD-free status (Figure 3).

In recent years SVD outbreaks have been persistently identified in the regions that do not have disease-free status; however from time to time SVD outbreaks have occurred in other regions, both in central and in northern Italy (in 2002 and 2004). These outbreaks have always been eradicated following the adoption of rigorous control measures, and on some occasions extraordinary surveillance plans have been implemented to verify SVD eradication (Lombardy, 1999 and 2002) (18, 19).

In 2004 Abruzzo was regionalised and lost its SVD-free status because 65 outbreaks were identified and the origin of the infection remained unknown. At the end of 2006 a new SVD epidemic occurred in the northern region of Italy; the relevant data are not included in this report because eradication activities are still ongoing.

In the 5 years between 2001 and 2005, a total of 352 outbreaks were detected in Italy:

- 32 (9.1%) on breeding farms; of these, 19 (59.3%) were primary outbreaks, while 13 (40.3%) were secondary
- 71 (20.2%) on dealers’ premises; of these, 59 (83.1%) were primary outbreaks and 12 (16.9%) were secondary
- 249 (70.7%) on fattening farms; of these, 19 (7.6%) were primary and 230 (92.3%) were secondary outbreaks.

Fattening farms were most commonly involved in these outbreaks; SVD was usually detected by tracing animals back to infected dealers’ premises. In fact, the outbreaks on fattening farms were very often secondary outbreaks. These farms, given the specific role they play in the production cycle, have an extremely limited part in the spread of the disease. Moreover, 84% of the cases concerned small-sized holdings with fewer than 10 animals, which were often reared for home consumption and represented a dead end for disease spread.
An important role in the spread of SVD was played by dealers’ premises, which gave rise to 261 (74.1%) of the outbreaks detected in the 5-year period. The origin of the infection in these holdings very often remained unknown (83.1% of primary outbreaks), this could be due to weaknesses in the systems of pig registration and recording of animal movements. Furthermore, when dealers’ premises represented secondary outbreaks, the origin was traced to another dealer’s premises in 100% of cases. This confirms the presence of a network among dealers that enhances disease spread.

According to the epidemiological investigations carried out during the outbreaks, SVD diffusion was mainly related to movements of pigs, transportation vehicles and contaminated material. Results of the epidemiological investigations confirmed that, unlike FMD, SVD has a limited tendency to spread even between pens of the same farm. Hence, the incursion of SVD on to a farm or in to a ‘new territory’ can be avoided by adopting routine biosecurity measures.

Despite the possibility of persistence of SVDV in meat originating from infected pigs, swill feeding has not been found to be an important cause of disease spread in Italy.

Since 2000 a change in the clinical trend of SVD was highlighted (Figure 4): no clinically affected animals were
found on 342 out of 357 holdings (95.8%) on which SVD outbreaks were confirmed in the period 2000 to 2005, and mortality in these outbreaks was almost nil (24/21, 048; 0.1%). The lack of clinical signs made clinical diagnosis impossible. The continued presence of SVD in Italy was detected by means of the surveillance included in the framework of the eradication programme put in place in 1995, which includes serological and virological monitoring of pig holdings, collection centres and dealers' premises.

Discussion

The results of the epidemiological investigations conducted during the 2000-2005 outbreaks in Italy confirmed that SVD spread is related to the movement of animals (growers and sows), personnel and contaminated vehicles from the infected farms. The virus is highly resistant in the environment, and hygiene measures on farms and transportation vehicles are not always correctly applied. The different types of pig farms involved in the production cycle, and hence the management and frequency of animal and vehicle movements, played a varying role in disease spread. Dealers' premises were found to play an important role in the spread of the disease, and on these farms the origin of the infection often remained unknown. Weaknesses in the swine registry and in recording of animal movements may compromise eradication activities.

Due to the frequently subclinical or undisclosed course of SVDV infection, surveillance and eradication activities are difficult to carry out; in fact, the early detection phase can be missed and often by the time the disease is identified it has already spread. Given the subclinical course of the infection, at present SVD in Italy does not represent a 'health problem' but a commercial issue. People operating in the sector do not understand the need for commercial restrictions following the confirmation of an outbreak because they do not perceive the economic importance of the disease, and, consequently, they do not participate in the control measures.

The Italian regions have never attained SVD-free status simultaneously. This is partly due to the different farm management situations that exist within Italian territory. Intensive breeding predominates in the northern and central parts of the country; therefore there are strong and understandable reasons why breeders here would like to maintain an SVD-free zone. The free zone enables them to export live animals and by-products abroad. The same economic and commercial motivations are not shared by southern breeders; in this region there are mainly small family holdings and farmers are therefore less interested in exporting their products.

In an SVD outbreak all the animals, their products and equipment have to be destroyed, and the owner is compensated for 100% of the market value (1, 2).

The detection and rate of reporting of certain contagious benign diseases, such as SVD, in densely populated areas should be considered in the light of market price trends at the moment of detection. It should be remembered that when the first outbreak of the 1998 to 1999 epidemic in the Lombardy region was initially detected, it coincided with the lowest point of a market downtrend and the disease notification came from the breeder himself (18) (Fig. 5).

According to Directive 92/119/EC, which was first enforced in 1992 and has not been subsequently updated, suspicion of SVD and imposition of the related control measures are based on clinical evidence. Given what we now know about this disease it is clear that this is no longer adequate: due to the subclinical course, clinical examination is ineffective and the infection may be missed.

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Currently, SVDV infection can only be confirmed or excluded by means of laboratory investigation. Therefore sero-surveillance is essential to detect subclinical or undisclosed clinical infection and to prove the absence of the virus.

Apart from Italy, the only other countries in the EU to have implemented extensive surveillance programmes for SVD are the Netherlands and Spain. A certain level of sero-surveillance is in place in several Member States, while in others there is very little, or, in some cases, none at all. These States still rely on SVD detection by farmers or veterinarians on the basis of clinical signs (12).

Hence, if the disease continues to be considered a serious threat, the current legislation should be modified to take into account the recent knowledge of SVD. Compulsory surveillance based on laboratory testing should be extended to the other Member States and surveillance criteria and guidelines should be established by the OIE. This would be necessary to ensure disease freedom and the immediate application of appropriate control measures to prevent spread should disease be detected.

The reason why SVD in Italy often has a subclinical course has not yet been explained; it could be related to the improved management and husbandry conditions on pig farms or the consequence of a decrease in virulence of the current Italian strain of the virus. If the latter is true we cannot exclude the possibility that the virus may re-acquire its original level of virulence.

Activités de surveillance et d’éradication de la maladie vésiculeuse du porc en Italie

S. Bellini, U. Santucci, G. Zanardi, E. Brocchi & R. Marabelli

Résumé
La maladie vésiculeuse du porc, confondue initialement avec la fièvre aphteuse, a été signalée pour la première fois en Italie en 1966. L’agent responsable de la maladie vésiculeuse du porc est un Enterovirus appartenant à la famille des Picornaviridae. La maladie figure dans la liste des maladies à déclaration obligatoire de l’Organisation mondiale de la santé animale (OIE) en raison de la similitude des lésions qu’elle induit avec celles attribuables à la fièvre aphteuse ; néanmoins, la maladie vésiculeuse du porc se présente souvent sous forme infraclinique et mineure.


Les derniers foyers de maladie vésiculeuse du porc survenus en Italie étaient des infections infracliniques ; il est rare que la maladie vésiculeuse du porc puisse être diagnostiquée sur la base des signes cliniques de sorte que le diagnostic doit être fait au laboratoire.

Les auteurs exposent l’épidémiologie de la maladie vésiculeuse du porc en Italie et examinent les mesures de lutte adoptées en Europe à la lumière des connaissances actuelles sur la maladie.

Mots-clés
Vigilancia y actividades de erradicación de la enfermedad vesicular porcina en Italia

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Resumen
En 1966 fue observada por primera vez en Italia la enfermedad vesicular porcina (EVP), diagnosticada en un principio como fiebre aftosa. El agente causal de la EVP fue clasificado como un Enterovirus de la familia Picornaviridae. La EVP fue incluida en la lista de enfermedades de notificación obligatoria a la Organización Mundial de Sanidad Animal (OIE) por la similitud de sus lesiones con las que induce la fiebre aftosa. La EVP, sin embargo, suele revestir carácter leve e incluso puede ocasionar infecciones asintomáticas.

En el último decenio se ha comunicado una y otra vez la presencia de EVP en Italia, por lo que ahora mismo hay medidas de vigilancia y erradicación en marcha. Las zonas central y septentrional del país fueron declaradas "libres de la enfermedad" en 1997, estatuto al que aún no han accedido las regiones meridionales. Sin embargo, en el centro y el norte de Italia se han seguido produciendo brotes ocasionales, que han sido erradicados gracias a estrictas medidas de control.

Los brotes más recientes de EVP en Italia han sido de carácter subclínico. Hoy en día pocas veces es posible diagnosticar la enfermedad atendiendo a la sintomatología, por lo que es preciso emplear el diagnóstico de laboratorio.

Los autores examinan la epidemiología de la EVP en Italia y las medidas adoptadas en Europa para controlarla, basadas en lo que actualmente se sabe de la enfermedad.

Palabras clave

References


