Potential animal health hazards of pork and pork products

S. Farez & R.S. Morley

Animal and Plant Health Risk Assessment Network, Canadian Food Inspection Agency, 3851 Fallowfield Road, Nepean, Ontario K2H 8P9, Canada

Summary
The animal health hazards associated with the importation of pork and pork products include four viral agents: foot and mouth disease, classical swine fever (hog cholera), African swine fever, and swine vesicular disease viruses. The safety of importing pork from a zone infected with one or more of these diseases can be adequately determined only through risk assessment. This also applies for the safety of importing pork products which have undergone some form of processing (fully cooked pork products are not counted here). For each disease, the agent (pH and temperature lability), target organs, agent survival in pork and pork products, and agent quantification are discussed. Agent quantification is an input of the risk assessment which measures the viral titres in waste pork and pork products in relation to the oral infective dose estimated for each disease. Two other viral diseases, transmissible gastroenteritis of pigs and porcine reproductive and respiratory syndrome, are presented to illustrate why these two diseases are not hazards when associated with pork and pork products.

Keywords
African swine fever - Classical swine fever (hog cholera) - Foot and mouth disease - Porcine reproductive and respiratory syndrome - Risk assessment - Swine vesicular disease - Transmissible gastroenteritis of pigs.

Introduction
The introduction of animal diseases through the importation of pork and pork products is a concern for all swine-rearing countries. While the trade in pork is intended for human consumption, the intentional or inadvertent feeding of any uncooked waste to pigs could introduce animal disease.

The Office International des Epizooties (OIE) International Animal Health Code (63, 69) describes the sanitary measures for the safe importation of pork and pork products from zones infected with certain swine diseases. The importation of pork (other than fully cooked products) from these zones is generally considered a high risk. It is therefore advisable to conduct a risk assessment for both chilled or frozen pork and for pork products which have been subjected to a process such as heat treatment, freezing, maturation and ageing, deboning, acidification, additives, curing, desiccation, etc.

Preceding risk assessment is the hazard identification. Hazard identification is the process which identifies potential risk agents and the conditions under which they may produce adverse reactions. Each hazard necessitates a risk assessment.

The risk assessment must determine, characterise, and quantify the following factors:

a) the potential of the source to release a risk hazard or risk agent
b) the intensity, frequency, and duration of exposure, and the nature of the animal and human populations which might be exposed
c) the relationship between exposure and the resulting biological and economic consequences.

The final outputs of this process are estimates of the magnitudes of possible adverse health and economic consequences, including a characterisation of the probabilities, uncertainties, or degree of confidence associated with these estimates (16).

In this paper, the authors discuss the potential hazards associated with trade in pork and pork products and present some of the inputs and the relevant information for a risk assessment on the safety of importing from zones where these hazards exist. The paper is restricted to a discussion of the
commodity-related inputs. These are as follows: agent (pH and temperature lability), target organs, agent survival in pork and pork products, and agent quantification. Agent quantification is an input of the risk assessment which quantifies the viral titres in waste pork and pork products in relation to the oral infective dose for each disease.

The risk assessment inputs of incidence or prevalence of disease, duration of infection/viraemia, swine population demographics, detection systems (in the herd, ante- and post-mortem inspections), surveillance and monitoring programmes, disease control and eradication programmes and exposure are more clearly elaborated on an individual country basis and are not discussed in this paper.

Hazard identification

The potential hazards associated with trade in pork and pork products are the viral agents of foot and mouth disease (FMD), classical swine fever (CSF) (hog cholera), African swine fever (ASF), and swine vesicular disease (SVD). The viral agents of transmissible gastroenteritis of pigs (TGE) and porcine reproductive and respiratory syndrome (PRRS), although perceived as potential hazards by some countries, are discussed to illustrate the differences between these two agents and actual hazards of pork and pork products. Excluded from the discussion is the parasitic agent of trichinellosis. Although the agent of this disease represents a potential animal health hazard of pork and pork products, the disease is principally a food safety concern that would manifest itself in humans much earlier and with a much greater impact than in pigs.

In the past and recently, pork and pork products have been incriminated as sources of disease introduction for all four hazards. The disease introduction often occurs as outbreaks in herds which are fed raw food waste in their swill, despite regulations that may be in place requiring the cooking of such waste before feeding. However, disease introduction of PRRS and TGE has never been attributed to the importation of pork and pork products from infected zones.

Many primary outbreaks of FMD have been attributed to imported meat and meat products, although pork and pork products were not necessarily implicated (6, 92).

Outbreaks of CSF have often been traced to the feeding of garbage from ships or aircraft. Notable examples include two occasions, in 1930 and 1953, when the disease was introduced into New Zealand (47). The feeding of swill, presumably containing uncooked waste pork and pork products, to pigs was incriminated in CSF outbreaks in the United Kingdom in 1986 (98), in Switzerland in 1993, in Bulgaria (65), Germany (66) and Poland (67) in 1994 and in Austria in 1994 (64) and 1995 (68).

Of 518 SVD outbreaks which occurred in Great Britain between 1972 and 1981, 80 were attributed to feeding of contaminated waste food (38).

There is no evidence of oral transmission of PRRS through uncooked waste of pork or pork products either in an enzootically infected zone or in the spread of the disease to a previously disease-free zone (5, 24, 61, 78). Experimental oral transmission is possible following administration of doses of $10^7$ TCID$_{50}$ virus (55).

Similarly, the transmission of TGE through the feeding of waste pork or pork products has never been reported. Only experimental oral infection of one-week-old piglets with homogenates of muscle of the hind leg, lymph nodes (internal iliac, sub-maxillary and cervical) and bone marrow from the femur of 16 six-month-old pigs which had been in contact with experimentally infected piglets has been demonstrated. Twelve three-week-old pigs fed about 1.5 kg of the same tissues each over 5 days failed to become infected, yet all became serologically positive for neutralizing antibody to TGE virus (27). Cook et al. (13) achieved transmission of TGE in six-day-old piglets by oral dosing daily for 4 days with a homogenate of brachiocephalic muscle and parotid lymph node. These two studies involved dosing with a rather artificial mixture considering the homogenisation of the tissues and the high proportion of lymph node tissue.

Inputs for risk assessments on the importation of pork and pork products

Agent pH and temperature lability

The pH and temperature lability of the viral agents represent the two most important biological properties for an assessment on the safety of importing meat. The anaerobic glycolysis which ensues in muscle after slaughter results in the conversion of glycogen to lactic acid. In one study on meat quality in different swine breeds, the pH of the semimembranosus and longissimus dorsi muscles at 45 minutes post mortem ranged from a low of 6.17 to a high of 6.71 (88). The final pH attained is generally about 5.4-5.5.
in typical mammalian muscles and is referred to as the ‘ultimate pH’. Between individual pigs there is a considerable variation in the time taken for the pH of latissimus dorsi muscle to fall from a pH of 6.5 to an ultimate pH of 5.5 at 37°C, a range of about 150 to just over 400 minutes (45). In the commercial production of pork, the time and temperatures at which the carcasses are held seldom allow the pork to fall below a pH of 5.7 (10). FMD virus is the only hazard which is labile to pH levels found in pork: however, viral inactivation as seen in the maturation of beef may not be as dramatic. Beef matured for 20-24 hours safely achieves a pH of 5.7 (3) and any virus present in the muscle tissues would be readily inactivated. The significance of this is that the pH of pork and pork products represents an important input in the risk assessment on pork from FMD infected zones. Other organs and tissues such as the parenchymatous organs, lymph nodes and bone marrow would be unlikely to achieve the low pH values as evidenced in muscle.

Temperature lability is the most important consideration for pork products which undergo some thermal processing. Although there is a difference between the temperature lability of the viral agents in culture media and that in pork, the laboratory data give some indication of the ease of inactivation. Generally the studies involving thermal processing of pork in which particular core temperatures achieve an apparent inactivation would be better to assess. For the four viral hazards, a temperature of 69°C which is reached by the very inner core of a portion of pork appears to readily inactivate the virus. A minimal time period of less than 15 minutes at this temperature, however, may be necessary to inactivate CSF virus in pork.

**Target organs**

Before discussing the target organs and predilection sites of the viral agents, it is necessary to make a clear distinction between the organs and tissues which constitute pork and pork products and non-pork organs. Pork and pork products are comprised principally of skeletal muscle, bone and fat. Bone marrow, blood within the capillaries of skeletal muscles and lymph nodes (prepectoral, presternal, precrural, superficial inguinal, popliteal, iliac, lumbar and renal) (32) amount to a very small fraction of the swine carcass. With respect to pork portions, lymph nodes and bone marrow may not be present as a result of trimming, deboning and depending on the particular cut. Many of the lymph nodes indicated above are removed through carcass trimming due to their superficial, fat-embedded location on the carcass. Obviously, the blood, the respiratory, gastro-intestinal and reproductive tracts, the head, the respective lymph nodes of these parts and the tonsils are not pork tissues.

The cell and tissues for which the agent has an affinity and in which the agent replicates are therefore an important consideration in the importation of pork. Virus titres in pork derived from infected pigs may be very low in comparison to the predilection sites of the agent (e.g. SVD virus replicates in the epithelium of the coronary band, tongue, snout and lips and in the myocardium, tonsils and brain stem (90). During the incubation period, the virus may only be found at the site of entry and the adjacent lymph nodes, such as the tonsils and mandibular and parotid lymph nodes for CSF, ASF and SVD or the popliteal lymph nodes of the hind legs for SVD following agent entry through broken skin on the foot. Despite this tissue tropism, titres are detected in almost all tissues during the state of viraemia. Viral titres in skeletal muscle of viraemic pigs with SVD or FMD may represent more a function of the viral titre within blood capillaries. Viral titres in skeletal muscle of pigs during the viraemic phase of ASF are a reflection of high blood titres, the target cells (lymphocytes, endothelial cells, reticular cells, monocytes and macrophages) (76) and loss in vascular integrity. Highly vascularised organs such as the spleen, kidney, and liver and bone marrow would be expected to exhibit viral titres during the state of viraemia. Generally, only the viraemic period and its duration are of significance in the importation of pork. The duration of viraemia may be quite short, as observed with SVD, or prolonged, as in some of the clinical forms of CSF and ASF. The mortality as seen in the peracute, acute and subacute forms of CSF and ASF naturally limits the viraemic period.

A carrier state is noted with ASF but not with CSF, SVD or FMD in pigs. The carrier state for diseases such as TGE and PRRS is characterised by predilection and maintenance of the virus in tissues other than pork, that is the tonsils, lungs and intestinal tract for TGE and the lung and tonsils for PRRS. The carrier state of ASF in surviving pigs is confined to lymph nodes, tonsils and to a lesser extent kidney, spleen and bone marrow (53).

**Agent survival in pork and pork products**

Survival in the product depends on the agent properties, especially lability to time, temperature and pH and on the inherent properties of the product (e.g. pH, water activity, moisture:protein ratio, temperature of processing and storage, salinity, additives, etc.). The interaction of these product properties and the enzymatic proteolysis which occurs in many products are significant factors in the inactivation of viral agents. Survival of agents in various pork products has been investigated and reported in the scientific literature. Most of these studies must be statistically interpreted, based on the number of samples tested for virus isolation or the data points on the reduction of viral titre of an agent over time. Multiple studies and other information may be combined and statistically evaluated to estimate the inactivation curve associated with some form of processing. Agent survival beyond the usual processing time and the time required for importation and distribution in the importing country are part of the considerations of this input. Changes in the technology of meat processing and curing, and the efficiency of modern abattoir practices and international transportation and distribution, must be considered here.
**Agent quantification**

**Viral titres**

Data on titres in tissues, in particular muscle, quantify the amount of agent present per gram. Quantification of viral titres is generally obtained from experimental infection of swine with a specific strain of virus. The viral titres in muscle and other tissues are measured, often at the peak of viraemia. Titres at other times in the disease course are not available, as experimental studies are frequently looking for the 'worst-case scenario' when titres are expected to be at their highest level. Employing these data to represent the titres for any day of the duration of infection or viraemia is an extrapolation. Nonetheless, the data may represent the best available biological information.

**Oral infective dose**

Uncooked waste of pork or pork products being ingested by swine by one or more scenarios represents the only mode of transmission of interest here. The target species is the pig, the portals of entry are the tonsils, oral abrasions, respiratory tract and the gastro-intestinal tract. After contact exposure, ASF virus primarily gains entry through the tonsils and in some instances across the nasal, bronchial and (possibly) gastric mucosae (34). Similarly with exposure to pigs infected with SVD, the tonsils appear to be the main site of entry, however, other entry sites may include the skin of the head and lower limbs, upper respiratory and digestive tract epithelium, and intestinal mucosae (56). Virus in pork or pork products has to contact the tonsils, the upper respiratory tract epithelium or gain entry through oral abrasions. Failing these routes of entry, the gastro-intestinal mucosae may serve as sites of entry providing the virus survives the low pH of the gastric juices. The amount of mastication of the food and the rapidity of swallowing a bolus of pork would determine the site of viral entry. It is for this reason that the oral pig infective dose 50% (IPID$_{50}$) for the viral diseases would be expected to be considerably higher for feeding of infected pork than for instilling virus in culture media on the tongue of a pig, feeding the virus suspended in a liquid medium such as milk or force-feeding homogenised infected tissues. The IPID$_{50}$ has not been estimated for FMD, CSF, and SVD. Instead, an infective dose at which infection was achieved experimentally in one or more pigs is available in the literature. In the case of SVD, skin abrasions, especially around the coronary bands of the feet may serve as a portal of entry following contact with infected waste pork (56).

**Foot and mouth disease**

**Agent pH and temperature lability**

FMD virus is acid, alkali and heat labile, but can survive for long periods at neutral pH (7 to 9) and under low temperature conditions (82). In tissue culture suspensions, the virus survived less than 15 seconds at pH 2.2 and pH 4, two minutes at pH 6 and for several weeks at pH 7 (25, 85). At a pH of 7.5, the virus in simple media survived 30 seconds at 61°C, two minutes at 55°C yet as long as 18 weeks at 4°C (1).

**Target cells and tissues**

The virus is distributed throughout the body of the infected animal and can be found in different concentrations for varying periods in the tissues. In pigs, the greatest quantities of virus are in the blood, epithelium, and liver (23, 86). The specific lesions in their early stages are microscopic and are limited to the epithelium at sites of predilection such as the mucosa of the mouth, including the tongue, lips, gums, pharynx, and palate (9).

The incubation period of FMD is generally within the range of 2-14 days (82). The virus is excreted one to ten days before clinical signs appear, and continues for four to ten days. Virus was detected in blood 32 hours and in muscle 20 hours before the appearance of aphthae or the beginning of a rise in temperature (21). In pigs, the virus does not persist for more than a month (57). One characteristic which distinguishes pigs from cattle and sheep is that they appear to harbour the virus only during clinical stages of the disease and therefore do not act as carriers (15).

**Survival in pork and pork products**

The apparent duration of survival of FMD virus in the listed pork and pork products is as follows:

- 30 days in different chilled organs such as lungs, stomach, tongue, intestine (83)
- 24 hours in chilled spleen, liver and kidney (83)
- 210 days in frozen lungs, intestine, stomach, tongue, kidney, spleen and liver (83)
- 170 days in Parma hams (52)
- 182 days in the white Serrano ham (58)
- 168 days in Iberian ham (58)
- 112 days in Iberian shoulder hams (58)
- 42 days in Iberian loins (58)
- 190 days in salted bacon, and 183 days in ham fat (22)
- 56 days in sausages (22)
- 250 days in processed intestinal casings (49)
- 7 days in salami (71)
- 10 days in tongue and 1 day in muscle (14).

Apparent thermal inactivation of FMD virus is obtained with an internal temperature of 69°C (50).

**Agent quantification**

**Viral titres**

Sellers (86) found high titres of $10^{7.2}$ plaque forming units (PFU)/ml in blood, $10^{6.6}$ PFU/g in bone marrow and
10\(^{5.6}\) PFU/g in liver of infected pigs. Similarly high virus titres of greater than 10\(^{5}\) PFU/ml were detected by extraction from both fat and muscle tissues of infected pigs (71). Viral titres were measured in each of 62 pigs two days after intravenous inoculation with 1 ml of a 1:10 dilution of a stock virus (serotype C) having a titre of 10\(^{8.9}\) TCID\(_{50}\) per ml. The mean and standard deviation of the viral titres in muscle, fat, bone marrow, blood and lymph node are presented in Table I (58). The mean viral titres in muscle and fat as presented in Table I are significantly lower than the titres detected by Panina et al. (71).

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Titre (plaque forming units [PFU] per ml or g)</th>
<th>Mean</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood</td>
<td>(10^{5.8})</td>
<td>(10^{1.6})</td>
<td></td>
</tr>
<tr>
<td>Lymph node</td>
<td>(10^{1.4})</td>
<td>(10^{1.7})</td>
<td></td>
</tr>
<tr>
<td>Bone marrow</td>
<td>(10^{1.8})</td>
<td>(10^{1.5})</td>
<td></td>
</tr>
<tr>
<td>Fat</td>
<td>(10^{5.5})</td>
<td>(10^{1.6})</td>
<td></td>
</tr>
<tr>
<td>Muscle</td>
<td>(10^{6.01})</td>
<td>(10^{0.2})</td>
<td></td>
</tr>
</tbody>
</table>

**Oral infective dose**

A viral titre of \(10^{5.0}\) TCID\(_{50}\) of FMD O-strain initiated infection in two of 30 pigs fed minced offal (liver, kidney and lymph nodes) (39).

**African swine fever**

**Agent pH and temperature lability**

The sensitivity of the ASF virus to different temperatures has been studied widely. Infected blood heated for 30 minutes at 60°C loses infectivity. At 56°C, the resistance of the virus will depend on the presence of serum. Some strains could remain virulent after 3.5 hours at 56°C (12). Kovalenko et al. (44) found that the virus could survive 6 years at 5°C with no light. ASF virus is very resistant to acid pH, even more than CSF virus. Plowright et al. (74) have found that the ASF virus could survive 22 hours at pH 3.1. The virus is rapidly inactivated at pH 11.5 and the inactivation rate decreases rapidly at lower pH values (12).

**Target cells and tissues**

The ASF virus usually enters the pig through the mouth or upper respiratory system, and infection is established in the nasopharyngeal region. The virus spreads rapidly to the mandibular lymph nodes and throughout the body through lymph and blood (46). Following the incubation period, the duration of viraemia may last 1-3 days, 4-8 days, and 6-10 days before death in the hyperacute, acute and subacute forms of the disease, respectively (70). With one ASF viral strain, viraemia was demonstrated two days before the onset of fever (34). Pigs surviving the subacute form may exhibit the chronic form of the disease which may last for several months. The carrier state in these chronically affected pigs and in pigs manifesting a subclinical or inapparent form of the disease may be confined to lymph nodes, tonsils and to a lesser extent kidney, spleen and bone marrow (53, 70, 97). The virus replicates principally in the cells of the lymphoreticular system, both the fixed cells in the lymph nodes, liver and spleen (58, 62); and in monocytes and macrophages (97). Endothelial cells lining blood vessels are severely damaged, with resulting oedema, haemorrhage and necrosis. Lesions are most prominent in the spleen, lymph nodes, lung and liver, but may be found in any lymphoid tissue or infiltrate of lymphoid cells (46).

**Survival in pork and pork products**

The apparent duration of survival of ASF virus in the listed pork and pork products is as follows:
- 104 days in frozen meat or chilled meat (44)
- 140 days in Iberian hams (58)
- 140 days in Iberian shoulder hams (58)
- 112 days in Iberian loins (58)
- 140 days in white Serrano hams (58)
- 399 days in Parma hams (52)
- 30 days in pepperoni sausage (49)
- 30 days in salami sausage (49)

Apparent thermal inactivation of ASF virus is obtained with an internal temperature of 69°C (50).

**Agent quantification**

**Viral titres**

Viral titres of meat samples from four pigs infected with ASF ranged from \(10^{5.25}\) to \(10^{5.75}\) haemadsorbing units 50% (HAd\(_{50}\)) per g in both whole and ground meat, two days after slaughter; \(10^{2.8}-10^{2.5}\) in salami, three days after slaughter; \(10^{2.0}-10^{2.25}\) in pepperoni, three days after slaughter; \(10^{2.5}-10^{2.75}\) in brined ham, two days after slaughter; \(10^{2.75}-10^{3}\) in pepperoni sausage, eight days after slaughter; and \(10^{-1}\) in salami sausage, nine days after slaughter (49). Viral titres were measured in each of 65 pigs five days after intramuscular inoculation of 1 ml of undiluted stock virus having a titre of \(10^{8.5}\) HAd\(_{50}\) per ml (58). The mean and standard deviation of the viral titres in muscle, fat, bone marrow, blood and lymph node are presented in Table II.

**Oral infective dose**

In one experimental study, the PID\(_{50}\) was \(10^{5.4}\) HAd\(_{50}\) (34). McVicar (53) found the PID\(_{50}\) of a moderately virulent strain of ASF virus to be \(10^{4.3}\) HAd\(_{50}\) by oral experimental infection. Experiments were conducted in which lymph node suspensions from naturally infected warthogs were administered to domestic swine. The results of these
African swine fever viral titres in tissues of 65 pigs five days after experimental infection (58)

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Titre (haemadsorbing units 50% [HAd50] per ml or g)</th>
<th>Mean</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood</td>
<td>$10^{9.3}$</td>
<td>$10^{9.2}$</td>
<td></td>
</tr>
<tr>
<td>Lymph node</td>
<td>$10^{9.5}$</td>
<td>$10^{9.1}$</td>
<td></td>
</tr>
<tr>
<td>Bone marrow</td>
<td>$10^{9.4}$</td>
<td>$10^{9.3}$</td>
<td></td>
</tr>
<tr>
<td>Fat</td>
<td>$10^{9.5}$</td>
<td>$10^{9.3}$</td>
<td></td>
</tr>
<tr>
<td>Muscle</td>
<td>$10^{9.3}$</td>
<td>$10^{9.3}$</td>
<td></td>
</tr>
</tbody>
</table>

experiments made it evident that well-homogenised tissue suspension, containing $10^{3.7}$ to $10^{6.1}$ HAd50 of ASF virus failed to infect pigs when administered either in liquid or moistened solid food (75).

Classical swine fever (hog cholera)

Agent pH and temperature lability

While the virus is very resistant at temperatures below 0°C, research has shown the sensitivity of the virus above this temperature. It can survive three days at 50°C, seven to 15 days at 37°C and years at -70°C. The virus is, however, susceptible to rapid changes in temperature such as thawing and refreezing. The effect of heat treatment on CSF virus is influenced by the physical medium in which the virus is heated. Thus cell culture fluid infectivity is lost after 30 minutes at 60°C, whereas in defibrinated blood the virus is not inactivated after 30 minutes at 68°C (94). The virus is stable over a wide pH range, but is rapidly inactivated below pH 4 and above pH 11 (89).

Target cells and tissues

The virus initially infects epithelial cells of the tonsillar crypts and subsequently spreads to the surrounding lymphoreticular tissue. From the tonsil, CSF virus is drained to the regional lymph nodes, where replication occurs. The virus reaches lymph nodes ranged from $10^{7}$ to $10^{10}$ TCID50/g and titres in muscle ranged from $10^{9.3}$ to $10^{10}$ TCID50/g. Titres in lymph nodes ranged from $10^{9.0}$ to $10^{7.5}$ TCID50/g. Titres

the viraemia persists at a high level until death which may occur between 10 and 20 days post-infection for the acute form and between 20 and 29 days post infection for the subacute form. In the chronic form, the duration of the disease could be 30 or more days (18). In the chronic form, the viraemia may subside during the course of the infection whereas viraemia persists at a high level for life in the late-onset form of CSF (94, 96). The late-onset disease, a sequel of congenital CSF virus infection, is characterised by a period of a few months during which pigs remain free of disease. Most pigs survive for more than 6 months but all eventually die. The viraemia persists for life (93). CSF virus strains of low virulence can induce mild disease with subsequent recovery without a carrier state (89). Virulent virus infects both epithelial and reticular cells, macrophages and tonsillar cells, whereas the growth of virus of reduced virulence is mainly restricted to cells of the epithelial tonsillar crypts (93).

Survival in pork and pork products

The apparent duration of survival of CSF virus in the listed pork and pork products is as follows:

- 4.5 years in frozen meat (96)
- 1 month in the meat of salt-cured pork (96)
- 2 months in the bone marrow of salt-cured pork (96)
- 90 days in salami (84)
- 75 days in Italian salami (72)
- 90 days in ham (muscle and fat) (84)
- 70 days in neck, lard and bone marrow (84)
- 226 days in frozen liver at -4°C and -6°C (12)
- 252 days in Iberian hams (58)
- 40 days in Iberian shoulder hams (58)
- 126 days in Iberian loins (58)
- 140 days in White Serrano hams (58)
- 189 days in Parma hams (52)
- 147 days in intestinal casings processed in water at 42.2°C for 30 minutes (50).

Apparent thermal inactivation of CSF virus is obtained using the following procedures:

- pasteurisation at core temperatures over 67°C of cured and canned hams (89)
- exposure of cubes (2 cm³) of ham to a ‘flash’ temperature of 71°C for 1 min (87)
- heating to 69°C for 15 min (50).

Agent quantification

Viral titres

Wood et al. (99) studied the titres of virus in tissues of pigs experimentally inoculated with CSF and slaughtered between seven and 25 days after infection. Pigs were infected by intranasal inoculation with $10^{6.5}$ TCID50/pig. CSF virus titres in muscle ranged from $10^{5.8}$ to $10^{6.9}$ TCID50/g and titres in lymph nodes ranged from $10^{5.0}$ to $10^{7.5}$ TCID50/g. Titles
Viral titres were measured in each of 64 pigs five days after intravenous inoculation with 1 ml of a 1:100 dilution of a stock virus having a titre of $10^{5.5}$ TCID$_{50}$ per ml (58). The mean and standard deviation of the viral titres in muscle, fat, bone marrow, blood and lymph node are presented in Table III.

### Table III

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Titre (plaque forming units [PFU] per ml or g)</th>
<th>Mean</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood</td>
<td>$10^{3.9}$</td>
<td></td>
<td>$10^{3.5}$</td>
</tr>
<tr>
<td>Lymph node</td>
<td>$10^{3.9}$</td>
<td></td>
<td>$10^{2.2}$</td>
</tr>
<tr>
<td>Bone marrow</td>
<td>$10^{3.8}$</td>
<td></td>
<td>$10^{2.7}$</td>
</tr>
<tr>
<td>Fat</td>
<td>$10^{3.0}$</td>
<td></td>
<td>$10^{1.0}$</td>
</tr>
<tr>
<td>Muscle</td>
<td>$10^{3.0}$</td>
<td></td>
<td>$10^{0.5}$</td>
</tr>
</tbody>
</table>

### Oral infective dose

A CSF virus titration in weaner pigs using the highly virulent strain 'Alfort' showed that the minimal infective dose resulting in fatal disease was less than $10^{3.5}$ TCID$_{50}$ (18). In comparison to the three other viral hazards, this represents a very low oral infective dose. It may indicate that an infective dose for pork and pork products is likewise comparatively very low.

### Swine vesicular disease

#### Agent pH and temperature lability

SVD virus is relatively stable over a pH range of 2-12, depending on temperature and time (38): 164 days at a pH of 5.10 and 7.54 at 5°C, but a reduction of over 6 log in titre occurred at pH values of 2.88 and 10.14 by 164 days and at pH values of 1.92 and 11.96 at 38 days (40). SVD virus is also stable in infected tissue kept at ambient or higher temperatures for four months or more (90). Carcass materials at –20°C were sampled after approximately 11 months and showed no significant drop in infectivity resulting from storage (20).

### Target cells and tissues

Swine vesicular disease virus has an affinity for, and replicates in, the epithelium of the coronary band, tongue, snout and lips, as well as the myocardium, the tonsils and the brain stem (90). The stratum spinosum is the primary site of viral replication in the epithelium (41). In cell cultures of epithelial and salivary gland tissues, SVD virus reached significant titres whereas little or no growth was detected in cell cultures of muscle, lymph node or parts of the digestive tract (56).

Eight healthy pigs were inoculated in the coronary band of the hind legs with cell culture SVD virus and were slaughtered at the peak temperature. Samples of different tissues were collected during necropsy. The virus could not be isolated from the muscles, but was detected in one fat sample. Tests were performed on fresh samples and at 12, 30, 40, 70 and 100 days after storage at 4°C (28).

The peak of virusaemia occurs 2-4 days post-exposure to SVD virus and persists for six days. Virus persists for at least 10 days in the tissues of the snout, tongue, coronary band, tonsil, cardiac muscle, and central nervous system. Following experimental infection, swine shed virus in their faeces for up to 23 days (41).

#### Survival in pork and pork products

The apparent duration of survival of SVD virus in the listed pork and pork products is as follows:
- 300 days in Parma hams (51)
- 200 days in dry salami sausage, dry pepperoni sausage and intestinal casings (48)
- 400 days in dried pepperoni and salami sausage (33)
- 780 days in processed intestinal casings (33)
- 40 days in salami and pepperoni sausages (38)
- 509 days in unprocessed intestinal casings (38)
- 28 days in Iberian loins (59)
- 112 days in Iberian shoulder hams (59)
- 560 days in Iberian hams (59)
- 539 days in white Serrano hams (59).

Apparent thermal inactivation of SVD virus is obtained by heating to at least 69°C (48, 50).

#### Agent quantification

**Viral titres**

Dawe (20) reported that 11-month-old frozen carcass material had $10^6$ TCID$_{50}$ per g of SVD virus in the skin, $10^4$ TCID$_{50}$ in intercostal muscle and $10^3$ TCID$_{50}$ per g in
both rib bone and kidney cortex. These virus titres were not significantly below titres from similar samples taken at the beginning of the storage. In hams held at 0-4°C for 72 hours after slaughter, mean titres, expressed as PFU per g of muscle, bone marrow and fat were $10^{4.5}$, $10^{2.3}$ and $10^{1.1}$, respectively.

Viral titres were measured in each of 64 pigs three days after intravenous inoculation with 3 ml of a 1:3 dilution of a stock virus having a titre of $4.5 \times 10^7$ PFU per ml (59). The mean and standard deviation of the viral titres in muscle, fat, bone marrow, blood and lymph node are presented in Table IV.

### Table IV

**Swine vesicular disease viral titres in tissues of 64 pigs three days after experimental infection** (58)

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Titre (plaque forming units [PFU] per ml or g)</th>
<th>Mean</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood</td>
<td>$10^2.8$</td>
<td></td>
<td>$10^1.1$</td>
</tr>
<tr>
<td>Lymph node</td>
<td>$10^2.2$</td>
<td></td>
<td>$10^1.2$</td>
</tr>
<tr>
<td>Bone marrow</td>
<td>$10^1.3$</td>
<td></td>
<td>$10^1.5$</td>
</tr>
<tr>
<td>Fat</td>
<td>$10^1.3$</td>
<td></td>
<td>$10^1.8$</td>
</tr>
<tr>
<td>Muscle</td>
<td>$10^3.2$</td>
<td></td>
<td>$10^3.8$</td>
</tr>
</tbody>
</table>

### Oral infective dose

A dose of $10^{6.8}$ PFU produced clinical signs within 2 to 7 days in three of the six pigs infected orally (instillation in the mouth), while the dose required to produce infection when the amount of virus was applied to abraded skin surfaces was $10^{3.6}$ PFU (56).

McKercher et al. (48) fed swine with infected meat which had a titre of $10^3$ to $10^4.5$ TCID$_{50}$ per gram. In some tests, the swine became infected when fed 60 g of infected meat and in other instances when fed 900 g. In one feeding test, the animals did not show any clinical manifestations of SVD; however, five of 18 developed neutralising antibody; three of these five were contact swine. Six feeding tests involving 39 swine were used to indicate that swine may become infected when fed products from meat from infected animals. However, in these tests, only 12 of the 39 swine showed clinical evidence of SVD.

### Porcine reproductive and respiratory syndrome

#### Agent pH and temperature lability

The PRRS virus has been classified an arterivirus (24, 26). The virus is sensitive to pH: infectivity titres are reduced by over 90% below pH 5 or above pH 7 (61). Bloemraad et al. (7) illustrated the short half-life time of the PRRS virus at pH 5.0: 0.65 hours at 37°C and 18.8 hours at 4°C. Survival time of PRRS virus in plasma is believed to be less than two days. At 70°C the virus retains infectivity for 18 months; at 4°C, for 1 month; at 37°C, 50% reduction occurs after 12 hours and complete inactivation after 48 hours and at 56°C, complete inactivation after 45 minutes (61).

#### Target cells and tissues

The porcine reproductive and respiratory syndrome is a multi-system disease characterised initially by viraemia with subsequent virus distribution and replication in multiple organs causing interstitial pneumonia, vasculitis, lymphadenopathy, myocarditis and encephalitis (79). The duration of viraemia as either plasma- or cell-associated viraemia has been portrayed experimentally by Mengeling et al. (60). The duration of viraemia in experimentally infected six-week-old pigs varied from two days to under 35 days. The duration of viraemia portrayed above is supported in part by other studies (11). The lymphoid and respiratory systems have the most remarkable lesions and appear to be the major sites of replication of PRRS virus (35). A tropism for vascular tissues is assumed from the lesions of vasculitis of the medium sized arteries and occasionally of the veins, not only in the lungs but also in other organs, including the meninges and the brain where a perivascular lymphoplasmacytic infiltrate has been described (19). Persistence of the virus occurs mainly in alveolar macrophages and in the oropharyngeal lymphoid tissue. Viral persistence has been detected as long as 157 days after initial challenge (4, 5, 31).

### Survival in pork and pork products

There are no studies on the survival of PRRS virus in pork and pork products.

#### Agent quantification

#### Viral titres

Bloemraad et al. (7) intranasally exposed four 6-month-old SPF pigs to $10^{5.0}$ TCID$_{50}$ of the PRRS virus. Virus was detected in the semimembranosus muscle in the two pigs killed five days post-exposure, at 24 hours post mortem (a titre of $10^{4.9}$ TCID$_{50}$ in one of the pigs) but not at 48 hours. No virus was detected in any of three samples representing 0, 24 and 48 hours post mortem from neck musculature, longissimus dorsi and bone marrow (a total of 9 samples). In the two pigs killed at 10 days post-infection, only one of the 24 samples from neck musculature, longissimus dorsi, semimembranosus and bone marrow at 0, 24 and 48 hours post mortem revealed virus. The virus positive sample was obtained from the neck musculature at 24 hours post mortem. Titres were obtained in certain viscera and lymph nodes and in serum samples at 0, 24 and 48 hours post mortem. The authors add that the sporadic recovery of virus from muscle tissue most likely originated from blood plasma.
and that the titres in muscle diminished after slaughter during the 12 hour cooling period, probably due to a decrease in the pH of the muscle to a pH of between 5 and 6.

Mengeling et al. (60) isolated virus only from ham and bone marrow of one pig but not from 30 samples of bone marrow, intercostal muscle, loin, shoulder, and rump from six other viraemic pigs. The six-week-old pigs were experimentally infected through oronasal inoculation with 2 ml of 10^6 median cell culture infective dose 50% (MCID_{50})/ml of PRRS virus.

Magar et al. (54) could not detect PRRS virus or antigens in either lymph nodes or muscle of two experimentally infected slaughter-age (6 months old) pigs 14 days post-exposure. At 7 days post-exposure in two pigs, virus was detected in these tissues. The pigs had been inoculated intranasally and orally with 10^{5.5} TCID_{50} and 10^{5.3} TCID_{50} of PRRS virus per pig. Samples from lung and tonsils revealed PRRS virus at both 7 and 14 days post-exposure. The muscle tissues included a pool of samples from neck, chest and pelvic region and a pool of samples from longissimus dorsi and semimembranosus muscles.

**Oral infectious dose**

A minimum oral infectious dose or an oral PID_{50} has not been reported for PRRS.

**Transmissible gastroenteritis of pigs**

**Agent pH and temperature lability**

Transmissible gastroenteritis virus is very stable when stored frozen. Storage of the virus in cell cultures at -20°C, -40°C, and -80°C for 365 days did not result in any significant drop in titre. However, storage at 37°C for four days eliminated infectivity. Similarly, virus of pig intestine origin held at 37°C exhibited one log_{10} reduction in viral titre every 24 hours. Field strains of TGE virus are stable at pH 3 (36).

**Target cells and tissues**

Ingestion is the most common route of virus entry, although nasal and airborne infections can occur (8). After ingestion, the virus passes unharmed through the stomach and replicates in the enterocytes of microvilli throughout the small intestine, resulting in rapid and massive epithelial cell sloughing, marked villous shortening or atrophy, leading to malabsorption and diarrhoea (73). The pathogenicity and the sites of viral multiplication were compared for three strains of TGE virus, administered orally to one-week-old weaned piglets. Two of the strains multiplied within the intestinal tract in the enterocytes of the jejunum and ileum, Peyer's patches and mesenteric lymph nodes. In view of the small numbers of infected cells in the tonsils, spleen, kidney, liver and lung, these tissues were not considered to be preferential multiplication sites. An attenuated strain replicated only in the ileum and the mesenteric lymph nodes. The variation in the tropism for particular parts of the intestine (with the preferential localisation of the virus in the ileum rather than the jejunum), could be related to the high degree of attenuation (17).

TGE virus has been transmitted by homogenates of kidney, spleen, liver, lungs and brain, as well as gastrointestinal tract of young pigs (2). In pigs aged 12 hours to 6 days, which were orally infected with TGE virus or were allowed to come in contact with these inoculated pigs, the virus was recovered from the digestive tract, nasal and tracheal membranes, lung and lymph nodes draining the affected organs. Virus was recovered from the parenchymatous organs such as liver, spleen, kidney, brain and blood (29, 37). In older pigs of 4-5 months of age inoculated orally or intratracheally, virus was recovered from the same tissues but not from the parenchymatous organs or blood (37). No virus was isolated from blood samples taken daily from 16 six-month-old pigs in contact with TGE infected piglets. TGE virus was detected in intestinal samples but not in pharyngeal swabs, muscle of the hind leg, lymph node or bone marrow samples (27).

TGE virus has an incubation period as short as 12 to 18 h (30) and up to three days (81). In growing and finishing pigs, a watery diarrhoea developed and lasted 5-7 days (73). Pigs infected with TGE virus may become convalescent carriers, the virus being isolated from intestinal and respiratory tissues for up to 104 days post-exposure (91). However, long-term shedding of viable virus and the role of TGE virus carriers in transmitting the disease have not been fully assessed (80). Long-term carrier pigs may be the exception rather than the rule (73). Viraemia does not appear to be a component of the pathogenesis of TGE in the growing and finishing pig (27, 37). Although the virus was recovered from the milk of sows during the acute stage of infection, these sows had been intravenously, intranasally, or intramammary inoculated with high titres of the virus (43).

**Survival in pork and pork products**

There are no studies on the survival of TGE virus in pork and pork products, presumably because the viral agent is not found in pork tissues of TGE-infected slaughter age pigs.

**Agent quantification**

**Viral titres**

Viral titres in pork tissues of slaughter age pigs do not exist since a viraemic phase of TGE does not occur in this age of pigs.
Oral infective dose

A minimum oral infective dose or an oral PID$_{50}$ has not been reported for TGE, other than a significant difference in the virus titre required to infect slaughter age pigs versus neonatal pigs.

Conclusions

The potential animal health hazards associated with the importation of pork and pork products, excepting fully cooked products, are FMD, ASF, CSF and SVD. PRRS and TGE viruses are not hazards of these commodities. The importation of any pork and pork products from zones infected with one of these hazards necessitates a risk assessment to ensure the animal health security of the importing country. Commodity related inputs which are required for the risk assessment include: the agent pH and temperature lability, target organs, agent survival in pork and pork products, and the quantification of the agent as to viral titres in relation to a minimum infective dose.

Risques zoosanitaires potentiels liés à la viande de porc et aux produits de charcuterie

S. Farez & R.S. Morley

Résumé

Les risques zoosanitaires liés à l’importation de viande de porc et de produits de charcuterie concernent quatre virus : ceux de la fièvre aphteuse, de la peste porcine classique, de la peste porcine africaine et de la maladie vésiculeuse du porc. Seule l’évaluation des risques permet de déterminer correctement l’innocuité des importations de viande de porc à partir d’une zone infectée par une ou plusieurs de ces maladies. Cela vaut également pour l’importation de produits dérivés du porc ayant subi un certain degré de transformation (abstraction faite de la charcuterie cuite). Les auteurs analysent, pour chaque maladie, la sensibilité de l’agent responsable (aux variations de pH et de température), les organes cibles, la persistance de l’agent dans la viande de porc et la charcuterie ainsi qu’une évolution quantifiée de la présence de cet agent. Cette évaluation est une donnée de l’appréciation du risque qui consiste à mesurer le titre de virus présent dans les déchets de viande de porc et de charcuterie par rapport à la dose infectieuse orale estimée pour chaque maladie. Les auteurs présentent également deux autres maladies virales du porc, la gastro-entérite transmissible et le syndrome dysgénésique et respiratoire, et expliquent pourquoi elles ne présentent pas de risque pour la viande de porc et la charcuterie.

Mots-clés
Potenciales riesgos zoosanitarios asociados a la carne de cerdo y embutidos

S. Farez & R.S. Morley

Resumen
Cuatro son los agentes víricos que representan los principales riesgos zoosanitarios asociados a la importación de carne de cerdo y embutidos, a saber, los virus de la fiebre aftosa, la peste porcina clásica, la peste porcina africana y la enfermedad vesicular porcina. La evaluación de riesgos constituye el único modo correcto de determinar el nivel de seguridad que ofrece la importación de productos porcinos procedentes de una zona afectada por una o varias de esas enfermedades. Esto también se aplica a la importación de productos porcinos previamente sometidos a alguna forma de tratamiento (excluidos los embutidos cocinados). Para cada una de las enfermedades antedichas, los autores examinan los órganos diana, la labilidad del agente en función del pH y la temperatura, su persistencia en el cerdo y embutidos y por último su cuantificación. La cuantificación del patógeno es un dato de la evaluación de riesgos que calcula la relación entre el título vírico presente en los restos de cerdos y productos porcinos y la dosis infecciosa por vía oral que se estima para cada enfermedad. Se exponen también los casos de otras dos enfermedades porcinas de origen vírico, la gastroenteritis transmisible y el síndrome disgenésico y respiratorio porcino, con objeto de ilustrar por qué estas dos enfermedades no entrañan riesgo alguno ligado a la carne de cerdo y embutidos.

Palabras clave
Enfermedad vesicular porcina - Evaluación de riesgos - Fiebre aftosa - Gastroenteritis transmisible del cerdo - Peste porcina africana - Peste porcina clásica - Síndrome disgenésico y respiratorio porcino.

References


