Annex to:

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Annex C – Extensive literature search on the effectiveness of risk mitigation treatments for products of animal origin and products of non-animal origin

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1 Context

EFSA has been requested to provide scientific opinions to support the European Commission in the production of amending and implementing acts related to Regulation 2016/429 (the 'Animal Health Law' (AHL)) which lays down rules for the prevention and control of the following animal diseases:

- Foot and Mouth Disease (FMD)
- Infection with Rinderpest virus (RP)
- Infection with Rift Valley Fever virus (RVF)
- Infection with Lumpy Skin Disease virus (LSD)
- Infection with *Mycoplasma mycoides* subsp. *mycoides* (Contagious Bovine Pleuropneumonia) (CBPP)
- Sheep Pox and Goat Pox (SPGP)
- Infection with Peste des Petits Ruminants virus (PPR)
- Contagious Caprine Pleuropneumonia (CCPP)
- African Horse Sickness (AHS)
- Infection with *Burkholderia mallei* (Glanders)
- Classical Swine Fever (CSF)
- African Swine Fever (ASF)
- Highly Pathogenic Avian Influenza (HPAI)
- Infection with Newcastle Disease virus (NCD)

One of these EFSA scientific opinions consists of assessing the effectiveness of the risk-mitigating treatments for products of animal origin and other materials produced or processed in the restricted zone set out in the Delegated Act Annex VII and VIII, and, if relevant, suggest new treatments or procedures that can be effective to mitigate or to eliminate such risk (ToR 4.2). As part of this, EFSA has asked P95 (in the context of FWC OC/EFSA/ALPHA/2020/02 LOT 2) to carry out an extensive literature search (ELS).

The current report contains a brief overview of the study objectives as well as the main ELS findings.

2 Objective

As described in the final protocol (**Appendix 1**), the reference inclusion criteria for the ELS were based on the following PI[CO] strategy:

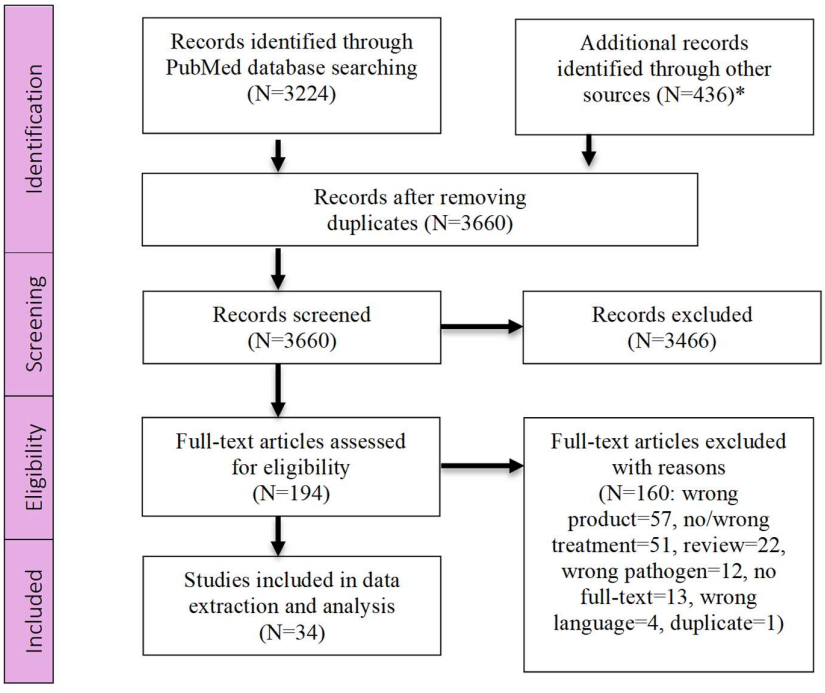
Population	Products: i. Meat (including blood), casings, milk and eggs (`products of animal origin`)[1] ii. Feed materials of plant origin and straw ('other materials') With presence of the causing pathogen of: FMD/Rinderpest/RVF/LSD/CBPP/PPR/CCPP/CSF/ASF/HPAI/NCD[2]
Intervention	Treatment to inactivate the pathogen
Comparison	Not Applicable
Outcome	Pathogen inactivation (as measured by diagnostic method)

[1] Originating from animals of listed species (Commission implementing Regulation (EU) 2018/1882 of 3 December 2018).
[2] The products to consider will depend on the pathogen. For three Cat. A diseases (SPGP, AHS and Glanders) no treatments are foreseen in Annexes VII and VIII.

3 Results

Screening and selection of references

We carried out the PubMed search on 07/04/2021. Adding records retrieved through reference list checking and hand searching via Google Scholar, a total of 3,660 unique references were identified. From these references, 34 were selected for data extraction. The full selection process is displayed in **Figure 1**.



* Records identified through reference list checking and hand searching in Google Scholar

Figure 1. Prisma diagram for ToR4.2 extensive literature search

Overview extracted data

Table 1 provides an overview of the 34 references from which data has been extracted.

Table 1. Overview of extracted references by disease of interest

Disease	#hits	References
FMD	15	(Blackwell and Hyde 1976; Cunliffe et al. 1979; Dhennin and Labié 1976; Hyde, Blackwell, and Callis 1975; Kästli and Moosbrugger 1968; Masana et al. 1995; Mebus et al. 1997; Salwa and Gaber 2007; Sellers 1969; Sonder et al. 1990; Tomasula et al. 2007; Walker et al. 1984; Wieringa-Jelsma et al. 2011; Wijnker, Haas, and Berends 2007, 2012)
RP	0	NA
RVF	0	NA
LSD	1	(Kononov et al. 2019)
CBPP	0	NA
CCPP	0	NA
PPR	0	NA
CSF	10	(Cowan et al. 2015; Helwig and Keast 1966; Jelsma et al. 2019; McKercher et al. 1980; McKercher et al. 1987; McKercher, Hess, and Hamdy 1978; Mebus et al. 1997; Stewart et al. 1979; Wieringa-Jelsma et al. 2011; Wijnker, Depner, and Berends 2008)
ASF	7	(Jelsma et al. 2019; McKercher et al. 1980; McKercher et al. 1987; McKercher, Hess, and Hamdy 1978; Mebus et al. 1997; Petrini et al. 2019; Wieringa-Jelsma et al. 2011)
HPAI	7	(Chmielewski and Swayne 2011; Chmielewski, Beck, and Swayne 2012; Isbarn et al. 2007; Swayne 2006; Swayne and Beck 2004; Thomas, King, and Swayne 2008; Thomas and Swayne 2007)
NCD	5	(Alexander and Manvell 2004; Chmielewski, Beck, and Swayne 2012; Gough 1973; Thomas, King, and Swayne 2008; Swayne and Beck 2004)

4 Disease-specific data extracted

Foot-and-mouth disease

Overall properties

FMD virus (FMDV) is inactivated by temperatures above 50°C. Although there is some variation between strains in resistance to temperature, exposure to 56°C for 30 min is sufficient to destroy most strains (AUSVETPLAN 2014). Heat inactivates FMDV, with a D-value of 6.06-10.87s at 70ºC, measured for virus suspended in saline solutions (Kamolsiripichaiporn et al. 2007). FMDV survives in lymph nodes and bone marrow at neutral pH (Sanson 1994), but is destroyed in muscle at pH below 6.0 (i.e., after rigor mortis)(Sellers et al. 1968). Meat matured at 2–7°C for 24–76 h (reaching pH< 6) and deboned is unlikely to contain infectious virus (ILSI 2009). Residual virus survives in milk and milk products during High temperature short time pasteurisation (HTST), but is inactivated by ultra-high-temperature (UHT) pasteurisation (OIE 2021a). FMDV may survive up to 74 days on pasture at 8–18°C and high relative

humidity, and 26–200 days in soil, sacking, hay or straw, depending on storage or climatic conditions (AUSVETPLAN 2014).

Meat

The following treatments are prescribed in the current EU legislation for meat:

- *Treatment 1: Heat treatment in an hermetically sealed container, to achieve a minimum F0 value (calculated killing effect on bacterial spores) of 3 (i.e. the coldest point in the product has been heated sufficiently to achieve the same killing effect as 121°C in 3 minutes with instantaneous heating and chilling)*
- *Treatment 2: Heat treatment to achieve a core temperature of 80°C*
- *Treatment 3: Heat treatment to achieve a core temperature of 70°C*
- *Treatment 4: Heat treatment (to meat previously de-boned and defatted) to achieve a core temperature of 70°C for a minimum of 30 minutes*
- *Treatment 5: In a hermetically sealed container, applying 60°C for a minimum of 4 hours*
- *Treatment 6: Natural fermentation and maturation for bone-in meat: minimum 9 months, to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 7: Natural fermentation and maturation for de-boned meat: minimum 9 months, to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 8: Drying after salting Spanish style bone-in hams and loins: 1. Iberian hams: minimum 252 days; 2. Iberian shoulders: minimum 140 days; 3. Iberian loins: minimum 126 days; 4. Serrano hams: minimum 140 days*

Masana et al. (Masana et al. 1995) evaluated the effectiveness against FMDV of low-temperature long-time cooking of meat over a range of 63 to 75°C. Both processing conditions of 71°C for 10.66 h and 75°C for 5.75 h successfully inactivated FMDV, as determined by cell culture and cattle inoculation.

Mebus et al. (1997) determined the following curing and processing times of for the inactivation of FMDV: 1. Iberian hams: minimum 168 days; 2. Iberian shoulders: minimum 112 days; 3. Iberian loins: minimum 42 days; 4. Serrano hams: minimum 182 days.

The OIE Terrestrial animal health code for FMDV (OIE 2021b) recommends in Article 8.8.31. the following procedures for the inactivation of FMDV in meat:

1. Canning: Meat and meat products are subjected to heat treatment in a hermetically sealed container to reach an internal core temperature of at least 70°C for a minimum of 30 minutes or to any equivalent treatment, which has been demonstrated to inactivate FMDV.
2. Thorough cooking: Meat, previously deboned and defatted, and meat products are subjected to a heat treatment that results in a core temperature of at least 70°C for a minimum of 30 minutes.
3. After cooking, they should be packed and handled in such a way they are not exposed to a source of FMDV.
4. Drying after salting: When rigor mortis is complete, the meat is deboned, treated with salt (NaCl) and 'completely dried'. It should not deteriorate at ambient temperature. 'Completely dried' is defined as a moisture protein ratio that is not greater than 2.25:1 or an Aw that is not greater than 0.85.

Based on the general properties of FMDV and on the available literature, the current EU-prescribed treatments are most likely effective.

Casings

The following treatments are prescribed in the current EU legislation for casings:

- *Treatment 1: Salting with sodium chloride (NaCl) either dry or as saturated brine (Aw< 0.80), for a continuous period of 30 days or longer at an ambient temperature of 20°C or above*
- *Treatment 2: Salting with phosphate supplemented salt 86.5% NaCl, 10.7% Na2HPO4 and 2.8% Na_3PO_4 either dry or as saturated brine (Aw< 0.80) for a continuous period of 30 days or longer at an ambient temperature of 20°C or above*

Wijnker, Haas, and Berends (Wijnker, Haas, and Berends 2007) showed that both treatments 1 and 2 were efficient on experimentally infected porcine and ovine casings stored at 20 ℃, whereas treated casings stored at 4°C still contained infectious virus.

Wieringa-Jelsma et al. (Wieringa-Jelsma et al. 2011) used a 3D collagen matrix model for animal casings to determine the inactivation of FMDV. Cells infected with FMDV were embedded in a bovine collagen type I gel matrix in order to investigate the effect of storage in saturated salt (NaCl) and in phosphate-supplemented NaCl at different temperatures over a period of 30 days. The detection limit in the non-treated samples was reached after 21, 7, 3 and 2 days of incubation at 4, 12, 20 and 25°C, respectively. Incubation with NaCl at the different temperatures resulted in curves that were similar to those for the untreated FMDV samples. The reduction of FMDV titres by phosphate salt treatment was significantly higher than the control treatment during days of incubation.

Wijnker, Haas, and Berends (Wijnker, Haas, and Berends 2012) carried out another study with casings of infected bovines, using the same treatments 1 and 2. Based on the combined results of the in-vivo and in-vitro experiments, it was concluded that the two treatments were sufficiently effective to inactivate FMDV in beef casings, and that the usage of phosphate-salt did not clearly enhance the inactivation of FMDV infectivity.

The OIE Terrestrial animal health code (OIE 2021b) in Article 8.8.38. also recommends Treatments 1 and 2, but with a cut-off temperature of 12°C instead of 20°C.

Based on this information, the current EU-prescribed treatments are most likely effective. However, according to the OIE, the temperature threshold could be lowered to 12°C instead of the 20°C described for both treatments.

Milk

The following treatments are prescribed in the current EU legislation for milk:

- *Treatment 1: Heat treatment (sterilization process) to achieve a minimum F0 value of 3*
- *Treatment 2: Heat treatment UHT Minimum 132°C for a minimum of 1 second*
- *Treatment 3: Heat treatment UHT Minimum 135°C for a suitable holding time*
- *Treatment 4: Heat treatment HTST (High temperature short time pasteurisation) if milk pH is lower than 7, minimum 72°C for a minimum of 15 seconds*
- *Treatment 5: Heat treatment HTST if milk pH is 7 or higher, minimum 72°C for a minimum of 15 seconds applied twice*
- *Treatment 6: Heat treatment HTST combined with a physical 72°C, treatment to achieve pH value below 6 for a minimum of 1 hour or to achieve a minimum of combined with desiccation*
- *Treatment 7: Pasteurisation consisting in a single heat treatment with an effect at least equivalent to that 72°C achieved by applying for 15 seconds*

Kästli and Moosbrugger (Kästli and Moosbrugger 1968) showed that FMDV is inactivated if milk is heated at 65°C for 30 s.

Sellers (Sellers 1969) found that for milk at pH 6.7, 99.99% inactivation of the virus (5 log reduction) was achieved in 17 s at 72°C, whereas, at pH 7.6, FMDV was inactivated after 55 s at 72°C.

Hyde, Blackwell, and Callis (Hyde, Blackwell, and Callis 1975) found that FMDV survived HTST treatment for 15-17 s. In addition, virus from infected milk survived heating at 80°C for the same time.

Blackwell and Hyde (Blackwell and Hyde 1976) showed that FMDV can survive in skimmed milk, cream and the pelleted cellular debris components of milk obtained from FMDV-infected cows after pasteurization at 72°C for 0.25 min.

Dhennin and Labie (Dhennin and Labié 1976) observed that, when skimmed milk was heated between 70°C and 90°C for 20 s, FMDV was not completely inactivated. However, when heated at 60°C for at least 320 s, the titre was almost null.

Cunliffe et al. (Cunliffe et al. 1979) found that heating milk at 148°C for at least 3 s (UHT) can inactivate FMDV in milk from infected dairy cows.

Walker et al. (Walker et al. 1984) observed that when heating FMDV-infected whole milk at temperatures (100°C), more than 20 min were needed to inactivate the virus, whereas at 148°C, 2.5 s were sufficient.

Sonder et al. (Sonder et al. 1990) showed that neither acidification nor hydrogen peroxide treatment was reliable for the inactivation of FMDV in skimmed milk. Either the virus was incompletely inactivated or the consistency of the milk was destroyed, making it unfit for consumption.

Tomasula et al. (Tomasula et al. 2007) observed that, although HTST treatment did not completely inactivate FMDV infectivity in whole and 2% milk, it greatly reduced the risk of natural transmission of FMDV by milk.

Salwa and Gaber (Salwa and Gaber 2007) studied the effect of several thermal processing methods on milk, and found that only UHT and double set pasteurisation proved to be effective for inactivating FMDV in milk.

In addition, EFSA (Authority 2006) mentioned in an Opinion that HTST treatment (72°C, 15 s) reduces the infectivity of FMDV in whole milk by 4-5 log, and that UHT treatment (132°C, 1s) causes a 10-fold higher inactivation than HTST treatment.

The OIE Terrestrial animal health code (OIE 2021b) recommends in Article 8.8.35. the EU-prescribed treatments 2, 4 and 5 for the importation of milk for human consumption. For the inactivation of FMDV present in milk for animal consumption (Article 8.8.36), one of the following procedures should be used: HTST applied twice; or HTST combined with another physical treatment, e.g., maintaining a pH 6 for at least one hour or additional heating to at least 72°C combined with desiccation (i.e., EU treatment 6); or UHT also combined with another physical treatment.

Based on the overall properties of FMDV and on the available literature, the current EU-treatments are most likely effective. It is noteworthy that some treatments (e.g., treatments 3 and 7) might need to be combined with another physical treatment (e.g., acidification) to be totally effective.

Products of non-animal origin

The following treatments are prescribed in the current EU legislation for feed materials of plant origin or straw:

- *Treatment 1: Heat treatment, minimum temperature of 80°C and for a minimum of 10 minutes, steam in a closed chamber.*
- *Treatment 2: Storage in package or bales under shelter at premises situated not closer than 2 km to the nearest outbreak and releasing from the premises do not take place before at least three months have elapsed following the completion of cleaning and disinfection according to Article 15.*

No data were available in the literature for these specific disease/product combinations. In general, Hay stored in uncovered bales of different diameters and different moisture contents were shown to reach maximum temperatures of 77.2°C (Coblentz and Hoffman, 2009), bales covered in tarpaulin reached temperatures of 40.7-44.9°C, depending on location of storage and tarpaulin colour (Guerrero et al., 2010).

The OIE Terrestrial animal health code (OIE 2021b) recommends in Article 8.8.28. for the importation of straw and forage one of the following treatments: 1.steam in a closed chamber such that the centre of the bales has reached a minimum temperature of 80°C for at least 10 minutes; 2.formalin fumes (formaldehyde gas) produced by its commercial solution at 35-40% in a chamber kept closed for at least 8 hours and at a minimum temperature of 19°C; Or have been kept in bond for at least 4 months before being released for export.

Based on this information, the current EU-prescribed treatments are most likely effective. In addition, the use of formalin fumes as abovementioned could be added to the list.

Rinderpest

Overall properties

RP virus (RPV) may survive in culture for at least 4 months at –20°C, 8 weeks at 4°C, 1 week at 20–25°C and more than 2.6 days at 37°C (Plowright 1968). The half-life periods of RPV infected lymph glands, spleens and bloods are 5 min and 2-3 days at 56°C and 7°C, respectively (Scott 1959). RPV is inactivated at pH values of less than 5.6 or more than 9.6 (Geering, Forman, and Nunn 1995). It is quickly inactivated in the environment, as it is sensitive to light, drying and ultraviolet radiation. Data suggests that it is inactivated at temperatures above 70°C (Doyle 2010; De Boer and Barber 1964). However, the virus can survive in shaded pastures and buildings for at least 48 h and may remain viable for long periods in chilled or frozen tissue. (OIE 2021a).

Products of non-animal origin

The following treatments are prescribed in the current EU legislation for feed materials of plant origin or straw:

- *Treatment 1*: *Heat treatment, minimum temperature of 80°C and for a minimum of 10 minutes, steam in a closed chamber.*
- *Treatment 2: Storage in package or bales under shelter at premises situated not closer than 2 km to the nearest outbreak and releasing from the premises do not take place before at least three months have elapsed following the completion of cleaning and disinfection according to Article 15.*

As indicated in **Table 1**, no data were available for these specific disease/product combinations.

It is noteworthy that the following recommendation is mentioned in the OIE Terrestrial animal health code (OIE 2021b) for international trade in livestock and their products (Article 8.16.4.): 'when authorising import or transit of livestock and their products, Veterinary Authorities should not require any rinderpest-related conditions'. In addition, the time needed to recover rinderpest free status of a country/zone/containment zone is either three months after the last case, or three months after the slaughter of all vaccinated animals, depending on the methods employed to achieve the elimination of the infection (Article 8.16.6.).

The EU-prescribed treatments for RPV inactivation are also prescribed by the EU and the OIE for the inactivation of FMDV in products of non-animal origin as described previously (section 3.3.1.5).

Given that RPV is quickly inactivated in the environment as it is sensitive to light, drying and ultraviolet radiation, and that the virus is inactivated at temperatures above 70ºC (Doyle 2010; De Boer and Barber 1964; OIE 2020), both treatments described in section 3.3.1.5 will most likely be effective in case of RPV contaminated feed materials of plant origin or straw.

Rift valley fever

Overall properties

RVF virus (RVFV) can survive for months at refrigeration temperatures and for years in the freezer. The virus is destroyed by heat treatment of serum for 120 min at 56°C and is resistant in alkaline environments but inactivated at pH below 6.8 (Bres 1981; OIE 2021a). It is inactivated by pH below 6.2 and would therefore be destroyed in meat that has matured and may be destroyed by stomach acid following ingestion.(Doyle 2010; Meegan 1979). RVFV is excreted in milk during the viraemic phase in animals (Jouan et al. 1989; Gerdes 2004; Authority 2005).

Meat

The following treatment is prescribed in the current EU legislation for meat:

• Treatment 1: Maturation of carcasses at a minimum temperature of 2°C for a minimum of 24 hours following slaughter

As indicated in **Table 1**, no data were available for this specific disease/product combination.

The OIE Terrestrial animal health code for RVF (OIE 2021b) recommends in Article 8.15.11. the same treatment in case of importation of fresh meat and meat products from ruminants from countries or zones not free from RVF.

The fall in pH normally results in the elimination of the RVFV 4-8 h after slaughter (Mims 1956). When meat is matured and chilled for 24 h, acid pH in meat inactivates virus (Chambers and Swanepoel 1980; ILSI 2009; AUSVETPLAN 2021). It is noteworthy that, if febrile animals are killed, rigor mortis and lactic acid formation do not occur and the pH changes may be minimal (Yedloutschnig, Dardiri, and Walker 1981). However, historically, there is no evidence to suggest that human or animal RVFV infections have been produced in this manner (Authority 2005).

Based on this information, the current EU-prescribed treatment is most likely effective.

Milk
The following treatment is prescribed in the current EU legislation for milk:

• Treatment 1: Pasteurisation consisting in a single heat treatment with an effect at least equivalent to that achieved by applying 72°C for 15 seconds

As indicated in **Table 1**, no data were available for this specific disease/product combination.

The OIE Terrestrial animal health code for RVF (OIE 2021b) recommends in Article 8.15.12. for the importation from countries or zones not free from RVF that milk and milk products are either subjected to pasteurisation or to a combination of control measures with equivalent performance as described in the Codex Alimentarius Code of Hygienic Practice for Milk and Milk Products.

There is circumstantial evidence that unpasteurised milk is infectious, but pasteurisation temperatures would be expected to result in rapid inactivation of the virus (AUSVETPLAN 2021; Chambers and Swanepoel 1980; Authority 2005). EFSA reviewed the risk related to the presence of RVFV in milk (Authority 2006) and reported that no data were available on the effect of pasteurisation at 72°C. EFSA concluded that by using heat treatment with acidification, RVFV survival may become negligible, whereas heat treatment alone may not completely remove virus infectivity. Based on this conclusion, the current prescribed treatment might not be totally effective.

Lumpy skin disease

Overall properties
LSD virus (LSDV) is remarkably stable, surviving for long periods at ambient temperature, especially in dried scabs. LSDV is very resistant to inactivation, surviving in necrotic skin nodules for up to 33 days or longer, desiccated crusts for up to 35 days, and at least 18 days in air-dried hides (Weiss 1968). It can remain viable for long periods in the environment. (OIE 2021a). There is no evidence of the virus persisting in the meat of infected animals, but it may be isolated from the milk in the early stages of the fever (ILSI 2009; Davies 1991). LSDV is inactivated at 55°C/2 hours and 65°C/30 minutes. It is susceptible to alkaline or acid pH. There is no significant reduction in titre when held at pH 6.6–8.6 for 5 days at 37°C (OIE 2021a).

Meat
The following treatment is prescribed in the current EU legislation for meat:

- *Treatment 1: Removal of offal*[1]

As indicated in **Table 1**, only one reference was available for LSD (Kononov et al. 2019); the authors found that infected animals are reservoirs of live LSDV in lymph nodes and testicles, whereas deep skeletal meat does not carry live virus and the risk of transmission through this product seems very low.

The OIE Terrestrial animal health code for LSD (OIE 2021b) mentions in Article 11.9.2. that 'When authorising import or transit of the following commodities, Veterinary Authorities should not require any LSD-related conditions regardless of the status of the animal population of the exporting country: skeletal muscle meat; casings; gelatine and collagen; tallow; hooves and horns.

Based on this information, the current EU prescribed treatment is most likely effective.

Casings

The following treatment is prescribed in the current EU legislation for casings:

- *Treatment 1: Removal of offal*[1]

No data were available for this specific disease/product combination.

The OIE Terrestrial animal health code for LSD (OIE 2021b) mentions in Article 11.9.2. that 'When authorising import or transit of the following commodities, Veterinary Authorities should not require any LSD-related conditions regardless of the status of the animal population of the exporting country: skeletal muscle meat; casings; gelatine and collagen; tallow; hooves and horns.

Based on this information, the current EU prescribed treatment is most likely effective.

Milk

The following treatment is prescribed in the current EU legislation for milk:

- *Treatment 1: Pasteurisation consisting in a single heat treatment with an effect at least equivalent to that achieved by applying 72°C for 15 seconds*

No data were available for this specific disease/product combination.

The OIE Terrestrial animal health code for LSD (OIE 2021b) recommends in Article 11.9.11. for the importation that milk and milk products were subjected to pasteurisation or any combination of control measures with equivalent performance as described in the Codex Alimentarius Code of Hygienic Practice for Milk and Milk Products.

A report from ILSI (ILSI 2009) mentions that there is evidence that heating at 56°C for 30 min or 60°C for 10 min inactivates virus in milk for suckling calves (references from ILSI 2009: personal communication T. Gerdes; OIE 2021a). In addition, EFSA reviewed the risk related to the presence of LSDV in milk (Authority 2006) and concluded that, although no data were available on the effect of LTST, by using heat treatment with acidification, the virus survival may become negligible. Based on this information, the current prescribed treatment might not be totally effective and require an additional acidification.

Contagious bovine pleuropneumonia

Overall properties

Mycoplasma mycoides subsp. mycoides (*Mmm*) does not survive for long in the environment and transmission requires close contact. *Mmm* is inactivated within 60 min at 56°C and 2 min at 60°C. It is

[1] 'offal' means fresh meat other than that of the carcass as defined in (7), even if it remains naturally connected to the carcass; (7): carcass of an ungulate' means the whole body of a slaughtered or killed ungulate after: bleeding, in the case of slaughtered animals; evisceration; removal of the limbs at the carpus and tarsus; removal of the tail, the udder, the head and the skin, except in porcine animals.

inactivated by acid and alkaline pH. Mycoplasma species do not grow below pH 6 (Mitscherlich and Marth 2012). *Mmm* survives outside the host for up to 3 days in tropical areas and up to 2 weeks in temperate zones. Frozen, it may survive more than 10 years (OIE 2021a).

Meat

The following treatment is prescribed in the current EU legislation for meat:

- *Treatment 1: Removal of offal*[2]

As indicated in **Table 1**, no data were available for this specific disease/product combination.

The OIE Terrestrial animal health code for CBPP (OIE 2021b) mentions in Article 11.5.2. that 'When authorising import or transit of the following commodities, Veterinary Authorities should not require any CBPP-related conditions, regardless of the CBPP status of the domestic bovids and water buffalo population of the exporting country, zone or compartment: milk and milk products; hides and skins; meat and meat products (excluding lung).' These are considered as 'safe commodities'.

Based on this, the current prescribed treatment is most likely effective.

Milk

In the current EU legislation for milk:

Safe commodities

As indicated in **Table 1**, no data were available for this specific disease/product combination.

The OIE Terrestrial animal health code for CBPP (OIE 2021b) mentions in Article 11.5.2. that 'When authorising import or transit of the following commodities, Veterinary Authorities should not require any CBPP-related conditions, regardless of the CBPP status of the domestic bovids and water buffalo population of the exporting country, zone or compartment: milk and milk products; hides and skins; meat and meat products (excluding lung).' These are considered as 'safe commodities'.

The current EU legislation is line with the OIE code.

Peste des petits ruminants

Overall properties

PPR virus (PPRV) is considered fragile and does not survive more than 4 days in the environment; however, it may survive for long periods in chilled and frozen tissue. As all members of the *Paramyxoviridae* family, PPRV is very heat sensitive (Diallo et al. 2007; AUSVETPLAN 2020). The half-life of the virus at 37°C and 56°C is 3 h and 2.2 min, respectively. PPRV is inactivated at pH below 4 and above 11 (Health and Welfare 2012; Doyle 2010; OIE 2021a).

Meat

The following treatments are prescribed in the current EU legislation for meat:

- *Treatment 1: Heat treatment in an hermetically sealed container, to achieve a minimum calculated killing effect on bacterial spores (F0 value) of 3 (i.e., the coldest point in the product has been heated sufficiently to achieve the same killing effect as 121°C in 3 minutes with instantaneous heating and chilling*
- *Treatment 2: Heat treatment to achieve a core temperature of 80°C*
- *Treatment 3: Heat treatment to achieve a core temperature of 70°C*

[2] 'offal' means fresh meat other than that of the carcass as defined in (7), even if it remains naturally connected to the carcass; (7): carcass of an ungulate' means the whole body of a slaughtered or killed ungulate after: bleeding, in the case of slaughtered animals; evisceration; removal of the limbs at the carpus and tarsus; removal of the tail, the udder, the head and the skin, except in porcine animals.

- *Treatment 4: Heat treatment (to meat previously de-boned and defatted) to achieve a core temperature of 70°C for a minimum of 30 minutes*
- *Treatment 5: In an hermetically sealed container, applying 60°C for a minimum of 4 hours*
- *Treatment 6: Heat treatment to achieve a core temperature of 65°C for a period of time to achieve a minimum pasteurisation value of 40*

As indicated in **Table 1**, no data were available for these specific disease/product combinations.

The OIE Terrestrial animal health code for PPR (OIE 2021b) recommends in Article 14.7.22. for the importation that meal and flour from blood, meat, defatted bones, hooves, claws and horns should be processed using heat treatment to a minimum internal temperature of 70°C for at least 30 minutes.

Data for PPRV is lacking, but similarly to RPV, it is expected to be rapidly inactivated at temperatures above 70°C. The virus is destroyed by heating at 50°C for 60 min. (AUSVETPLAN 2020; Knight, Haines, and Zuber 2013; Doyle 2010).

Based on this information and on PPRV survival characteristics, the current prescribed treatments are most likely effective.

Casings

The following treatments are prescribed in the current EU legislation for casings:

- *Treatment 1: Salting with sodium chloride (NaCl) either dry or as saturated brine (Aw < 0.80), for a continuous period of 30 days or longer at an ambient temperature of 20°C or above*
- *Treatment 2: Salting with phosphate supplemented salt 86.5 % NaCl, 10.7 % Na_2HPO_4 and 2.8 % Na_3PO_4 either dry or as saturated brine (Aw < 0.80) for a continuous period of 30 days or longer at an ambient temperature of 20°C or above*

As indicated in **Table 1**, no data were available for these specific disease/product combinations.

The OIE Terrestrial animal health code for PPR (OIE 2021b) recommends in Article 14.7.26, the same procedures for the inactivation of PPRV in casings of sheep and goats.

An EFSA scientific opinion on mitigation treatments for animal casings (Health and Welfare 2012) reported here that no data were available on inactivation of PPRV on casings from experimentally infected animals, in spiked casings or in a 3D matrix model, but that from the general characteristics of these viruses, it could be assumed that they will be inactivated within 30 days at room temperature.

Based on this information, the current prescribed treatments are most likely effective.

Milk

The following treatments are prescribed in the current EU legislation for milk:

- *Treatment 1: Heat treatment UHT: Minimum 132°C for a minimum of 1 second*
- *Treatment 2: Heat treatment HTST if milk pH is lower than 7, minimum 72°C for a minimum of 15 seconds*
- *Treatment 3: Heat treatment HTST if milk pH is 7 or higher, minimum 72°C for a minimum of 15 seconds applied twice*

As indicated in **Table 1**, no data were available for these specific disease/product combinations.

The OIE Terrestrial animal health code for PPR (OIE 2021b) recommends in Article 14.7.19. the same three treatments for the importation of milk for human consumption. For the inactivation of PPRV present in milk for animal consumption, one of the following procedures should be used: HTST applied twice; or HTST combined with another physical treatment, e.g., maintaining a pH 6 for at least one hour or additional heating to at least 72°C combined with desiccation; or UHT also combined with another physical treatment. The treatments recommended by the OIE for PPRV inactivation are based on the abovementioned treatments for FMDV.

In an EFSA scientific opinion (Authority 2006), it was reported that, although no specific data for milk were available, based on the inactivation kinetics at lower temperatures for virus in various solutions, HTST treatment is likely to give at least a 10^6-fold reduction in infectivity and thus remove any risk of

PPRV being transferred to susceptible species. Single HTST treatment is likely to render the milk safe for use as animal feed and double heat treatment is likely to provide a very good safety margin (Chandra et al. 1999; Authority 2006). It is also stated that UHT treatment is likely to cause a 10-fold higher inactivation than HTST, thus, the risk of UHT treated milk is likely to be reduced 10-fold compared to single HTST treated milk. Nevertheless, it is stressed that directive 2002/99/EC "laying down the animal health rules governing the production, processing, distribution and introduction of products of animal origin for human consumption", which governs products for human use, does not approve HTST, double HTST or UHT in relation to PPRV, and only recognises these treatments in relation to FMDV, presumably due to lack of direct experimental evidence regarding PPRV (Authority 2006).

Based on this information, Treatment 3 is most likely effective for both human and animal consumption milk. On the other hand, based on the OIE recommendations for the inactivation of PPRV present in milk for animal consumption, a preliminary treatment of milk might be needed for Treatments 1 and 2 to be completely effective.

Contagious caprine pleuropneumonia

Overall properties

Mycoplasma capricolum subsp. capripneumoniae (Mccp), the agent of CCPP, is inactivated within 60 min. at 56°C and within 2 min. at 60°C, but can survive more than 10 years in frozen, infected pleural fluid. Mccp is very fragile and not able to survive long in the external environment. On average, it only survives outside the host for up to 3 days in tropical areas and up to 2 weeks in temperate zones. Cultures can be inactivated by ultraviolet radiation within a few minutes (OIE 2021a).

Meat

The following treatment is prescribed in the current EU legislation for meat:

- *Treatment 1: Removal of offal*[3]

As indicated in **Table 1**, no data were available for this specific disease/product combination.

The OIE Terrestrial animal health code for CCPP (OIE 2021b) does not mention anything related to meat.

The CCPP agent is transmitted mainly by means of contaminated aerosol emitted by the affected lungs of mycoplasma-shedding animals. The pathogen is not present in the meat (Rapoport and Shimshony 1997). Therefore, the removal of offal is most likely a sufficient measure to ensure that meat is safe for consumption.

Milk

In the current EU legislation for milk:

Safe commodities

As indicated in **Table 1**, no data were available for this specific disease/product combination

The OIE Terrestrial animal health code (OIE 2021b) does not mention anything related to milk.

The current EU legislation is in line with the OIE code.

[3] 'offal' means fresh meat other than that of the carcass as defined in (7), even if it remains naturally connected to the carcass; (7): carcass of an ungulate' means the whole body of a slaughtered or killed ungulate after: bleeding, in the case of slaughtered animals; evisceration; removal of the limbs at the carpus and tarsus; removal of the tail, the udder, the head and the skin, except in porcine animals.

Classical swine fever

Overall properties

CSF virus (CSFV) can survive in fresh pig meat and some processed pig meat products. Survival can be prolonged for months when meat is stored cool, or even for years when it is stored frozen (Terpstra 1991). CSFV survives in meat during salt curing and smoking up to more than 180 days depending on the process used. The virus is readily inactivated by heating meat to 65.5°C for 30 min or 90–100°C for 1 min. Cell culture fluid infectivity is lost after 10 min at 60°C (Walton 1993). CSFV is rapidly inactivated at pH below 4.0 or above 11 (Terpstra 1991); (Farez and Morley 1997; OIE 2021a).

Meat

The following treatments are prescribed in the current EU legislation for meat:

- *Treatment 1: Heat treatment in a hermetically sealed container, to achieve a minimum calculated killing effect on bacterial spores (F0 value) of 3 (i.e., the coldest point in the product has been heated sufficiently to achieve the same killing effect as 121°C in 3 minutes with instantaneous heating and chilling)*
- *Treatment 2: Heat treatment to achieve a core temperature of 80°C*
- *Treatment 3: Heat treatment to achieve a core temperature of 70°C*
- *Treatment 4: Heat treatment (to meat previously de-boned and defatted) to achieve a core temperature of 70°C for a minimum of 30 minutes*
- *Treatment 5: In a hermetically sealed container, applying 60°C for a minimum of 4 hours*
- *Treatment 6: Natural fermentation and maturation for bone-in meat: minimum 9 months, to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 7: Natural fermentation and maturation for de-boned meat: minimum 9 months, to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 8: Natural fermentation for loins: minimum 140 days to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 9: Natural fermentation for hams: minimum 190 days to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 10: Drying after salting Italian style bone-in hams: minimum 313 days*
- *Treatment 11: Drying after salting Spanish style bone-in hams and loins: 1. Iberian hams: minimum 252 days; 2. Iberian shoulders: minimum 140 days; 3. Iberian loins: minimum 126 days; 4. Serrano hams: minimum 140 days*

McKercher et al. (McKercher, Hess, and Hamdy 1978) showed that heating infected hams until the internal temperature reached 69°C successfully inactivated CSFV.

Stewart et al. (Stewart et al. 1979) found that a flash temperature of 71°C during 1 min caused inactivation of CSFV in ham. CSFV was inactivated from canned ham when an internal temperature of 65°C was sustained during 90 min.

McKercher et al. (McKercher et al. 1987) obtained conclusive proof of virus inactivation after 313 days of processing of Prosciutto di Parma (Parma ham).

Mebus et al. (Mebus et al. 1997) found the following curing and processing times for the inactivation of CSFV: 1. Iberian hams: minimum 252 days; 2. Iberian shoulders: minimum 140 days; 3. Iberian loins: minimum 126 days; 4. Serrano hams: minimum 140 days.

Cowan et al. (Cowan et al. 2015) studied the effect of heating infected meat at 68°C. This temperature was selected to determine the extent of virus survival if treatments do not reach the required temperature prescribed by the EU legislation (i.e., 70°C). At 68°C, a $\log_{10}$ reduction in the amount of virus was achieved in muscle within 0.41 min. The authors highlighted that the EU legislation does not specify a time for which the 70°C target must be applied for, but estimation of $D^4$70 values indicate that the target temperature would have to be achieved for over 1 min to reduce material with a titre of 10^4 TCID50/g to zero. Much longer periods would be required if conservative estimates based on the upper

[4] decimal reduction time

95%CI are applied. The authors estimated that the D80 value for muscle is 2.8 s, indicating that adopting this temperature as a minimum for acceptable treatments would eliminate the possibility for viable CSFV to remain.

The OIE Terrestrial animal health code for CSF (OIE 2021b) recommends in Article 15.2.23. for the inactivation of CSFV in meat either heat treatment (treatments 1 or 3), natural fermentation and maturation (EU treatments 6 to 9), and dry cured pork meat (treatments 10 and 11).

Based on the overall characteristics of CSFV and the available literature, the EU-prescribed treatments are most likely still the most effective.

Casings

The following treatments are prescribed in the current EU legislation for casings:

- *Treatment 1: Salting with sodium chloride (NaCl) either dry or as saturated brine (Aw< 0.80), for a continuous period of 30 days or longer at an ambient temperature of 20°C or above*
- *Treatment 2: Salting with phosphate supplemented salt 86.5% NaCl, 10.7% Na_2HPO_4 and 2.8% Na_3PO_4 either dry or as saturated brine (Aw 0.80) for a continuous period of 30 days or longer at an ambient temperature of 20°C or above.*

Helwig and Keast (Helwig and Keast 1966) found that salted and standard processed casings infected with CSFV were inactivated within 17 days.

McKercher et al. (McKercher et al. 1980) showed that CSFV survived for at least 147 days in infected natural casings treated with saturated NaCl brine and stored at 4°C.

Wieringa-Jelsma et al (Wieringa-Jelsma et al. 2011) used a 3D collagen matrix model for animal casings to determine the inactivation of CSFV in casings. Cells infected with CSFV were embedded in a bovine collagen type I gel matrix in order to investigate the effect of storage in NaCl and storage in phosphate-supplemented NaCl at different temperatures over a period of 30 days. Phosphate supplemented salt treatment increased the effect temperature had on inactivation of CSFV. In contrast, the NaCl treatment only increased CSFV inactivation at the higher temperatures (20°C and 25°C). These results were later confirmed by an in vivo experiment (Jelsma et al. 2019).

In addition, Wijnker, Depner and Berends (Wijnker, Depner, and Berends 2008) showed that processing casing with citrate-supplemented salt solution containing 89.2 % NaCl, 8.9 % trisodium citrate dehydrate and 1.9 % citric acid monohydrate (wt/wt/wt), with pH 4.5, was successful in removing infectivity only after 30 days, and only when casings were preserved at 20ºC (Health and Welfare 2012).

The OIE Terrestrial animal health code for CSF (OIE 2021b) in Article 15.2.24. recommends also the two EU-prescribed treatments.

Based on the available literature, the EU-prescribed treatments are most likely effective.

African swine fever

Overall properties

ASF virus (ASFV) remains viable for long periods in blood, faeces and tissues. In meat products, ASFV may persist for several weeks or months in frozen or uncooked meat. The virus is highly resistant to low temperatures but is inactivated at 56°C for 70 min and at 60°C for 20 min. Infected blood heated for 30 min at 60°C loses infectivity. ASFV is very resistant to acid pH (Plowright and Parker 1967), but is inactivated by pH below 3.9 or above 11.5 in serum-free medium (Farez and Morley 1997; Health and Welfare 2012; OIE 2021a).

Meat

The following treatments are prescribed in the current EU legislation for meat:

- *Treatment 1: Heat treatment in a hermetically sealed container, to achieve a minimum calculated killing effect on bacterial spores (F0 value) of 3 (i.e., the coldest point in the product has been heated sufficiently to achieve the same killing :effect as 121°C in 3 minutes with instantaneous heating and chilling*

- *Treatment 2: Heat treatment to achieve a core temperature of 80°C*
- *Treatment 3: In a hermetically sealed container, applying 60°C for a minimum of 4 hours*
- *Treatment 4: Natural fermentation and maturation for de-boned meat: minimum 9 months, to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 5: Natural fermentation for loins: minimum 140 days to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 6: Natural fermentation for hams: minimum 190 days to achieve maximum values of Aw of 0.93 and pH of 6*
- *Treatment 7: Drying after salting Spanish style bone-in hams and loins: 1. Iberian hams: minimum 252 days; 2. Iberian shoulders: minimum 140 days; 3. Iberian loins: minimum 126 days; 4. Serrano hams: minimum 140 days*

McKercher et al. (McKercher, Hess, and Hamdy 1978) showed that the heating of infected hams until the internal temperature reached 69°C successfully inactivated ASFV. In another study, McKercher et al. (McKercher et al. 1987) obtained conclusive proof of virus inactivation after 399 days of processing of Prosciutto di Parma (Parma ham).

Mebus et al. (Mebus et al. 1997) found the following curing and processing times of for the inactivation of ASFV: 1. Iberian hams: minimum 140 days; 2. Iberian shoulders: minimum 140 days; 3. Iberian loins: minimum 112 days; 4. Serrano hams: minimum 140 days.

Petrini et al. (Petrini et al. 2019) tested the effect of dry curing process on inactivation of ASFV in different traditional Italian dry-cured meat products (i.e., drying after salting), salami, pork belly, and loin prepared from experimentally infected pigs. For loin, conclusive proof of virus inactivation was obtained after 137 days of processing.

The OIE Terrestrial animal health code for ASF (OIE 2021b) mentions in Article 15.1.2. that 'When authorising import or transit of the following commodities, Veterinary Authorities should not require any ASF-related conditions, regardless of the ASF status of the exporting country or zone: meat in a hermetically sealed container with a Fo value of 3 or above' (i.e. treatment 1 in current EU legislation). In Article 15.1.23., the following procedures are mentioned for the inactivation of ASFV in meat: 'Meat should be subjected to heat treatment for at least 30 minutes at a minimum temperature of 70°C, which should be reached throughout the meat; or the meat should be cured with salt and dried for a minimum of six months.'

No data on EU-prescribed treatments 4 and 6 for ASFV were available in the literature. However, as mentioned above, the OIE Terrestrial animal health code for CSFV (OIE 2021b) recommends these two treatments for the inactivation of CSFV in meat.

Based on the general properties of ASFV and on the available literature, the EU-prescribed treatments are most likely still the most effective.

Casings

The following treatments are prescribed in the current EU legislation for casings:

- *Treatment 1: Salting with sodium chloride (NaCl) either dry or as saturated brine (Aw< 0.80), for a continuous period of 30 days or longer at an ambient temperature of 20°C or above*
- *Treatment 2: Salting with phosphate supplemented salt 86,5% NaCl, 10.7% Na_2HPO_4 and 2.8% Na_3PO_4 either dry or as saturated brine (Aw< 0.80) for a continuous period of 30 days or longer at an ambient temperature of 20°C or above*

Wieringa-Jelsma et al (Wieringa-Jelsma et al. 2011) used a 3D collagen matrix model for animal casings to determine the inactivation of ASFV in casings. Cells infected with ASFV were embedded in a bovine collagen type I gel matrix in order to investigate the effect of storage in NaCl and storage in phosphate-supplemented NaCl at different temperatures over a period of 30 days. Both NaCl and phosphate salt were able to inactivate ASFV. At 4°C, low titres were still observed after NaCl treatment up to day 15, while phosphate salt treatment resulted in titres reaching the detection limit by day 2 at all temperatures. These results were later confirmed by an in vivo experiment (Jelsma et al. 2019), (Health and Welfare 2012).

The OIE Terrestrial animal health code for ASF (OIE 2021b) in Article 15.1.24 recommends also treatments 1 and 2, except that the storage temperature may be decreased till 12°C instead of 20°C.

Based on the available literature, the EU-treatments are most likely effective. It is noteworthy that the threshold temperature could be decreased, based on the OIE code (2021b).

Highly pathogenic avian influenza

Overall properties

HPAI virus (HPAIV) can be isolated in breast thigh meat from infected birds despite no apparent clinical symptoms (Doyle 2010; ILSI 2009; Knight, Haines, and Zuber 2013; OIE 2021a). In samples from experimentally infected chickens stored at 4°C, HPAIV can survive up to 240 days in feather tissues, 160 days in muscle, and 20 days in liver (Yamamoto, Nakamura, and Mase 2017). Survival of the virus is mainly temperature dependant. Cooking contaminated meat at 70°C for 5 s, 56-60°C for 60 min. is sufficient to inactivate the virus. Extreme pH values (pH 1-3 or pH 10-14), ionizing radiation, salinity and chemical agents can inactivate the virus. The latter are not always compatible with standard food processing requirements (De Benedictis, Beato, and Capua 2007; Emmoth 2015; ILSI 2009; Knight, Haines, and Zuber 2013).

Meat

The following treatments are prescribed in the current EU legislation for meat:

- *Treatment 1: Heat treatment in a hermetically sealed container, to achieve a minimum killing effect on bacterial spores (F0 value) of 3 (i.e., the coldest point in the product has been heated sufficiently to achieve the same killing effect as 121°C in 3 minutes with instantaneous heating and chilling)*
- *Treatment 2: Heat treatment to achieve a core temperature of 80°C*
- *Treatment 3: Heat treatment to achieve a core temperature of 70°C*
- *Treatment 4: Core temperature of 73.9°C for a minimum of 0.51 seconds*
- *Treatment 5: Core temperature of 70.0°C for a minimum of 3.5 seconds*
- *Treatment 6: Core temperature of 65.0°C for a minimum of 42 seconds*
- *Treatment 7: Core temperature of 60°C for a minimum of 507 seconds*

Swayne et al. (Swayne 2006) investigated experimentally thermal inactivation conditions in chicken meat. Data indicated that thermal inactivation was dependent on virus infectivity titres, but that no HPAIV was isolated after 5 s of treatment at 70°C. The findings also highlighted that given similar chemo-physical properties, strain-to-strain variation is expected minimal.

Isbarn et al. (Isbarn et al. 2007) demonstrated that incubation at 63°C for 2 min and 500MPa at 15°C for 15 s inactivated similarly more than 10^5 PFU/ml, thereby showing that HPAIV is not only heat-sensitive but also baro-sensitive, and that combining pressure with heat can reduce the heat-time ratio required for inactivation.

Thomas et al. (Thomas and Swayne 2007) studied the thermal inactivation of HPAIV in naturally infected chicken meat and found D-values[5] between 241.2 s and 33.1 s for 57°C to 61°C. Predicted D-values for 70°C and 73.9°C were 0.28 s and 0.073 s, receptively.

Thomas et al. (Thomas, King, and Swayne 2008) found for infected chicken meat D-values between 267.6 s and 23.6 s for 57°C to 61°C. The cooking tests at 70 and 73.9°C were negative for virus isolation regardless of time at heart.

Chmielewski et al. (Chmielewski and Swayne 2011) found for infected chicken meat D-values of 4 min to 0.03 s, at temperatures of 57.8 and 73.9°C, respectively. The authors also concluded that the standard cooking temperature of 70°C for poultry meat would adequately inactivate HPAIV in chicken meat.

[5] decimal reduction time

The OIE Terrestrial animal health for HPAI (OIE 2021b) in Article 10.4.24. recommends for poultry meat the EU-prescribed treatments 4-7.

Based on the overall properties of HPAIV and the current literature, the EU-prescribed treatments are most likely effective. However, it was found that other methods could be suitable for viral inactivation. For instance, HPAIV was found as well to be baro-sensitive and could be inactivated more effectively if specific heat and pressure combinations were used synergistically (Isbarn et al. 2007).

Eggs

The following treatments are prescribed in the current EU legislation for eggs:

- *Whole egg:*
  - *Treatment 1: Heat treatment: 60°C - 188 seconds*
  - *Treatment 2: Heat treatment: completely cooked*
- *Whole egg blends:*
  - *Treatment 1: 60°C - 188 seconds*
  - *Treatment 2: completely cooked*
  - *Treatment 3: 61.1°C - 94 seconds*
- *Liquid egg white:*
  - *Treatment 1: 55.6°C - 870 seconds*
  - *Treatment 2: 56.7°C - 232 seconds*
- *Plain or pure egg yolk:*
  - *Treatment 1: 60°C - 288 seconds*
- *10% salted yolk:*
  - *Treatment 1: 62.2°C - 138 seconds*
- *Dried egg white:*
  - *Treatment 1: 67°C - 20 hours*
  - *Treatment 2: 54.4°C – 50.4 hours*
  - *Treatment 3: 51.7°C – 73.2 hours*

Swayne et al. (Swayne and Beck 2004) tested different heat-inactivation methods in chicken eggs and products. In order to reach a 2 $\log_{10}$ reduction (i.e., probability of 1/100 of having 1 EID50/ml), the following pasteurisation times were found: (i) Whole egg: 60°C, 188 s, (ii) Whole egg blends: 60°C, 188 s, or 61.1°C, 94 s, (iii) Liquid egg white: 55.6°C, 256 s, or 56.7°C, 228 s, (iv)10% salted yolk: 62.2°C, < 138 s, or 63.3°C, < 138 s, (v): dried egg white: 54.4°C, 21.38 d, or 67°C, 0.83 d.

Chmielewski et al. (Chmielewski and Swayne 2011) showed that for whole homogenised eggs, liquid egg white and 10% salted eggs, the USDA pasteurisation standards are sufficient for inactivating HPAIV. Furthermore, heating at 63°C at minimum 20 s enabled a $\log_{10}$ reduction.

Chmielewski et al. (Chmielewski, Beck, and Swayne 2012) showed that the USDA pasteurisation processes for fortified (61.1°C, 6.2 min or 62.2°C, 3.5 min), sugared/salted (62.2°C, 6.2 min or 63.3°C for 3.5 min) egg or plain yolk (60°C, 6.2 min or 61.1°C, 3.5 min), and whole egg (60°C, 3.5 min) products resulted in more than 5-log reductions in HPAIV at the lower temperature–longer times. In addition, a > 5-log reduction was also demonstrated for the five products at the higher temperatures–shorter times of USDA-approved pasteurisation processes.

The OIE Terrestrial animal health code for HPAI (OIE 2021b), in Article 10.4.23, recommends the same treatments as EU-prescribed treatments, except that EU also considers for whole egg and whole egg blends heat-treatment as 'completely cooked'.

Based on the overall properties of HPAIV and on the literature findings, the current EU-prescribed treatments are most likely effective. However, for some egg products such as liquid egg white, longer heating pasteurisation times might be required. Furthermore, other methods could be suitable for HPAIV inactivation. For instance, HPAIV was found as well to be baro-sensitive and could be inactivated more effectively if specific heat and pressure combinations were used synergistically (Isbarn, 2007).

Newcastle disease

Overall properties

NCD virus (NCDV) survives for long periods at ambient temperature, especially in faeces. It can survive in frozen poultry carcases (bone marrow and muscle), at refrigerator temperatures, for up to 4 months. In eggs at room temperature, NCDV may survive for months, and more than one year at 4°C. The virus is inactivated by acid pH lower than 2 and by treating at by 56°C for 3 h or 60°C for 30 min (OIE 2021a; ILSI 2009; Knight, Haines, and Zuber 2013).

Meat

The following treatments are prescribed in the current EU legislation for meat:

- *Treatment 1: Heat treatment in a hermetically sealed container, to achieve a minimum killing effect on bacterial spores (F0 value) of 3 (i.e., the coldest point in the product has been heated sufficiently to achieve the same killing effect as 121°C in 3 minutes with instantaneous heating and chilling)*
- *Treatment 2: Heat treatment to achieve a core temperature of 80°C*
- *Treatment 3: Heat treatment to achieve a core temperature of 70°C*
- *Treatment 4: Core temperature of 73.9°C for a minimum of 0.51 seconds*
- *Treatment 5: Core temperature of 70.0°C for a minimum of 3.5 seconds*
- *Treatment 6: Core temperature of 65.0°C for a minimum of 42 seconds*
- *Treatment 7: Core temperature of 60°C for a minimum of 507 seconds*

Alexander et al. (Alexander and Manvell 2004) studied heat inactivation methods of NCDV in artificially infected chicken meat homogenate. The following D_t-values for meat homogenate were estimated: at 65°C, 120 s; at 70°C, 82 s; at 74°C, 40 s and at 80°C, 29 s.

Thomas et al. (Thomas, King, and Swayne 2008) studied the thermal inactivation of NCDV in artificially and naturally infected chicken meat. NCDV was inactivated similarly in naturally and artificially infected meat. Linear regression models predicted that the current USDA–Food Safety and Inspection Service time-temperature guidelines for cooking chicken meat to achieve a 7-log reduction of *Salmonella* also would effectively inactivate the NCDV strains tested (i.e., 57.8°C for 63.3 min). 56.7

The OIE Terrestrial animal health code for NCD (OIE 2021b), in Article 10.9.21 does not recommend the exact same treatments as EU-prescribed treatments. While EU prescribes for meat the same heat treatments as for HPAI, the OIE prescribes different treatments (i.e., core temperature of 65, 70, 74, 80°C should be maintained for 39.8, 3.6, 0.5, 0.03 seconds respectively). Furthermore, EU recommends as well as heat treatments to achieve a core temperature of 80°C and/or 70°C as effective, regardless of time as well as heat treatments in a hermetically sealed container, to achieve a minimum F0* value of 3 for NCD.

The ELS findings above confirm that heat treatment seems the most appropriate inactivation method as stated elsewhere (ILSI 2009). Heat treatment of infected meat at temperature above 70°C for at least 82 s seems to be sufficient for inactivation (Alexander and Manvell 2004; Thomas, King, and Swayne 2008).

Eggs

The following treatments are prescribed in the current EU legislation for eggs:

- *Whole egg:*

Treatment 1: 55°C – 2521 seconds
Treatment 2: 57°C - 1596 seconds
Treatment 3: 59°C - 674 seconds
completely cooked

- *Liquid egg white:*

Treatment 1: 55°C – 2 278 seconds
Treatment 2: 57°C - 986 seconds
Treatment 3: 59°C – 301 seconds

- *10% salted egg yolk:*

Treatment 1: 55°C - 176 seconds

- *Dried egg white:*

Treatment 1: 57°C - 54 hours

Gough et al. (Gough 1973) looked at the thermostability of NCDV in liquid whole egg. Measurable viral population of virus resistant to heat was found at 64.4°C for up to 200 s. The study revealed that the pasteurisation process (64.4°C for > 2.5min) as prescribed by the regulations of 1963 was effective for inactivating NCDV.

Swayne et al. (Swayne and Beck 2004) demonstrated that all NCDV were inactivated in all egg products (homogenised whole egg, liquid egg white, 10% salted egg yolk) when treated using USDA industry standard pasteurisation protocols.

Chmielewski et al. (Chmielewski, Beck, and Swayne 2012) evaluated the USDA egg pasteurisation processes on the inactivation of velogenic NCDV in processed egg products. The pasteurisation processes for fortified (61.1°C, 6.2 min or 62.2°C, 3.5 min), sugared/salted (62.2°C, 6.2 min or 63.3°C for 3.5 min) egg or plain yolk (60°C, 6.2 min or 61.1°C, 3.5 min) products resulted in at least 5-log reductions in NCDV at the lower temperature–longer times of USDA–approved *Salmonella* pasteurisation processes. In addition, a > 5-log reduction was demonstrated at the HTST of USDA-approved pasteurisation processes, in only fortified and plain egg yolk products. For the salted and sugared egg yolk products at 63.3°C, an additional 0.65 and 1.6 min of treatment, respectively, were necessary to inactivate 5 log of NCDV.

The OIE Terrestrial animal health code for NCD (OIE 2021b), in Article 10.9.20., recommends the same treatments as EU-prescribed treatments, except for the heat treatment of dried egg white (while for dried egg white a heat treatment at 57°C for 54 h is prescribed by the EU, a heat treatment at 57°C for 50.4 hours is prescribed by the OIE).

The ELS findings confirm that heat treatment seems the most appropriate inactivation method for NCDV in eggs (ILSI, 2009).

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6 List of abbreviations

AHS(V) African horse sickness (virus)

ASF(V) African swine fever (virus)

CBPP Contagious bovine pleuropneumonia

CCPP Contagious caprine pleuropneumonia

CSF(V) Classical swine fever (virus)

ELS Extensive literature search

FMD(V) Foot-and-mouth disease (virus)

LSD(V) Lumpy skin disease (virus)

PFU Plaque forming unit

PPR(V) Peste des petits ruminants (virus)

RP(V) Rinderpest (virus)

RVF(V) Rift valley fever (virus)

SPGP(V) Sheep pox goat pox (virus)

Appendix A – Extensive literature search protocol

Review question

The specific objective of this ELS is to answer the following question:

'Which, if any, of the 14 pathogens of interest, as described in the current mandate, can be found:
i) in the meat (including blood), milk, casings, and eggs (`products of animal origin`) obtained from an infected animal, or
ii) in other products not of animal origin (´other materials`), originating from the protection zone,
after these products are treated with the treatments described in Annexes VII and VIII of the Delegated Act, respectively?'

Criteria for including studies

To meet the above stated objective of the ELS , the study inclusion criteria are based on the Population – Intervention – Comparison – Outcome (PICO) strategy:

Population	Products: i. Meat (including blood), casings, milk and eggs (`products of animal origin`)[1] ii. Feed materials of plant origin and straw ('other materials') With presence of the causing pathogen of: FMD/Rinderpest/RVFV/LSD/CBPP/SPGP/PPR/CCPP/AHS/Glanders/CSF/ASF/HPAI/NCD[2]
Intervention	Treatment to inactivate the pathogen
Comparison	NA
Outcome	Pathogen inactivation (as measured by diagnostic method): log titer reduction. length of survival

[1] Originating from animals of listed species (Commission implementing Regulation (EU) 2018/1882 of 3 December 2018).
[2] The products to consider will depend on the pathogen.

The following criteria for inclusion will be used to select studies to be included in the review:

i. Studies on products of animal origin and/or other materials infected with one of the pathogens of interest
ii. Studies evaluating a treatment to inactivate the pathogen of interest
iii. Studies from all countries in the world are eligible
iv. No timeframe restriction
v. Studies published in either English, Spanish, German, Dutch, Portuguese or French
vi. Original studies

Exclusion criteria

The references will be excluded from the ELR if they meet one or more of following criteria:

i. Published after search date (07/04/2021)

ii. Published in another language than English, Spanish, German, Dutch, Portuguese or French.

iii. Review papers. However, original studies included in the review papers complying with the inclusion/exclusion criteria will be included.

Information sources

Electronic databases

P95 will conduct a literature search in MEDLINE (via PubMed) to obtain peer-reviewed, scientific publications related to the ELS.

Reference checking and hand searching

The reference list of relevant studies retrieved from the electronic database search will be hand searched to identify additional studies.

Search strategy

The following search strategy will be used in PubMed:

#	Search string	# of results
1	meat OR casings OR milk OR egg OR eggs OR carcass OR carcasses OR tissue OR blood OR "product of animal origin" OR "products of animal origin" OR "animal origin product" OR "animal origin products" OR offal OR feed OR "feed material" OR "material of plant origin" OR straw OR litter OR bedding	8,341,278
2	#1 AND "foot and mouth disease" OR "hoof and mouth disease" OR rinderpest OR morbillivirus OR "rift valley fever" OR phlebovirus OR "lumpy skin disease" OR capripoxvirus OR capripox OR "mycoplasma mycoides" OR "contagious bovine pleuropneumonia" OR "sheep pox and goat pox" OR "sheep and goat pox" OR sheeppox OR goatpox OR "peste des petits ruminants" OR "contagious caprine pleuropneumonia" OR "mycoplasma capricolum" OR "african horse sickness" OR orbivirus OR glanders OR "burkholderia mallei" OR "classical swine fever" OR pestivirus OR "hog cholera" OR "african swine fever" OR asfivirus OR "highly pathogenic avian influenza" OR H5N1 OR H7N9 OR H5N8 OR H5N5 OR "newcastle disease" OR "avian paramyxovirus" OR orthoavulavirus	21,631
3	#2 AND heat OR heating OR thermal OR temperature OR heated OR UHT OR "ultra high temperature" OR "high temperature short time pasteurisation" OR HTST OR pasteurization OR pasteurization OR pasteurized OR pasteurized OR extrusion OR boiling OR boil OR boiled OR bleached OR fermentation OR fermented OR maturation OR matured OR drying OR dry OR dried OR "water activity" OR salt OR salted OR salting OR "sodium chloride" OR NaCl OR phosphate OR bleaching OR bleached OR bleach OR package OR packaged OR stored OR store OR storing OR bales OR disinfect OR disinfected OR disinfection OR disinfectant OR spray OR sprayed OR spraying OR irradiation OR ultraviolet OR ozone OR "high pressure" OR clean OR cleaned OR cleaning OR wash OR washed OR washing OR steam OR steamed OR steaming OR "saturated brine" OR brine OR Na2HPO4 OR Na3PO4 OR mature OR burn OR burned OR burning OR "food process" OR "food processes" OR "food processing" OR decontamination OR decontaminated OR decontaminate OR inactivation OR destruction	3,600

4	#3 AND "log reduction in titer" OR "log reduction in titers" OR "reduction in titer" OR "reduction in titers" OR "titer reduction" OR logs OR titer OR count OR survival OR infectivity OR concentration OR reduced concentration	3,207

Review methods

Selection of the studies

In the first review phase, the resulting list of references will be exported to Rayyan[6] to proceed with the title, abstract and key words screening and study selection.

To decrease the risk of selection bias, two P95 reviewers will independently review the list of references obtained by screening the titles/abstracts to identify studies that fulfil the above-mentioned inclusion criteria. Discrepancies will be discussed, and if not resolved, a third reviewer will make the final decision.

The complete selection process will be documented in an Endnote file, containing folders that reflect the selection criteria.

Data extraction

In the second review phase, full papers for all selected studies will be assessed for eligibility. Data from the eligible full-text papers identified will then be extracted by two/three reviewers using a standardized extraction form in MS Excel (see Annex 1) to ensure that all relevant data are extracted systematically. In addition, the sections of the pdf manuscript from where data are collected will be highlighted.

Analysis and reporting

During the selection process, the results of the literature search will be imported into Endnote where a clear track of the selection process will be maintained, and the flow of publications will be noted. Based on these numbers, a flowchart of the studies selected in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines will be prepared for use in the report.

The final report will consist of an update of the two tables in the Delegated Act Annex VII and VIII, as well as a narrative review summarizing the key findings that brought to the conclusions presented in the tables. The report will also include the results obtained for alternative treatments.

[6] https://rayyan.qcri.org