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# Literature review and qualitative risk assessment on the role of feed materials in African Swine Fever Virus transmission

## *Literaturübersicht und qualitative Risikobewertung zur Rolle von Futtermitteln bei der Übertragung des Virus der Afrikanischen Schweinepest*

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**Summary**

In Germany, the African swine fever virus (ASFV) was first detected in a wild boar in September 2020 in the German federal state of Brandenburg near the border to Poland. It has since been spreading with currently more than 3,960 confirmed cases in wild boar and five outbreaks in domestic pigs (TSIS 2022). Feed has been mentioned as a possible source of ASFV transmission into domestic pig farms and stability of ASFV in feed and bedding has been defined as a knowledge gap on European and global level.
A literature study was performed to examine the role different categories of feed materials could play for virus transmission to domestic pigs considering the impact of feed processing, transport and storage. The available information was used to estimate the risk of individual feed groups for domestic pigs with regard to their virus transmission probability.
For processed by-products, e.g. grains, extraction meals and compound feed, it can be assumed that ASFV will become most likely inactivated during processing. Although recontamination with virus after the manufacturing process may occur, this scenario is assumed to be unlikely under general hygiene and HACCP principles.
Due to the infectivity of ASFV and its resistance to environmental factors, virus transmission into domestic pig farms cannot be excluded for certain feed categories such as unprocessed, directly-fed feed materials. In contrast, processed feed probably does not play a role in ASFV transmission. According to literature, ASFV may be also retained in blood meal and spray-dried plasma after processing in the very unlikely case that blood or plasma from highly-infectious domestic pigs is used.
Overall, the literature review pointed out that future research is needed to generate data on the main factors influencing the survival and transmission of ASFV in feed material for domestic pigs during processing and storage.
**Keywords:** ASF, contamination, feed material, pig, processing

**Zusammenfassung**

Das Virus der Afrikanischen Schweinepest (ASPV) wurde bei Wildschweinen in Deutschland erstmalig im September 2020 im Bundesland Brandenburg nahe der polnischen Grenze nachgewiesen. Seitdem breitet es sich weiter aus mit zurzeit mehr als 3.960 bestätigten Fällen in Wildschweinen und fünf Ausbrüchen in Hausschweinen (TSIS 2022). Als eine mögliche Übertragungsquelle von ASPV auf Schweine wurden Futtermittel benannt und die Stabilität des ASPV in Futtermitteln und Einstreumaterialien wurde als Wissenslücke auf europäischer und globaler Ebene definiert. Aus gegebenem Anlass wurden die in der Literatur verfügbaren Informationen zur Rolle von Futtermitteln für Schweine bei der Virusübertragung unter Einbezug des Einflusses von Verarbeitung, Transport und Lagerung zusammengestellt. Die verfügbaren Informationen wurden genutzt, um das Risiko einzelner Futtermittelgruppen für Schweine im Hinblick auf ihre Virusübertragungswahrscheinlichkeit abzuschätzen.

Für verarbeitete Nebenprodukte, Getreide, Extraktionsschrote und Mischfuttermittel ist von einer weitestgehenden Inaktivierung von ASPV während der Verarbeitung auszugehen. Hier kann lediglich durch Rekontamination mit ASPV nach dem Herstellungsprozess ein Infektionsrisiko für Schweine bestehen. Bei Einhaltung allgemein geltender Hygiene- und HACCP-Prinzipien ist dies jedoch unwahrscheinlich.
Aufgrund der Widerstandsfähigkeit von ASPV-Partikeln gegenüber äußeren Einflüssen und der Infektiosität des ASPV ist eine Übertragung der Viren in Schweinebestände durch bestimmte Futtermittelkategorien wie zum Beispiel Futtermittel, die keiner weiteren Behandlung unterzogen und direkt verfüttert werden, nicht vollständig auszuschließen. Futtermittel hingegen, die technologisch verarbeitet werden, spielen vermutlich keine Rolle bei der ASPV-Übertragung.
Untersuchungen zeigen, dass infolge der Herstellung von Blutmehl und sprühgetrocknetem Plasma aus Blut oder Plasma hochinfizierter Schweine eine infektiöse ASPV-Dosis erhalten bleiben kann.
Die Literaturübersicht hat gezeigt, dass künftige Forschungsarbeiten erforderlich sind, mit deren Hilfe Daten zu den Einflussfaktoren auf das Überleben und die Übertragung von ASPV in Futtermittel-Ausgangserzeugnissen für Hausschweine während der Verarbeitung und Lagerung erhoben werden.
**Schlüsselwörter:** Afrikanische Schweinepest, Kontamination, Futtermittel, Schweine, Verarbeitung

## Introduction

African Swine Fever (ASF) is caused by the African swine fever virus (ASFV), a large double-stranded DNA virus of the Asfarviridae family (Gaudreault et al. 2020). It is endemic in many countries of sub-Saharan Africa where it is involved in a sylvatic cycle among warthogs and soft ticks of the genus *Ornithodoros*. In these reservoir hosts, ASFV does not cause overt disease (Sanchez-Vizcaino et al. 2015). However, in naïve wild boar and domestic pigs, ASFV infection leads to severe haemorrhagic disease with high fatality (Howey et al. 2013). The ASFV strains involved in the current epidemic belong to the p72 genotype II with a high mortality rate for domestic pigs, leading to substantial economic losses in domestic pig husbandry (Brown et al. 2021). The first case in Germany was identified in September 2020 in the border region to Poland (Sauter-Louis et al. 2020). Since then, German federal states of Brandenburg, Saxony and Mecklenburg-Western Pomerania together reported more than 3,960 ASF cases in wild boar and four outbreaks in domestic pigs. Very recently, a single outbreak was reported from a free-ranging pig farm in the federal state of Baden-Württemberg (TSIS 2022).

African swine fever virus shows a high tenacity which may be the reason for long-lasting cycles in the wild boar population in Europe. The virus is relatively temperature-resistant, heating at 56°C for 70 minutes or 60°C for 20 minutes is needed to inactivate the virus (Friedrich-Loeffler-Institut 2020). At room temperature, ASFV can remain contagious around 18 months and up to 6 years at 4°C, while at 37°C the virus stays infectious in urine and faeces for four and five days, respectively (Blome et al. 2020). Humidity, a protein-rich environment and lower ambient temperature favour the virus survival. African swine fever virus keeps its infectivity at a pH range between 3 and 13.4; however no time frame was mentioned (Friedrich-Loeffler-Institut 2020).

Transmission of ASFV can occur through direct contact between infectious and healthy animals (wild boar as well as domestic pigs). However, indirect transmission through carcasses, contaminated blood, excretions or pork-containing food waste as well as virus-infected surfaces (tools, vehicles, shoes/clothing etc.) are important routes of infection, particularly in the wild boar population (Pepin et al. 2020).

In the absence of competent tick vectors in Western and Central Europe, the transmission occurs mainly via the oro-nasal route where ASFV reaches the animal organism via the tonsils as primary place of replication (Niederwerder et al. 2019).

After an incubation period of two to seven days, infected animals develop high fever and severe unspecific clinical signs such as dullness, motion disorders, diarrhea, and highly increased respiratory rate. These signs are accompanied with a significant reduction of feed intake. Within seven to ten days, pigs die from the infection. The case fatality rate is almost 100% under experimental conditions (Gabriel et al. 2011, Friedrich-Loeffler-Institut 2020).

In 2007, ASFV was introduced to Georgia in the Caucasus presumably by ship transports, i.e. improperly disposed and contaminated catering waste in the Black Sea harbour of Poti (Rowlands et al. 2008), and has spread in the Trans-Caucasian region and Europe since then. As most domestic pigs are traditionally kept in free range in Georgia, access to infected food waste material is likely. There is evidence in Russia that kitchen and food waste caused disease outbreaks in domestic pig stocks (Rowlands et al. 2008, Gogin et al. 2013). With the implementation of Regulation (EC) No 1069/2009 (health rules concerning animal by-products [and derived products] not intended for human consumption), the EU prohibited swill feeding of farm animals. Thus, this transmission route is unlikely in domestic pigs on commercial farms in Germany, if legal provisions are complied with.

In principle, the oral route of infection with ASFV requires substantially more infectious particles compared to parenteral routes. In the case of the moderately virulent ASFV strain "DR´79", McVicar

calculated a factor of 140,000:1 and concluded that $10^4$ tissue culture infectious dose 50 ($TCID_{50}$) led to ASF-associated clinical signs in all domestic pigs after oral uptake (McVicar 1984). In contrast, for the highly virulent strain "Lisbon '60" the difference of the effective dose was much smaller between the oral and the parenteral route. Similar results were obtained with ASFV isolate "Malawi Lil-20/1" (McVicar 1984, Howey et al. 2013). However, for domestic pigs with an impaired health status, a lower dose was sufficient (Howey et al. 2013).

Niederwerder et al. (2019) investigated the minimal infectious dose for the ASFV-isolate "Georgia 2007/1" under natural intake conditions. Here, the virus inoculum was mixed either with drinking water or with a small amount of regular complete feed. A single dose of $10^4$ $TCID_{50}$/animal was sufficient to infect one hundred percent of animals in the group when drinking water was used to deliver the virus. However, none of the doses between $10^0$ $TCID_{50}$ and $10^8$ $TCID_{50}$ led to a complete infection rate when the virus particles were fed once via feed. On this basis, the dose to infect fifty percent of the animals ($ID_{50}$) of $10^{1.0}$ was calculated for the intake via drinking water and $10^{6.8}$ for the intake via complete feed. This is in contrast to investigations by Blázquez and colleagues. They mixed non-pelleted commercial feed with ASFV contaminated domestic pig plasma. Both infectious doses tested ($10^{4.3}$ and $10^{5.0}$ $TCID_{50}$/pig/day) did not lead to disease in domestic pigs (Blázquez et al. 2020).

## ASFV entry routes and influence of feed processing on virus infectiousness

Crops, hay and straw from commercial trade are rarely contaminated with ASFV (EC 2020). Still, there is a high uncertainty concerning the possible infection path through feed in affected areas, also particularly because of the discussions of "transmission of ASFV via feed" in the media. The role of diverse feed materials, drinking water and contaminated objects are matters of controversial discussions, and all named vectors were linked to ASFV transmission (Olsevskis et al. 2016). In a current opinion, the European Food Safety Authority (EFSA) assessed the risk of ASFV transmission through different matrices and classified the overall risk as low (EFSA 2021).

The use of feed obtained from fields with prior dwelling of infected wild boar, could lead to the introduction of ASFV into domestic pig holdings. In the present paper, the risk of ASFV infection in domestic pigs through different feed materials frequently used for domestic pig feeding has been assessed. Therefore, the probability of feed materials to get in contact with wild boar or its excretions on fields were considered and furthermore, different feed processing steps that can inactivate ASFV in the feed material. Based on available literature, a decision tree was generated to allow a qualitative risk assessment for ASFV in available feed materials for domestic pigs in endemic areas. Chemical risk mitigation strategies, like usage of antimicrobial active substances (e.g. organic acids, aqueous formaldehyde) in contaminated feed ingredients, are beyond the focus of this article but were reviewed elsewhere (Niederwerder 2021).

## Results and Discussion

### Feed material categories

#### *Forages and roughage*

In Latvia and Estonia, most disease outbreaks in domestic pigs occurred in summer. This was associated with feeding potentially contaminated forages (Olsevskis et al. 2016). Forages and roughages are defined as alfalfa, alfalfa green meal, clover, clover green meal, forage (obtained from forage plants), green meal, hay, silage, cereal straw and root vegetables for foraging. Feeding of these materials is required in organic domestic pig production according to Regulation (EC) No 834/2007. In conventional pig farming, roughage (hay, straw) is used for pregnant sows to meet the minimum supply of crude fibre specified in the German Animal Welfare Ordinance (TierSchNutztVO). In addition, hay and straw serves as enrichment material, which enables oral intake of larger amounts of plant material. Alfalfa, clover and grass fed to pigs in the ration is mainly dried and ground. During the technical process the forage is dried with hot air reaching a temperature up to 200°C, before grinding and further processing. Corn can be fed directly to pigs as so-called green corn made from freshly cut plant parts that were grown above ground. Hay and straw dried on the field or under aeration is usually pressed into bales and stored. Residual moisture can result in microbial fermentation. Consequently, reheating processes may occur during the storage (Jeroch et al. 1993).

Systematic studies on influence of storage and drying conditions on ASFV-infected forages and roughages are not available. However, the German Swine Fever Regulation (Schweinepest-Verordnung/SchwPestV 1988, last amended in November 2020) allows straw harvested in regions at risk of ASFV only as feed material after storage for 6 months secured from wild boar or after heat treatment for at least 30 minutes at 70°C.

In organic domestic pig farming, roughage such as clover grass or corn silage (whole plant silage) are also used as feed (Werner and Sundrum 2008). For latter, during the ensiling process, the plant material acidifies to a pH of 4–5, depending on the starting material and the dry matter. The low pH in combination with the usual storage time should be sufficient to inactivate the ASFV. From this point of view, silages presumably pose a low risk for ASFV transmission (EFSA 2021). An overview of risk assessment factors assumed to have impacted of the ASFV transmission from forages and roughages is shown in Table 1. The table indicate that caution should be exercised especially when fresh forages is fed to domestic pigs.

#### *Cereals and cereal by-products*

Cereals are used extensively in pig feeding for compound feed production. Wheat has by far the greatest importance, followed by barley and corn, while triticale and rye usually play a minor role (Statista 2022). In wheat, rye and triticale, threshing separates the glume from the grain. However, in barley and oats the glume is firmly attached to the grain and is mainly processed and fed along with it. Thus, a potentially virus-contaminated surface is not removed. Feeding of fresh grain to domestic pigs is not recommended due to an increased activity of plant-derived enzymes and the risk of gastrointestinal problems. Feeding of sprouted grains to pigs is not advised as well due to lower vitamin E levels

and the risk of mycotoxin contamination (Jeroch et al. 1993). Usually, cereals are dried post-harvest and stored in dry air to enable seed maturation. However, durable and spoilage-free storage is only possible at a dry matter content of at least 86% and miscellaneous impurities of less than 1% (Jeroch et al. 1993). Therefore, to provide stable storage conditions, cereals are processed by various conservation procedures (drying, airtight storage, cooling, chemical conservation, or ensiling). Fischer et al. studied the influence of drying on the ASFV infectivity in contaminated cereals (Fischer et al. 2020). Different cereal varieties were either left untreated or experimentally contaminated with ASFV (Isolate "Armenia08", $9x10^5$ $TCID_{50}$) and subsequently processed using common methods storage and heating. Analyses showed that after a two-hour storage at room temperature, virus DNA was detectable. However, the hemagglutination test, as a measure of infectivity, was negative for all samples. Heat-treated samples (one hour at 40–75°C) experimentally contaminated with ASFV gave the same results. The authors discussed the hygroscopic effect as a cause of virus inactivation. Despite certain limitations regarding the test systems used and a limited predictive power of in vitro experiments, the authors conclude that there is a low probability of ASFV transmission if cereals are stored dry and at a minimum of room temperature (20°C) for 24 hours before being fed to domestic pigs (Fischer et al. 2020). However, as a complete virus inactivation cannot be guaranteed following this protocol, cereals harvested in ASF risk areas should be fed only after treatment appropriate to inactivate the virus (as specified in the Annex of Commission Implementing Decision 2014/709/EU).

On the field, corn kernels are virus-protected by husk leaves. If corn cob meal is used, virus contamination of plant parts by wild boar is possible, as these frequently reside in corn fields. The virus transmission probability through wild boar contact is lower for cobs of upright corn plants that are mechanically collected during harvest. Dried Distillers Grains with Solubles (DDGS) is a by-product of alcohol production and is frequently used as part of domestic pig feed ration (Kersten et al. 2010, EC 2020). The procedure is a multi-level process, thus, various temperatures between 30 and 70°C (saccharification step and fermentation) as well as up to about 80–200°C (distillation and concentration steps) are applied. After distillation of the alcohol from the mash, DDGS is produced as a by-product. Fresh DDGS has a dry matter (DM) content of 35–40%, while dried DDGS has a DM content of 86% (compare Table 1).

Since DDGS is produced in a controlled temperature-regulated process, inactivation of any ASF viruses that may be present can be assumed. Therefore, transmission to domestic pigs is estimated to be unlikely. Overall, the likelihood of ASFV transmission through grain feeding in domestic pig is low (see Table 1).

*Expeller, extraction meals*

In domestic pig feeding, expeller and extraction meals made from soy beans, rape seed, and sunflower seed and to a lesser extend linseed are used. ASFV contamination in the field is assumed unlikely due to the protection by hulls (soy beans, rape seed and linseed) or shells (sunflower), respectively. Assuming a presence of ASFV on oil seeds, a strong reduction is likely during the expelling and extraction process. During the extraction process the oil seeds are first cleaned and crushed before being thermally treated (>100°C). The oil extraction is a combination of pressing and treatment with a hexane-oil-mixture. The de-oiled meal is toasted with hot steam (105–110°C) to obtain a solvent free extraction meal (Kersten et al. 2010). In addition to by-products from the German vegetal oil industry, extraction meals for farm animals are imported from main cultivation countries such as the USA, Brazil and Argentina to cover the demand. Two model studies describe ASFV survival in soy extraction meal transported across borders. They show that virus particles are still detectable in ASFV contaminated soy extraction meal after a 30-day storage at mean temperatures of 12.3 or 15°C (Dee et al. 2018, Stoian et al. 2019). Thus, an indirect ASFV transmission through respective feed materials seems possible (Dee et al. 2018). However, this would require high viral loads and is therefore unlikely under typical commercial conditions. It should be noted that the scenarios described above assume that expellers or extraction meals get into direct contact with infected animals or infected material after leaving the oil mill. This scenario is unlikely if industries along the production chain are compliant to stipulated hygiene and storage regulations. The ASFV transmission via expeller and extraction meals into domestic pig herds is therefore considered to be very unlikely. Reinfection is unlikely provided infected animals (wild boar, but also pest rodents) have no access to the feed storage (as summarized in Table 1).

*Tubers and roots*

Root crops (e.g. potatoes, sugar beets, fodder beets, carrots) or their processed products can be components of feed rations for domestic pigs. Potatoes and beets can come in direct contact with ASFV-infected wild boar in the field. Sugar beet processing produces mainly molasses and sugar beet pulp (wet, pressed or dried) as potential feedstuff. After harvesting sugar beets are washed and chopped before the sugar is extracted at temperatures between 73 and 78°C for 60–120 minutes. The by-product beet pulp is usually pressed and ensiled to obtain pressed beet pulp or dried using hot air to produce dried beet pulp. The resulting sugar containing raw juice is then purified and evaporated into thick juice before white sugar and molasses are extracted in a multi-stage boiling process. Due to the thermal treatment of sugar beets to extract sugar beet pulp and sugar, it is assumed that this leads to inactivation of potentially present ASFV particles. Therefore, we assume that virus transmission to domestic pigs can be excluded.

If fodder beets are fed directly to domestic pigs the situation has to be assessed differently. Here, roots are merely cleaned and chopped and then used. Fodder beets are especially suitable for low-bearing sows. The preservative storage of fodder beets is done in storage halls at an optimum temperature of 1–5°C and 90% humidity (Jeroch et al. 1993). However, no thermal treatment of roots takes place; therefore, an ASFV inactivation of contaminated roots cannot be expected. Because raw potatoes contain ingredients that inhibit feed intake and performance of domestic pigs, potatoes are thermally treated before feeding (steaming at 90–100°C for more than 20 minutes) to break down the potato starch and to inactivate inhibitors. This thermal treatment would also decontaminate ASFV. Overall, an ASFV transmission to domestic pigs via feeding of processed

root crops products is assumed to be unlikely if the feeding stuff is thermally treated (Table 1).

Overall, cautions should be taken before feeding fresh fodder beets directly to domestic pigs.

*Grain legumes (peas, horse beans, lupines)*

Native grain legumes are used as protein rich animal feed. Due to the increase in use of protein-rich feed in livestock diets, especially in the regional organic farming, growing of grain legumes is gaining importance in Germany. However, antinutritive ingredients limit their use (Bernsmann et al. 2011). Various processing can be applied to reduce these antinutritive properties and thereby elevate the feed value. Typically, these include mechanical treatment (seed peeling) and thermal/hydrothermal treatment (autoclaving, expanding, extruding, toasting or steam pelleting) (Freitag 2006). These mechanical or heating procedures are suitable to reduce an existing virus load. Since a hull protects sweet lupines, horse beans and peas anyway, the probability of direct contact of the seeds with ASFV and subsequent transmission to domestic pigs with the feed is seen as low (as summarized in Table 1).

*Animal protein, blood and plasma products*

Blood meal or spray-dried plasma (SDP) derived from coagulated blood of slaughtered animals is used in compound feed for domestic pigs, in particular for weaned piglets. Both feed materials are characterized by a very high protein level being optimal for virus propagation. The processing steps to obtain blood meal include boiling the starting product for at least three hours or chemical conservation by acids and salts. Subsequently, the product is rolled or spray-dried, respectively (Jeroch et al. 1993). For the production of SDP, the cellular blood fraction is separated by centrifugation using an anticoagulant. The plasma is then concentrated by vacuum evaporation, by filtration with inverse osmotic membrane or by ultra-centrifugation and subsequently dried using spray technology. During the spray drying, plasma proteins are only shortly exposed to high temperatures, which prevents protein denaturation and preserves the biological activity (Torralardona 2010). Gerber et al. demonstrated that the spray-drying processing steps with typical industrial temperatures of 166°C (inlet) and 80°C (outlet), a flow-rate of 820 ml/hour and a pressure of 0.1 MPa were sufficient to inactivate porcine epidemic diarrhea virus (PEDV) experimentally added to porcine blood (Gerber et al. 2014). The authors conclude that SDP, produced under commercial conditions, does not pose a significant risk of PEDV transmission. In a review by Blázquez et al. (2020), a depletion rate of various viruses by commercial spray drying technology was compared. A 4-log reduction (99.99%) of ASFV could be determined after spray-drying. Furthermore, Fischer et al. (2021) showed that storage of contaminated spiked SDP for two week at room temperature completely inactivated ASFV. However, further studies would be useful. If blood from ASF-infected domestic pigs is used for the production of blood meal or SDP, sufficient inactivation of ASFV might not occur due to the short-term thermal drying process. However, this is not realistic under normal conditions. ASFV-infected domestic pigs are completely prohibited from the meat processing chain and respective material is not allowed as feed material. Moreover, transport of domestic pigs or swine animal by-products from ASFV high risk areas is strongly regulated. Therefore, it is not anticipated that blood products contaminated with ASFV will enter domestic pig feed on a significant scale (see Table 1).

*Compound feed*

ASFV is inactivated at temperatures above 50°C over a longer period of time. Industrial feed technology applies various thermal and physical techniques to provide necessary nutrients in compound feed, to inactivate antinutritives and to reduce the germ count through hygienization (Bernsmann et al. 2011). The most important procedures in compound feed production are pelleting, extrusion and expansion. As a conditioning procedure, all these processes are preceded by steaming (about 130–170°C at 2.5–8 bar) (Kersten et al. 2010). ASFV inactivation through pelleting can be assumed if the pellets leave the press at a temperature up to 90°C. Here, the exposure time is up to 30 seconds. The expander treatment occurs at temperatures up to 130°C. However, the processing time is only a few seconds. During extruding, temperatures of 100–180°C are reached for 30–150 seconds. Therefore, ASFV inactivation can be assumed using thermal/physical processing techniques in compound feed production and that transmission to domestic pigs seems unlikely. Dee et al. (2018) could detect ASFV particles after a transatlantic transport simulation of experimentally (4–14°C for 37 days or 10–20°C for 30 days) contaminated complete feed. It has to be kept in mind, however, that the dose for contamination was exceptionally high and would not reflect most field conditions. In case of compound feed, an ASFV transmission cannot be excluded due to recontamination after production.

It is therefore essential to prevent recontamination of compound feed with ASFV-containing material. Dry storage would help mitigating residual risks (see Fischer et al. 2021).

## Conclusions

The most important factor for inactivation of virus particles during processing is a sufficiently high process temperature, and many of the feed storage and processing methods apply effective heating; therefore, in summary, feed materials play at most a minor role in ASFV transmission. However, such risk must be considered in a differentiated manner based on the type of feed used (Table 1).

Transmission through compound feed for domestic pigs can be ruled out, provided that it was produced and transported in compliance with industry standards of hygiene and storage conditions. The physical and thermal processing procedures should lead to an inactivation of virus particles. This also applies to extraction meals commonly used in feeding.

An ASFV transmission through cereals used for feeding is considered to be unlikely since, either protection by glume or subsequent storage and drying reduce the transmission risk substantially. However, the risk cannot be excluded completely. Therefore, for raw materials from affected areas appropriate protective measures need to be taken. ASFV transmission through by-products of the processing of field crops (DDGS, molasses etc.) is considered very unlikely due to prevalent conditions. However, for raw or insufficiently heated crops (e.g. fodder beets) an ASFV transmission cannot be excluded if these materials are used directly in domestic

**TABLE 1:** *Assessment of the probability of ASFV transmission from feed material to domestic pigs through protective seed coverings mechanisms, feed processing and storage conditions using risk factors*

| Feed material | Protective seed coverings | Processing | Storage conditions | ASFV transmission risk factor | Probability of ASFV transmission |
|---|---|---|---|---|---|
| **Forages and roughages** | | | | | |
| forages (alfalfa, clover, grass, green corn) | No (3) | –/D (3/1) | Few, days, OT (3) | 2 | cannot be excluded |
| roughages (hay, straw) | No (3) | D (1) | Several month, OT/ RT (1) | 1 | Unlikely[1] |
| silage | No (3) | S (2) | At least 90 days; OT (1) | 2 | Cannot be excluded* |
| **Grain and grain by-products** | | | | | |
| grains | Partly (2) | D/CP/S (1) | RT (1) | 1 | Unlikely[3] |
| corn | Yes (1) | D (1) | RT[2] (1) | 1 | Unlikely |
| Corn cob meal | Partly (2) | S (2) | RT (1) | 1,5 | Unlikely |
| DDGS | Partly (2) | H & D (1+) | RT (1) | 1 | (Very) Unlikely |
| expeller, extraction meals | Yes (1) | H & CP (1+) | RT (1) | 1 | (Very) Unlikely[4] |
| **Tubers und roots** | | | | | |
| fodder beet | No (3) | S (2) | Several month, 4°C, 90% AH (3) | 3 | Likely |
| sugar beet pulp | No (3) | H (1+) | RT (2) | 1 | Unlikely |
| potatoes | No (3) | H & S[2] (1) | RT (1) | 1,5 | Unlikely |
| **Others** | | | | | |
| grain legumes (peas, horse beans, lupines) | Yes (1) | H (1) | RT (2) | 1 | Unlikely |
| blood and plasma products | n/a (2) | H & CP (1) | 4°C (2) RT (1)[§] | 2 | Cannot be excluded |
| compound feed | Partly[5] (2) | H (1) | Several weeks, RT (1) | 1 | Unlikely |

–: none; D: drying; S: ensiling; CP: chemical preservation; C: cooling; H: heat; n/a: not applicable; RT: room temperature (ca. 18–20 °C[#]); OT: outdoor temperature (7.4–10.5°C[+]); AH: air humidity

[1] A transmission is very unlikely if the feed has been stored secured from wild boar for more than six months

[2] Sealed airtight

[3] A transmission is very unlikely if grains with at least 86% dry matter was stored on a cool and dry place for at least 24 hours before feeding (Fischer et al. 2020)

[4] In the case of feed materials that have been stored at temperatures of 12–15°C during transport, transmission is very unlikely

[5] mechanical protection same as raw material (e.g. corn, grain)

* no data available

[+] https://de.statista.com/statistik/daten/studie/914891/umfrage/durchschnittstemperatur-in-deutschland/

[#] Kersten et al. 2010

[§] Fischer et al. 2021

pig feeding. Owing to the short-term thermal treatment during production, blood and plasma products used for weaned piglets can pose an ASFV transmission risk if contaminated starting material was used for production. Prevailing animal health regulations in the EU mean to prevent this scenario, but it could have a higher significance for third country imports.

Roughage, which is used primarily in organic domestic pig farming as feed, but also in conventional farms, for example as bedding material, is usually not subjected to any further treatment. Therefore, contaminated material can remain infectious long after harvesting. However, the provisions of the Swine Fever Regulation in Germany (Schweinepest-Verordnung) regarding the storage conditions minimize this risk. In grass or corn silage, the pH values usually achieved in the ensiling process which is assumed to lead to inactivation of ASFV. However, experimental data are missing and should be systematically collected to assess the role of silage as transmission route.

The assessments described here are based mostly on data collected experimentally, which cannot be entirely extrapolated to the complex environment of domestic pig production and feeding. They should be understood as general indications and less as a detailed risk assessment. Moreover, the stability and inactivation of ASFV is largely determined by the surrounding matrix and the amount of virus particle. Generally, it should be noted that storage alone does not sufficiently reduce the viral load in contaminated products. Therefore, the prevention of ASFV recontamination of products is of utmost importance.

Although ASF has gone pandemic for several years, risk arising from feed are quite unknown. Knowledge gaps exists for the survival of ASFV in domestic pigs' feed

during different processing steps and common storage conditions as well as treatment strategies of infected feed for mitigation. The knowledge gaps indicated here well complement those research gaps analysed and published by EFSA (2019). From a risk assessment perspective, data on stability and transmission of ASFV in or respectively from feed are urgently needed, thus we recommend to fill the knowledge gaps as well and as quickly as possible.

## Materials and methods

In order to examine the role of different categories of feed material in ASFV transmission to domestic pigs we performed a literature study to identify those feed materials that are relevant for domestic pig feeding and to collect all available information on ASF virus survival.

Peer-reviewed literature, textbooks, scientific reports by EFSA and Friedrich-Loeffler-Institute [FLI] between 1984 and 2021 in English and German were included. The literature databases Pubmed, OpenAgrar and the library of the Federal Institute of Risk Assessment (BfR) were searched systematically. The aim of the literature search was to identify data on ASPV survival in matrices and typical processing factors for common domestic pig feed materials. Using information on virus survival factors, typical domestic feed processing techniques as well as common storage conditions, mitigation risk factors could be identified which were used to estimate the transmission risk to domestic pigs.

Thereafter, information was summarized for each feed material on I) plant protective coverings, II) the thermal or chemical processing steps, and III) the usual feed storage conditions as most important evaluation criteria in Table 1. Each criterion was evaluated in terms of the likelihood of its ASFV transmission, whereas a value of 1, 2 or 3 were set if transmission is assumed to be either unlikely, not to be excluded or likely. For example, ASFV protection by plant protective coverings were categorized into "unlikely", "not to be excluded" and "likely" if plant compartment intended for pigs' feeding is e.g. completely protected by hulls or glume (1) or partly protected by husk leaves which may open at the end of harvest (2), or if there are no protection at all (3).

For the criterion feed processing, each process step was individually evaluated with a value from 1 "unlikely" to 3 "likely". ASFV transmission risk of feed that went through several processing steps was summarized by an arithmetic mean factor. After the combination of the individual judgments, the general ASFV transmission risk was finally expressed as one factor shown in Table 1.

All results are illustrated in a decision tree (Figure 1). Here, the user can quickly estimate the ASFV transmission risk from feed to domestic pigs by answering a few questions about the plant protective coverings and the performed feed processing steps of the used feed material.

## List of abbreviations

| | |
|---|---|
| ASF | African swine fever |
| ASFV | African swine fever virus |
| DDGS | Dried Distillers Grains with Solubles |
| DM | Dry matter |
| EFSA | European Food Safety Authority |
| FLI | Friedrich-Loeffler-Institute |
| $ID_{50}$ | Dose to infect fifty percent of the animals |
| PEDV | Porcine epidemic diarrhea virus |
| SDP | Spray-dried plasma |
| $TCID_{50}$ | Tissue culture infectious dose 50 |

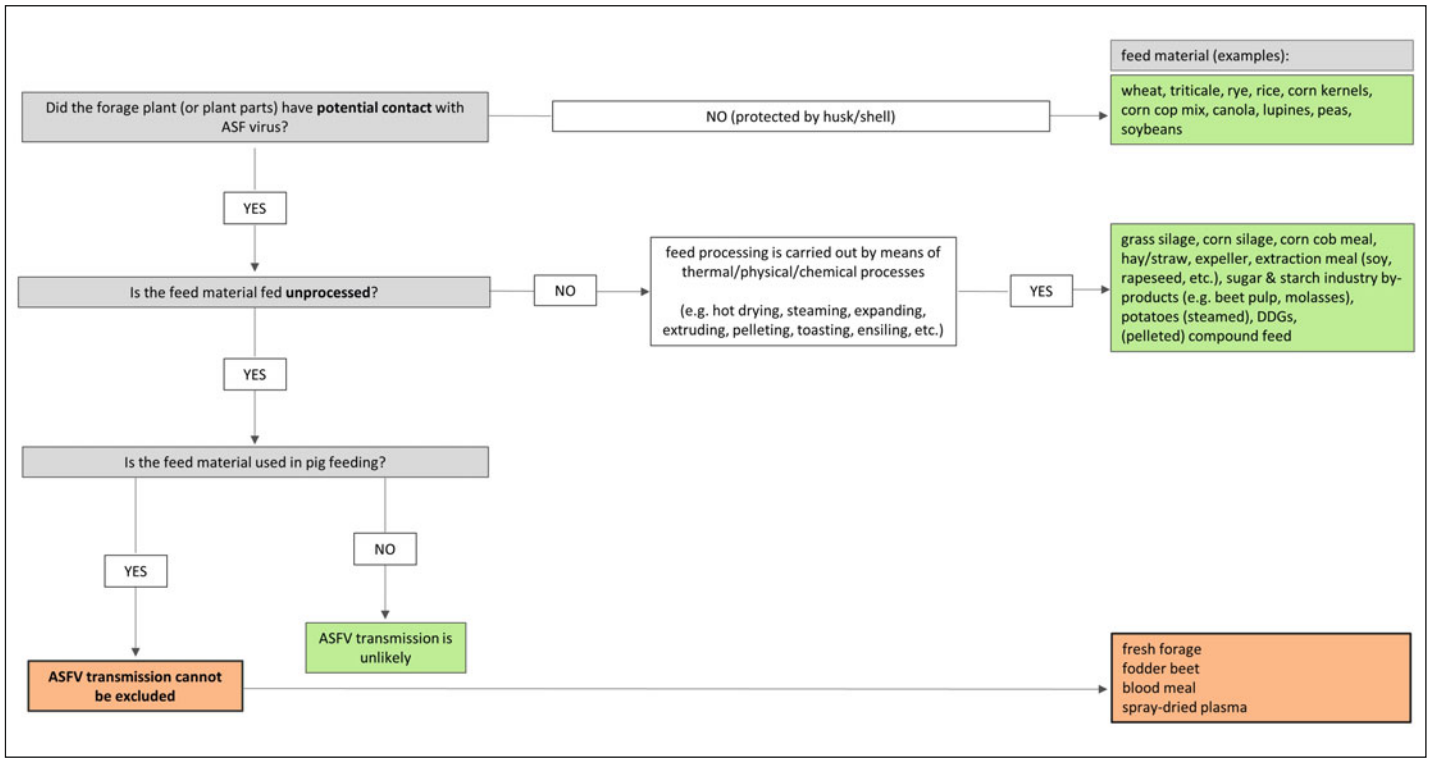


**FIGURE 1:** *Scheme for estimating the possibility of African swine fever virus (ASFV) transmission to domestic pigs through feed. Orange: a transmission of the ASFV cannot be excluded; Green: Transmission of ASFV is unlikely. Copyright: Robert Pieper.*

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## Ethical approval

The authors hereby declare that they have followed the universally accepted guidelines of good scientific practice while preparing the present paper.

## Conflict of interest

The authors hereby declare that they have no proprietary, professional or other personal interests in any product, service and/or company that could have influenced the contents or opinions expressed in this publication.

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## Authors contribution

Conceptualization, writing – original draft preparation: J.K., N.N.B., A-M.E, F.K.
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