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Research priorities to fill knowledge gaps on ASF seasonality that could improve the control of ASF

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Abstract

The European Commission requested EFSA to provide study designs for the investigation of four research domains according to major gaps in knowledge identified by EFSA in a report published in 2019: i) the patterns of seasonality of ASF in wild boar and domestic pigs in the EU; ii) the ASF epidemiology in wild boar; iii) ASF virus (ASFV) survival in the environment and iv) ASF transmission by vectors. In this Scientific Opinion, the first research domain on ASF seasonality is addressed. Therefore, five research objectives were proposed by the working group and broader ASF expert networks, such as ASF stop, ENETWILD, VectorNet, AHAW network and the AHAW Panel Experts. Of the five research objectives, only two were prioritised and elaborated into a general protocol/study design research proposal, namely: 1) to monitor the herd incidence of ASF outbreaks in EU Member States (MS) and 2) to investigate potential (seasonal) risk factors for ASF incursion in domestic pig herds of different herd types and/or size. To monitor the incidence in different pig herd types, it is advised to collect, besides ASF surveillance data, pig population data describing at least the following parameters per farm from the first moment of incursion in an affected MS: the numbers of pigs (e.g. number of breeding pigs sows and boars, weaners and fatteners) and the location and the type of farm (including details on the level of biosecurity implemented on the farm and the outdoor/indoor production). We suggest collecting data from all ASF-affected MS through the SIGMA data model, which was developed for this purpose. To investigate potential risk factors for ASF incursion in domestic pig herds, we suggest a matched case–control design. Such a study design can be run either retrospectively or prospectively. The collected data on the pig herds and the ASF surveillance data in the SIGMA data model can be used to identify case and control farms.

Keywords: African swine fever, epidemiology, risk factor, seasonality, wild boar, domestic pigs, management, prevention

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Summary

This Scientific Opinion follows up on a Scientific Report published in 2019 by EFSA titled 'Research gap analysis on African swine fever'. That Scientific Report provided a review of the most significant ASF knowledge gaps as perceived by the EU Veterinary Services and other stakeholders involved in pig production and wild boar management. The aim of that Scientific Report was to identify gaps in knowledge that could improve short-term ASF risk management once addressed, and to facilitate evidence-informed decision-making on ASF prevention and spread.

Based on this report, the European Commission requested EFSA to provide study designs to investigate four research domains according to major gaps in knowledge identified by EFSA in the report published in 2019: i) the patterns of seasonality of ASF in wild boar and domestic pigs in the EU; ii) ASF epidemiology in wild boar; iii) ASF virus (ASFV) survival in the environment and iv) ASF transmission by vectors. In this Scientific Opinion, the first research domain is addressed, focussing on the identification of the main factors that determine ASF seasonal patterns, as this could support risk managers in the control of ASF.

To address this first ASF research domain on seasonality, five specific research objectives were proposed by the working group and broader ASF expert networks, such as ASF stop, ENETWILD, VectorNet, AHAW network and the AHAW Panel Experts. Of these five research objectives, only two research objectives were prioritised, namely: 1) to monitor the herd incidence of ASF outbreaks in EU Member States, and 2) to investigate potential (seasonal) risk factors for ASF incursion in domestic pig herds of different herd types and/or sizes. The prioritisation was based on the following set of criteria: 1) the impact on ASF management; 2) the feasibility or practicality to carry out the study; 3) the potential implementation of study results in practice; 4) a possible short time frame study (< 1 year); 5) the novelty of the study and 6) if it was a priority for risk managers.

As these two prioritised research objectives are interlinked, one research protocol was developed to monitor both the incidence of ASF and to investigate potential risk factors for ASF incursion in domestic pig herds of different herd types together.

To monitor the incidence in different pig herd types, we advise to collect, besides ASF surveillance data, pig population data describing the following parameters per farm from the first moment of incursion in an affected Member State (MS): the numbers of pigs (e.g. number of breeding pigs sows and boars, weaners and fatteners), the type of farm (including details on the level of biosecurity implemented on the farm and the outdoor/indoor production). We suggest collecting data from all ASF-affected MS through the SIGMA data model, which was developed for this purpose.

To investigate potential risk factors for ASF incursion in domestic pig herds, we suggest a matched case–control design. Such a study design can be run either retrospectively or prospectively. The collected data on the pig herds and the ASF surveillance data in the SIGMA data model can be used to identify case and control farms.

For a retrospective study with a matched case–control design, all outbreaks of ASF until the end of a preset date can be used. However, there is a risk of recall bias associated during interviews in a retrospective study and there is a need for historical data, which might be difficult to obtain. This type of study design, however, does have the advantage of knowing the exact numbers of outbreaks that have occurred up to the end of the preset date, and thus, there is less risk of not obtaining the envisaged sample size.

A prospective case study, on the other hand, will not have constraints such as recall bias. The same potential risk factors as in the retrospective study can be investigated, but with the additional benefit that some information can be collected only through interviews of farmers during farm visits on the included farms, for instance by direct observations by the interviewer. However, there is a risk of not reaching the envisaged sample size when an insufficient number of outbreaks occur during the study period. This could be of particular relevance for commercial pig farms, where less outbreaks have occurred up to now in affected EU Member States. To overcome this, we propose to carry out the prospective study in several MS together. In addition, if only a limited number of outbreak farms with ASF are detected over the first season, it is advisable to carry out the study for two seasons.

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1. Introduction

1.1. Background and Terms of Reference as provided by the requestor

ASF is an infectious lethal disease affecting domestic pigs and wild boar. It can be transmitted via direct animal contact, dissemination of contaminated food or equipment and, in some regions, via biological vectors. This disease has serious economic implications for pig meat production and related sectors, including indirect costs related to trade restrictions. The persistence of the disease in wild boar and the limited number of control measures available represents a challenge for the pig breeding sector in the EU, in particular for the pig farming industry. There is no licensed vaccine or cure despite active ongoing research. From the beginning of 2014 up to now, ASF has been notified in the following EU Member States: Belgium (officially free again since October 1, 2020), Bulgaria, the Czech Republic (free again since March 2019), Estonia, Germany, Greece, Hungary, Latvia, Lithuania, Poland, Romania and Slovakia. The disease has also been reported in Belarus, Moldova, Russia, Serbia and Ukraine, which creates a constant risk for all the Member States bordering with these third countries. The virus strains involved in this ongoing epidemic that started 2007 in Georgia, belong to genotype II. Apart from this, ASF virus strains of genotype I have been present in Italy (Sardinia only) since 1978.

There is knowledge, legislation, scientific, technical, and financial tools in the EU to face properly ASF. In addition, Member States and the Commission are continuously updating the 'Strategic approach to the management of African Swine Fever for the EU' and the related legislation. On 27 August 2019, EFSA published a scientific report titled 'Research gap analysis on African swine fever.[1] The Scientific Report provided a review of the most significant ASF knowledge gaps as perceived by the EU Veterinary Services and other stakeholders involved in pig production and wild boar management. The aim of this scientific report was to improve short-term ASF risk management and to facilitate evidence-informed decision making on ASF prevention and spread. Four major gaps were identified: 'wild boar', 'African swine fever virus (ASFV) survival and transmission', 'biosecurity', and 'surveillance'. The EU is in need to further address some of the major research gaps as identified by EFSA in the Scientific Report, in particular: 'wild boar' and 'ASFV survival and transmission' are crucial to practically implement risk management actions to prevent and control ASF. For this, it is necessary that EFSA complements its previous Scientific Report providing new scientific input and technical assistance to the Commission on those crucial topics identified by the stakeholders as perceived major research gaps and suggests additional studies to fill the knowledge gaps.

1.2. Terms of Reference (TOR)

In accordance with Article 29 of Regulation (EC) No 178/2002, EFSA is requested to provide a Scientific Opinion addressing the following three TORs:

1) Design studies needed to evaluate: (i) the impact of reducing the wild boar population densities in relation to transmission of African swine fever virus (ASFV); (ii) the natural behaviour of wild boar to improve effectiveness of wild boar population management. EFSA should assess feasibility and provide support to design studies, or pilot trials, to verify suitability of new methods for wild boar population control such as immunocontraception (as a tool for population and health control of wild boar) and any other methods, including diverse types of hunting. EFSA should base the Scientific Output or Scientific Technical report on previous EFSA works on this subject and review existing literature, data and information to identify effective methods to reduce and to manage effectively wild boar populations.
2) Design studies needed to understand: (i) the role and impact of vectors, in particular arthropod vectors, in ASF transmission (biological and mechanical); (ii) ASF survival and transmission from contaminated environment and (iii) residual infectivity of buried wild boar carcasses, all this assessing its overall [relative] role in the epidemiology of ASF. EFSA should provide the state of the art of what is known and base the Scientific Output, or Scientific Technical report, on previous EFSA works on this subject. EFSA should review existing literature, data and information to investigate the role of vectors and of the environment to clarify the pathways that facilitate ASF persistence and transmission in affected areas over a number of years.
3) Design studies to investigate patterns of seasonality in wild boar and domestic pigs and identify main factors that determinate these patterns. Provide recommendations in particular

[1] https://www.efsa.europa.eu/en/efsajournal/pub/5811

in relation to risk mitigation options to address these factors, where relevant. EFSA should focus again its analysis on the European experience. EFSA should investigate if seasonal patterns differ across different areas (e.g. temporal spatial increase of already infected areas or seasonality of the so-called 'jumps').

1.3. Interpretation of the Terms of Reference

To facilitate the assessment, the three TORs were interpreted and divided into four general research domains according to their aim:

1) Wild boar management measures with the objective to reduce or stop the spread of ASFV; TOR 1 i) and ii)
2) Potential of ASFV transmission by vectors (including arthropod vectors and scavengers; TOR 2 i)
3) Potential survival of ASFV in the environment; TOR 2 ii) and iii)
4) Possible factors that determine seasonality of ASF in wild boar and/or domestic pig populations; TOR 3

Each of the four research domains is assessed in a separate Scientific Opinion sharing the same methodology to avoid lengthy opinions. This Scientific Opinion answers to research domain 4 (TOR 3), more in particular the assessment identifies and prioritises research that could address the knowledge gaps pertaining **the patterns of seasonality of ASF**. Filling this knowledge gap should contribute to reduce or eradicate ASF in wild boar (WB) populations and pig population. It should be noted that, although research domain 4 (TOR 3) requires the design of studies to investigate seasonal patterns of ASF, it was agreed that apart of research studies, harmonised data acquisition and interpretation play also a key role in addressing the identified knowledge gaps. For instance, knowledge could be generated through the synergetic effect of surveillance data collected in a harmonised way in different MS, as already envisaged by the SIGMA project (EFSA, 2018b). EFSA can have a coordinating role in facilitating this harmonised data collection (i.e. monthly incidence of ASF in different pig herd types).

Besides the harmonised data collection to fill gaps in knowledge about ASF seasonality, EFSA was requested to identify research objectives for primary studies and develop them in research protocols or guidances which could be addressed in a short time frame (preferably within 1 year), to generate information which could support risk managers in their decision-making.

2. Methodologies

2.1. Step 1: Identification of research objectives by working group

1) Brainstorm session during a web conference of the working group to identify possible research objectives for each research domain.
   According to the interpretation of TORs, the following domains of research were identified:

   1) Wild boar management measures with the objective to reduce or stop the spread of ASF;
   2) Potential of ASFV transmission by arthropods;
   3) Potential survival of ASFV in the environment and in buried carcasses;
   4) Possible factors that determine seasonality of ASF in wild boar and/or domestic pig populations.

For each domain of research, specific objectives of research were identified and discussed. For each objective, a brief description was provided, focusing on the main aim of the research regarding ASF management. In addition, keywords defining the research objectives and references showing a similar approach were also included.

2) Contributions by each individual working group member to the results generated during the brainstorm session

A table for each of the four research domains was circulated among the WG members. Each WG member worked separately on-line on the table and proposed all research objectives considered to be of interest for the particular research domains that could be achieved in a relatively short time frame (i.e. less than a year). Thereafter, proposals for each research objective were discussed during a web conference among all WG members. Overlapping research objectives were identified and amended in agreement with the working group. The final version of the table with research objectives was agreed among WG members and prepared to be circulated among networks.

2.2. Step 2: Identification of research priorities by broader networks

An online survey (Annex B) based on the table produced by the WG was distributed to the following networks of experts: ASF stop, ENETWILD, VectorNet, AHAW network and the AHAW Panel Experts. The experts in the networks had 2 weeks to complete the survey online, using the same tables of the research domains and their research objectives developed by the WG.

The WG conducted an analysis of the survey results, identifying new potential objectives and merging overlapping ones. The research objectives selected for the final list, which combined the research objectives suggested by the WG and networks were then prioritised according to procedure explained in Section 2.3.

2.3. Step 3: Prioritisation of research objectives

1) Inclusion criteria: The research objectives proposed by the working group and the different networks were included if they were related to the particular domain of research. In the case of this Scientific Opinion, the inclusion criterion was: Is the research objective related to the possible factors that determine seasonality of ASF in domestic pig populations (Research Domain 4)?

If the answer to this question was 'YES', the research objective was included; if it was 'NO', the research objective was excluded.

In addition, due to potential overlap between research domains, studies potentially dealing with seasonal risk factors pertaining to wild boar or vectors were included in the research domains: 1) Wild boar management measures and 2) Potential of ASFV transmission by arthropods, respectively.

2) Apply scoring criteria for each research objective according to the criteria listed in Table 1.

The working group scored the research objectives proposed by the working group and the different networks using the scoring criteria provided in Table 1. Each member of the WG did a blinded scoring of the different research objectives. The different criteria for ranking the priority of the research objectives and their definitions were discussed and agreed with the requestor of the mandate (the European Commission). For each criterion, a score of either 1 (low), 3 (medium) or 5 (high) was given per research objective according to Table 1.

For each scoring criterion provided, each of the WG members provided a rationale that was discussed afterwards, collectively, during another on-line meeting. Only criterion 6 (priority for the risk managers) was scored by the liaison of the European Commission, who attended the working group. Only a few criteria were not scored by some working group members, but the group scoring was provided by calculating the average of the group, as shown in Annex A and discussed and agreed upon by the whole working group.

Table 1: Criteria for prioritising research objectives

Nr	Criterion	High = 5 points	Medium = 3 points	Low = 1 point
1	Impact on ASF management	The results can have a high impact on the practical management of the disease spread. The topic is part of or is included in one or more of the main strategies for ASF control.	The results can have a medium impact on the practical management of the disease spread. The topic is part of, or includes, one or more of the secondary strategies for ASF control.	The results can have a low impact on the practical management of the disease spread. The topic is not included in any of the main or secondary strategies for ASF control.
2	Feasibility or practicality to carry out the study	Low complexity, methodology fully available	Medium complexity, methodology available but needs further development	High complexity methodology needs to be fully developed
3	Potential implementation of study results in practice	Results can be easily implemented in a short time in the current management of ASF	Results could somehow be implemented in the current management of ASF	Results are not easily implemented in a short time in the current management of ASF

Nr	Criterion	High = 5 points	Medium = 3 points	Low = 1 point
4	Short time frame study possible (1 year)	The study can be completely carried out in 1 year	Part of the study could be done in 1 year	The study cannot be completely carried out in 1 year
5	Novelty: other studies carried out on the same topic?	No previous studies available	Few previous studies available	High number of previous studies available
6	Priority for risk managers	The research gap was perceived as important by the stakeholders (experts and risk managers) in the previous gap analysis; experts and funding are available for the research objective and results will be useful in short term to manage the disease	The research gap was less perceived as important by the stakeholders (experts and risk managers) in the previous gap analysis; experts and funding are less available for the research objective and results will be less useful in short term to manage the disease	The research gap was not perceived as important by the stakeholders (experts and risk managers) in the previous gap analysis; experts and funding are not available for the research objective and results will not be useful in short term to manage the disease

Nr = number.

In order to ensure that proposed ROs (Research Objective) fulfilled the prioritisation requirements mentioned in Table 1, a minimum average score of 3.5 (70% of the maximum score) was agreed a priori by the working group as the cut-off for a research objective to be further developed into a protocol.

The standard deviation and the coefficient of variation were given to show the uncertainty in the initial expert judgements on the criteria for each of the objectives (Annex A). A consensus was reached on the average values of the scores and the working group discussed and agreed with the omission of those proposals that did not reach the score of 3.5.

2.4. Step 4: Development of short research protocols for research priorities

A short research protocol was developed for each of the research objectives that at least scored 3.5/5 points on average (and is therefore considered as a research priority). These protocols could be used by research agencies or funding agencies as a call for research proposals. These protocols should have the following minimum components:

Outline research guidance (3–5 pages per protocol)

- Objectives
  - Research hypotheses
- Introduction
  - Summary of what is known on topic up to date, and identification of the research gap
  - Potential impact on ASF control if the gap of knowledge was to be filled
- Methodology
  - Study design
  - Suggestions for statistical analysis
- Deliverables and milestones

The development of the research protocol has been outsourced to the University of Copenhagen and further discussed and elaborated by the working group. Thereafter, it was reviewed by the Panel on Animal Health and Welfare of EFSA.

3. Assessment

3.1. Step 1: Identification of research objectives by working group

During the web meeting/brainstorming exercise and further consultation by email from the working group, three research objectives were identified by the working group (Table 2).

Table 2: Identification by the working group of research objectives pertaining domain of research 4

Nr.	Research objective	Short description	Key words
1	Study on the seasonal pattern and abundance of potential vectors	The aim is to determine the possible contribution of potential vectors on the seasonality of ASF in affected areas. Different groups of vectors should be included, considering blood and non-blood feeders.	ASF seasonality, vectors
2	Comparative study of monthly ASF herd incidence risk in 2020 between EU member states	Compare the regional monthly ASF incidence in different types of domestic pig farms in affected Member States to understand disease dynamics over time	Monthly incidence, ASF seasonality, herd incidence
3	Harmonised case–control studies in pig herds for potential seasonal risk factor involving several ASF affected countries.	Case–control studies matched on the type of pig farm. This study could focus on commercial pig farms as the number of cases in previous case–control studies was too low to reach the power needed in seasonal risk factor studies. In backyard farms, on the contrary, case–control studies have already been performed with sufficient case farms. By involving several EU MS, it is possible to get a larger sample size and thereby investigate possible risk factors in these types of farms.	ASF seasonality, risk factors

3.2. Step 2: Identification of research objectives by broader expert networks

In addition to the research objectives proposed by the working group (Table 2), the following two research objectives were proposed by broader expert networks (Table 3).

Table 3: Identification by the broader expert networks of additional research objectives pertaining domain of research 4

Nr	Research objective	Short description	Key words
4	Study of ASF seasonal pattern in association with socio-cultural activities	The aim is to assess the possible association between seasonality of ASF in affected areas and human social activities, such as tourism season, festivities, slaughtering period, food festivals, particularly in countries where the domestic pigs breeding is mostly represented by backyard farms.	Social factors, ASF seasonality
5	Defining the spatio-temporal interfaces (and interactions) between WB and pigs in different regions and production systems	Defining the spatio-temporal interfaces (and interactions) between WB and pig in different regions and production systems will help to develop biosecurity practices adapted to specific conditions.	Spatio-temporal interface, WB and pigs, regional variations, production system

Nr: number.

3.3. Step 3: Prioritisation of research objectives

The results of the ranking of research objectives for domain of research 4 are listed in Table 4. From the total of five research objectives identified either by the WG (Table 2) and the broader experts' networks (Table 3), only three research objectives did meet the inclusion criterion, and of only two of them received an average score of 3.5 or more.

Details of the individual scoring and rationales can be found in Annex A.

Table 4: Results of priority ranking of research objectives pertaining domain of research 4

Nr.	Research objective	Inclusion criteria	Average score	Standard deviation	Priority rank
2	Comparative study of monthly ASF herd incidence risk in 2020 between EU member states (# 2. Table 2)	Yes	3.6	1.5	1
3	Harmonised case–control studies in pig herds for seasonal risk factor involving several ASF-affected countries. (# 3. Table 2)	Yes	3.6	1.3	2
4	Study of ASF seasonal pattern in association with socio-cultural activities (#4. Table 3)	Yes	2.8	1.5	3
1	Study on the seasonal pattern and abundance of potential vectors (#1. Table 2)	No	NS	NS	NS
5	Defining the spatio-temporal interfaces (and interactions) between WB and pig in different regions and production systems (#5. Table 3)	No	NS	NS	NS

NS: not scored; Nr: number.

3.4. Step 4: Development of short research protocols to study ASF Seasonality

3.4.1. Research objectives:

Two research objectives were prioritised to address the gap in knowledge on ASF seasonality (Research Domain 4):

1) To monitor the herd incidence of ASF outbreaks in EU Member States
2) To investigate potential risk factors for ASF incursion in domestic pig herds of different herd types and/or size.

As these research objectives are interlinked, they are tackled together in one research protocol. The research protocol follows the structure of the Outline research guidance provided in Section 2.4.

3.4.2. Introduction

African swine fever has been spreading among wild boar on the European continent since it was introduced into Georgia in 2007. From the introduction, the epidemic spread towards north east, and thereafter towards west, entering the EU member states in 2014.

Furthermore, seasonality in numbers of outbreaks of ASF in domestic pig farms shows a clear summer peak (EFSA, 2018, 2020, 2021), while in general, the ASF peak in wild boar is located in autumn. The observed seasonality has been explained by a range of potential contributing factors, such as: longer survival of virus at winter temperatures; pig farming activities, such as harvesting of crops and use of fresh grass during summer; wild boar ecology, such as hierarchical fights, dispersal, fluctuating population size at certain times of year; vector activity and human activity in areas with wild boar. However, it has not yet been possible to explain the difference in seasonality between wild boar and domestic pigs, neither to find a clear explanation for what links the summer peak for domestic pigs to the autumn peak in wild boar.

Cases of ASF in wild boar and outbreaks of ASF in domestic pig farms are registered by the MS and reported to OIE and to the EU (Table 5). This allows to follow the geographical spread of the epidemic, including how the front of the disease has moved with a relatively slow speed, and how focal introductions, most likely human mediated, have occurred from time to time at greater distances from the previously affected areas. The proportion of backyard holdings has been given as an explanation for the different ASF situation between MS (EFSA, 2020). However, outbreaks have been registered in large commercial pig farms in several MS (Nurmoja et al., 2018; Lamberga et al., 2020). In order to better understand the risk of introduction of ASFV to domestic pig farms of varying sizes and types, the outbreak data need to be related to population data regarding pig farms.

Table 5: Number of African swine fever virus genotype II outbreaks in domestic pigs and cases in wild boar notified to the Animal Disease Notification System in 2020

	Number of outbreaks domestic pigs in period	Number of cases in wild boar in period
	1 January 2020—31 December 2020	1 January 2020—31 December 2020
BELGIUM	0	3
BULGARIA	19	533
ESTONIA	0	68
GERMANY	0	403
GREECE	1	0
HUNGARY	0	4,052
LATVIA	3	320
LITHUANIA	3	230
POLAND	103	4,156
ROMANIA	1,060	906
SLOVAKIA	17	388
Total	**1,206**	**11,059**

Several risk factors for ASF incursion have been described, such as free-range pig management, presence of infected farms in the neighbourhood, visitors, swill feeding (Bellini et al., 2016), feed matrices contaminated with ASFV and blood-feeding invertebrates (Olesen et al., 2020). However, often the results are based on outbreaks in a very restricted number of farms (Lamberga et al., 2020). In domestic pig farms, few risk factor studies have previously been completed, revealing varying and sometimes contradicting results (Khomenko et al., 2013; Nurmoja et al., 2018; Boklund et al., 2020). For instance, Khomenko et al. (2013) found an increased risk for small pig farms, while Nurmoja et al. (2018) found an increased risk for larger farms. Boklund et al. (2020) revealed several risk factors for Romanian backyard farms, while for commercial farms, the only significant factor found was distance to nearest outbreaks. Based on the most often limited numbers of domestic pig farms affected in most MS, and especially the limited numbers of commercial pig farms affected, it can be difficult to reach a sufficient number of farms to include in studies, in order to have enough power in statistical analyses.

- *Potential impact on ASF control if the knowledge gaps were to be filled*

Calculation of the monthly incidence for different herd types and areas will help focussing surveillance in affected areas. Furthermore, increased awareness may lead to reduced incidence. Increasing knowledge on which farm types are at higher risk of ASF incursion, information campaigns can be directed to specific types of farmers and shared through specific channels reaching exactly the audience needed.

The use of results from risk factor studies will depend on which risk factors are found to significantly influence ASF incursion in different farm types. Specific risk factors, such as visitors or use of forage from affected areas, can be used directly for controlling the disease by providing guidance on biosecurity and use of different products from affected areas. Other factors, such as distance to nearest outbreak or case of ASF or weather or habitat in the surroundings of farms cannot be directly controlled, but can help support results or evidence from other studies, in terms of whether or not blood-feeding invertebrates might act as mechanical vectors in the spread of ASFV.

3.4.3. Methodologies

3.4.3.1. Monitoring of ASF incidence

Currently, the monitoring of ASF in domestic pig farms is based on reports of ASF outbreaks from each MS. By combining this information with updated population data, incidence can be calculated. The more detailed information available on populations, the more detailed incidence calculations can be performed. Especially, it is important to gain more knowledge on the seasonal incidence of herds of different types/sizes. Backyards have been described to be most often infected. However, the number of outbreaks in backyard farms should be related to the numbers of backyard farms in the area.

It is suggested to collect data from all MS, where ASF has been detected in domestic pigs, including at least the numbers of farms of chosen size strata in each country. It is recommended to

use the SIGMA data model for this purpose. From 2014 onwards, for the MS where ASF has been detected, preferably population data describing the following parameters for each individual herd should be collected for each year (2014–2021): numbers of pigs (e.g. breeding pigs, sows and boars, weaners and fatteners), the location (geo-coordinates or NUTS code level) and type of pig farm (including the level of biosecurity implemented on the farm, outdoor/indoor production). If it is not possible to collect these data, the alternative is to collect the number of pig farms for each NUTS level 3 of the following types: outdoor/indoor production, backyard (< 10 animals), small farm (10–100 fatteners), medium size farm (101–1,000 animals), large farm (> 1,000 animals), sows/no sows. This will give the opportunity to calculate incidence for each year and each herd type from 2014 onwards.

3.4.3.2. Potential risk factors for ASF incursion in domestic pig herds

For the investigation of potential risk factors for ASF incursion, a matched case–control design is suggested. Such a study design can be run either retrospectively or prospectively.

Retrospective studies have the advantage of knowing the exact numbers of ASF outbreaks that have occurred up to the end of the preset date, and thus, there is less risk of not obtaining the envisaged sample size.

Interviews of control farms are only suggested for prospective case–control studies. This is based on the risk of recall bias associated with interviews in a retrospective study. Questions about sources of feed and bedding, which might not change much over time, could be easily answered even for previous time periods. However, questions related to visitors will most likely be almost impossible to answer, unless strict biosecurity with notation of all visitors is performed. For case farms, recalling what happened in the period before the outbreak might be easier, as this will often be something the farmer has already speculated on. However, there is still a risk of unclear memories, and furthermore, farms might have changed owners or employees over time, making it more difficult to get valid answers on what happened in retrospective studies.

A. *Retrospective case–control study*

For a retrospective study with a matched case–control design, all outbreaks of ASF until the end of 2020 will be used. For each outbreak farm, two to five matched control farms are randomly selected, matched by herd size and MS. For backyard farms/small farms, two controls are suggested per outbreak farm, matched by NUTS level 3. Historic population data are needed, in order to correctly select control farms, i.e. a data set on population data for each affected MS for each year in the period 2014–2020.

Potential risk factors, which can be investigated with this approach, are herd size, distance to nearest outbreak in domestic farms, pig/farm density, distance to nearest wild boar case, number of wild boar cases within a certain distance, wild boar density estimates, wild boar habitat in the surroundings of farms, distance to or area covered by wetlands around included farms, as a proxy for abundance of blood-feeding invertebrates, and weather factors at the estimated time of virus introduction. For each outbreak farm, these factors are investigated for the time period in which ASFV might have been introduced into the farm, i.e. a 6-week period prior to detection in large farms, a 4-week period for medium-sized farms and a 2-week period for small farms or backyards. Similarly, for each control farm, the same potential risk factors are investigated for the same time period as the corresponding outbreak farm. The time period is especially important for weather factors. However, as farm size and the density of pigs/farms and wild boars have changed in Europe over time, and especially in areas affected by ASF, the availability of historic data on these parameters is especially important.

B. *Prospective case–control study*

In a prospective study, the same potential risk factors as in the retrospective study can be investigated, with an additional section on potential risk factors that can only be collected through interviews of farmers on included farms. These factors must include, but not be limited to, visitors on farms, sources of feed, storage of feed and bedding material and observations of wild boar in the surroundings. The challenges with questionnaire studies are the needed resources for visiting control farms. While outbreak farms are already visited by official veterinarians, and thorough epidemiological investigations are carried out on these farms, control farms are normally not visited during outbreaks, and therefore additional effort is needed. However, these visits of control farms could be combined, for instance, with visits on commercial farms that are carried out by the Veterinary Services to check the biosecurity and production data in the restricted zones.

The required sample size for each herd type has previously been estimated to 468 farms, based on an odds ratio of 2.5, a prevalence of exposure among control farms of 0.1, a power of 0.9 and a ratio of outbreak to control farm of 2 (Boklund et al., 2020).[2] A reduction of the power to 0.8, an increase in the expected odds ratio to 3 and an increase of the number of controls per case to 5 leads to a sample size of 56 cases and 280 controls, in total 336 included farms. In EU MS other than Romania, the total number of outbreaks in farms was 186 from 1 November 2018 to 31 October 2019 (EFSA, 2020) and 168 from 1 January to 2 October 2020. This means that overall, the study should be doable within one season. For large farms/farms with high levels of biosecurity, the required number will most likely be difficult to reach. Therefore, it is advised to increase the numbers of controls for large commercial farms. Increasing the expected odds ratio to 5, and the case control ratio to 5, leads to a sample size of 22 case farms and 110 controls, for a total of 132 large commercial farms.

The protocols suggested here are limited to short-time studies only. However, especially for risk factor studies in commercial farms, there is a need for continuing data collection over several seasons in order to be able to find significant risk factors with a relative risk below 5. If 32 case farms and 160 controls are included in the study, an expected odds ratio of 4 can be detected, while if 56 case farms and 280 controls are included, an odds ratio of 3 can be detected (Table 6). Only time will show how many cases of each herd type in the coming season will occur. Risk factor studies for large commercial farms are needed to be able to manage risk factors. For this reason, cross-border collaboration is absolutely necessary in order to reach the needed sample size and thereby a sufficient power in the statistical analyses, especially for large farms/farms with high levels of biosecurity. It is suggested to start out the project including countries with a considerable number of outbreaks in previous years for all herd types. When outbreaks occur in large farms/farms with high levels of biosecurity in other MS, the farms can be invited to be included in the study from that point in time. If only a limited number of large commercial farms are detected with ASF over the first season, it will be wise to expand the study for another season.

Control farms that experience ASF within 2 months after the relevant period as control farm should be excluded as controls and included as outbreak farms, for which new control farms would be selected.

Definitions of small/large farms or farms with high levels of biosecurity must be harmonised for all participating MS. Furthermore, questions included in the interviews must be harmonised. EFSA will provide guidance for the data model to be developed and centralising the data collection, to ensure that definitions are in accordance with the SIGMA data model.

Table 6: Comparison of sample sizes given differences in expected odds ratio, power and ratio between cases and controls

Case–control ratio	OR	n.total	n.case	n.control	n.total	n.case	n.control	n.total	n.case	n.control
		Power = 0.9			Power = 0.8			Power = 0.7		
2	**1.5**	2,922	974	1,948	2,169	723	1,446	1,698	566	1,132
	2	891	297	594	663	221	442	519	173	346
	2.5	468	156	312	348	116	232	273	91	182
	3	303	101	202	228	76	152	180	60	120
	3.5	219	73	146	165	55	110	132	44	88
	4	171	57	114	129	43	86	102	34	68
	4.5	138	46	92	105	35	70	84	28	56
	5	117	39	78	90	30	60	72	24	48
2	**5**				90	30	60			

[2] The sample size was calculated using the epi.sscc function of the epiR package v2.0.19 in R (R Core Team, 2018), which made it possible to calculate the sample size taking into account the power or minimum detectable odds ratio (OR). For a matched case–control study, the following parameters were used: OR = varied from 1.5 to 5, p0 = 0.1, n = NA, power = 0.7, 0.8 or 0.9, r = 2–10, rho.cc = 0.1, design = 1, sided.test = 2, conf.level = 0.95, method = 'matched'. OR: the odds ratio that is expected to be detected by the study; p0: the prevalence of exposure among the controls; power: the required study power; r: the number of subjects in the control group divided by the number of subjects in the case group; rho: the correlation between case and control exposures for matched pairs.

Case–control ratio	OR	n.total	n.case	n.control	n.total	n.case	n.control	n.total	n.case	n.control
		Power = 0.9			Power = 0.8			Power = 0.7		
3	**5**				104	26	78			
4	**5**				120	24	96			
5	**5**				132	22	110			
6	**5**				147	21	126			
7	**5**				168	21	147			
8	**5**				180	20	160			
9	**5**				200	20	180			
10	**5**				209	19	190			
5	**1.5**				3,300	550	2,750			
	2				996	166	830			
	2.5				522	87	435			
	3				336	56	280			
	3.5				246	41	205			
	4				192	32	160			
	4.5				156	26	130			
	5				132	22	110			

OR: Odds ratio; n.total: total sample size; n. case: number of cases; n control: number of controls.

3.4.4. Deliverables and milestones

Seasonal ASF incidence in domestic farms should be included in the External Scientific Reports, which are delivered on a yearly basis.

Description of the analyses of potential risk factors in participating countries should be reported in the External Scientific report, including a multivariable analysis of the result. The submission of a peer-reviewed article by the end of the study is encouraged (Table 7).

Table 7: Suggested milestones and deliverables for prioritised research objectives of research domain 4 (seasonality)

	Milestone	Deliverable
General (both objectives)	Participating MS identified (preferably all, otherwise those with high numbers of outbreaks/cases)	
	Definitions of herd types harmonised between participating MS	
Monitoring herd incidence		Seasonal incidence of ASF in all MS by size categories
Investigating potential risk factors	Protocol for selection of case and control farms	Protocol for selection of case and control farms
	Descriptive analysis of data (Y1)	Descriptive analysis of data
	Multivariable analysis of data (Y1)	Multivariable analysis of data
	Descriptive analysis of data	Descriptive analysis of data
	Multivariable analysis of data	Multivariable analysis of data
		Peer-review paper submitted

4. Conclusions

- From five research objectives proposed by the working group and the broader network for the Research Domain 4 (Possible factors that determine seasonality of ASF in wild boar and/or domestic pig populations-TOR 3), two research objectives were prioritised: 1) to monitor the herd incidence of ASF outbreaks in EU member states and 2) to investigate potential risk factors for ASF incursion in domestic pig herds of different herd types and/or size.

- To monitor the seasonal incidence of ASF, it is suggested to collect data from all MSs and/or from the SIGMA project
- To investigate potential risk factors for ASF incursion in domestic pig herds, retrospective case–control study and prospective case–control study are proposed.
- If only a limited number of farms with ASF are detected over the first season, expansion of the study over two seasons may be needed to achieve the necessary statistical power of the study.

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Abbreviations

ASFV	African swine fever virus
MS	Member State
RO	Research Objective
TOR	Terms of reference

Annex A – Priority scoring of research objectives that passed the inclusion criteria

Research objective	Rational	Quantitative Scores						Total average (StDev)	Variance Coefficient
		1. Impact on ASF management	2. Feasibility or practicality	3. Potential implementation in practice	4. Short timeframe	5. Novelty	6. Priority for risk managers*		
1 Comparative study of monthly ASF herd incidence risk in 2020 between EU member states									
Low	Already addressed, though not for all countries	1.0							
Low	Unclear what will be the resulting management measure			1.0					
Low	No rationale reported					1.0			
Medium	Important because pigs are key commercial hosts, but wild boar are not included.	3.0							
Medium	Monitoring herd incidence will inform you about stage of epidemic, but does not help much in management decisions on wild boar	3.0							
Medium	No rationale reported	3.0		3.0		3.0			
High	Apparently, there are not comparative studies among MS					5.0			
High	Current practice in veterinary services		5.0						
High	Should be easy to calculate		5.0						
High	Sound results can be obtained in a year				5.0				
High	This can be easily implemented in the current ASF control measures			5.0					
High	No rationale reported		5.0		5.0		5.0		
Average score of experts for criterion ((StDev)		**2.5 (0.9)**	5.0 (0)	3.0 (1.6)	**5.0 (0)**	**3 (1.6)**	**5.0 (0)**	**3.6 (1.5)**	**0.43**

Research objective	Rational	Quantitative Scores						Total average (StDev)	Variance Coefficient
		1. Impact on ASF management	2. Feasibility or practicality	3. Potential implementation in practice	4. Short timeframe	5. Novelty	6. Priority for risk managers*		
2. Harmonised case–control studies in pig herds for seasonal risk factor involving several ASF-affected countries.									
Low	Done for selected countries					1.0			
Low	Only on smaller regional or MS level					1.0			
Medium	This is a costly study, need to probably be based on case–control study in different countries		3.0						
Medium	Is already done for selected countries	3.0							
Medium	Selection of farms is feasible as well as the study of risk factors, but complexity may be limiting factor		3.0						
Medium	No rationale reported		3.0	3.0	3.0	3.0			
High	High importance. Knowledge of risk factors will help in preventing them	5.0		5.0					
High	Important to avoid introduction of ASF in farms and to estimate seasonal risk	5.0							
High	No similar studies are available among affected countries. Some are available at country level (i.e. Romania)					5.0			
High	The study can lead to sound results in one year in ASF EU affected areas				5.0				
High	Useful to set up biosecurity in different types of farms			5.0					
High	No rationale reported	5.0			5.0		5.0		
Average score of experts for criterion (StDev)		**4.5 (0.9)**	**3.0 (0)**	**4.0 (1)**	**4.0 (1)**	**2.5 (1.7)**	**5.0 (0)**	**3.6 (1.3)**	**0.36**

Research objective	Rational	Quantitative Scores						Total average (StDev)	Variance Coefficient
		1. Impact on ASF management	2. Feasibility or practicality	3. Potential implementation in practice	4. Short timeframe	5. Novelty	6. Priority for risk managers*		
3. Study of ASF seasonal pattern in association with socio-cultural activities									
Low	Difficult to define a priori what social activities are relevant for AHS persistence/dispersal		1.0						
Low	Only on smaller regional or MS level					1.0			
Low	Very difficult to show relation in only one year. Long-term studies will be needed				1.0				
Low	No rationale reported	1.0	1.0	1.0		1.0			
Medium	Costly study, need to probably be based on case–control study in different countries		3.0						
Medium	Difficult to implement a field level due to the variety of social activities and to determine its relevancy. Managing of social activities seems also difficult.			3.0					
Medium	High importance. Knowledge of risk factors will help in preventing them			3.0					
Medium	Just when related to animal movement. The other social activities may have little impact on the spread or persistence of the disease	3.0							
Medium	No rationale reported		3.0	3.0	3.0		3.0		
High	High importance. Knowledge of risk factors will help in preventing them	5.0							

Research objective	Rational	Quantitative Scores						Total average (StDev)	Variance Coefficient
		1. Impact on ASF management	2. Feasibility or practicality	3. Potential implementation in practice	4. Short timeframe	5. Novelty	6. Priority for risk managers*		
High	No similar studies are available among affected countries. Some are available at country level (i.e. Romania)					5.0			
High	No rationale reported	5.0			5.0	5.0			
Average scores of experts for criterion (StDev)		**3.5 (1.7)**	**2.0 (1)**	**2.5 (0.9)**	**3.0 (1.4)**	**3.0 (2)**	**3.0 (0)**	**2.8 (1.5)**	**0.53**

StDev: standard deviations.
Low score = 1 point; Medium score = 3 points; Large = 5 points.
*: only one expert attending the working group represented the risk managers and scored Score 6.

Annex B – Questionnaire: Request for Scientific and Technical Assistance on African Swine Fever

Why this questionnaire?

On 27 August 2019, EFSA published a scientific report titled 'Research gap analysis on African swine fever'. The Scientific Report provided a review of the most significant ASF knowledge gaps as perceived by the EU Veterinary Services and other stakeholders involved in pig production and wild boar management. The aim of this scientific report was to identify research gaps which could benefit **short-term ASF risk management** if addressed and which can facilitate evidence-informed decision-making on ASF prevention and spread. The EU is in need to further address some of the major research gaps as identified by EFSA in the Scientific Report, in particular related to the research domains: '**wild boar management**', '**ASFV transmission by arthropods**', '**ASFV survival in the environment and carcasses**' and '**risk factors contributing to ASF seasonality**'. In May 2020, EFSA was mandated by the European Commission to complement its previous Scientific Report providing new scientific input and technical assistance on those crucial topics identified by the stakeholders by identifying additional studies to fill the knowledge gaps, and to propose research protocols for the key research objectives.

EFSA has established a working group, which has started to identify possible research objectives for each of those domains in the attached file. We would kindly like to seek your expertise to verify if no research objectives are missing for any of the four research domains. If you would have additional suggestions, please could you provide a short title for the objective, a short description, a key word and possible references to similar studies LINK TO SURVEY?

The next steps will be to prioritise all research objectives based on several criteria, such as their possible impact on ASF management, the feasibility or practicality to carry out the study, the possibility for a short-time frame study (1 year), the novelty of the study and if the topic is a priority for risk managers. After prioritisation, short study protocols will be developed by experts from the working group and/or EFSA's networks, which will be published in June 2021 possibly identifying future calls for research proposals.

RESEARCH DOMAINS

Please consult the research objectives provided in the document attached. If you think some objectives are missing, kindly complete the table below.

Download

EFSA_-_List_with_possible_research_objectives.pdf

Research objectives pertaining wild boar management in view of ASF control

	Research Objective	Short description	Key word	References
1				
2				
3				
4				

Research objectives pertaining ASFV transmission by arthropods

	Research Objective	Short description	Key word	References
1				
2				
3				
4				

Research objectives pertaining ASFV survival in the environment and wild boar carcasses

	Research Objective	Short description	Key word	References
1				
2				
3				
4				

Research objectives pertaining risk factors contributing to ASF seasonality

	Research Objective	Short description	Key word	References
1				
2				
3				
4				