

Upholding of **Aujeszky's disease**-free status in 2014: improvement of detection in high-risk pig herds but decrease in field player vigilance

Clara Marcé (1)* (clara.marce@agriculture.gouv.fr), Céline Deblanc (2)**, Aurélie Oger (2)**, Olivier Bourry (2), Gaëlle Simon (2), Nicolas Rose (2), Marie-Frédérique Le Potier (2)**

(1) Directorate General for Food, Animal Health Office, Paris, France

(2) ANSES Ploufragan-Plouzané Laboratory, France

* Management team member of the French National Epidemiological Surveillance Platform for Animal Health (ESA Platform)

** OIE Reference Laboratory and National Reference Laboratory for Aujeszky's disease

Abstract

This article presents the results of surveillance of Aujeszky's disease in mainland France and Reunion Island in 2014. The results show an increase in the number of open air pigs tested, especially among farrow-to-finish farms. However, the proportion of pig farms screened seems to have decreased, especially among grow-to-finish farms. On the other hand, the number of pigs screened in nucleus and breeder-multiplier farms was similar in 2014 to previous years. A decrease in the number of suspicions, serological or clinical, has also been noticed. Despite the fact that no cases of Aujeszky's disease were detected in 2014, the priority for all stakeholders is to remain vigilant. It is especially important that veterinarians include Aujeszky's disease in their differential diagnosis when encountering symptoms (*influenza*-like illness, spontaneous abortions) that cannot be attributed with certainty to another disease.

Keywords

Regulated disease, Category 1 health hazard, Aujeszky's disease, Epidemiological surveillance, France, Official control, Swine

Résumé

Maintien du statut indemne de maladie d'Aujeszky en 2014 : amélioration du dépistage dans les élevages à risque mais baisse de la vigilance des acteurs de la filière
Cet article présente les résultats de la surveillance de la maladie d'Aujeszky en France continentale et dans l'île de la Réunion en 2014. Ces résultats rapportent une augmentation du nombre de porcs d'élevages plein-air dépistés, notamment en élevages naisseurs-engraisseurs. La proportion d'élevages dépistés apparaît en baisse, notamment chez les engraisseurs. Le nombre de porcs dépistés en élevages de sélection-multiplication reste stable. Une diminution du nombre de suspicions a été observée, qu'elles soient sérologiques ou cliniques. Bien qu'aucun cas de maladie d'Aujeszky n'ait été confirmé en 2014, le maintien de la vigilance de l'ensemble des acteurs reste la priorité. Il est notamment important que les vétérinaires incluent la maladie d'Aujeszky dans leur diagnostic différentiel lors de signes cliniques (*syndrome grippal*, *avortements*) ne pouvant être rattachés avec certitude à une autre maladie.

Mots clés

Maladie réglementée, danger sanitaire de 1^{ère} catégorie, maladie d'Aujeszky, épidémiologie, France, police sanitaire, suidés

Here, we present the results of the Aujeszky's disease (AD) surveillance programme (Box) in mainland France and Reunion Island for 2014.

Population counts used in this report come from holding registration forms filed by pig farmers on or before 31 December 2014 (compiled in the BDPORC, the national pig identification database and transmitted to the DGAL's information system, SIGAL). All pig keepers are required to make this declaration (Ministerial Order of 20 October 2010 amending the Ministerial Order of 24 November 2005). All new pig holdings must be registered and records must be updated if there are any changes to the initial information provided. Since the Aujeszky's disease surveillance programme is not implemented in Corsica (which does not have disease-free status), the pig numbers in this article do not include its two *départements*.

Sampling

Surveillance in nucleus and multiplier herds

Surveillance was conducted in 384 of the 505 nucleus and multiplier holdings identified in the pig holding registry (i.e. a 76% coverage rate).

On average, 45 samples were taken per holding and per year, i.e. 14 samples per quarter, for a total of 20,967 samples. This was a slight increase on the average of 12 samples per holding and per quarter for 2013 (to a level equivalent to 2012) (Marcé *et al.*, 2013; Marcé *et al.*, 2014). As a guideline, 12 samples per holding and per quarter are sufficient to detect a minimum intra-holding prevalence in the region of 25%, with a confidence level of 95%.

Overall, assuming that samples were only taken on breeding stock, and according to the pig population count recorded in the BDPORC database, 25% of breeding stock were tested in 2014, or 6% per quarter, i.e. the same levels as in previous years.

Surveillance in outdoor pig production holdings (farrow-to-grower, farrow-to-finish, wean-to-grow and grow-to-finish farms)

In all, 1,691 outdoor holdings (domestic swine or farmed wild boar) were listed as having been tested out of 2,659 holdings in the database (2,438 outdoor domestic pig holdings and 221 holdings with wild boars), i.e. a 64% coverage rate, with 15,669 samples taken.

The rate of implementation of programmed surveillance varied with the type of domestic swine holding, from 66% in farrow-to-finish holdings to more than 100% in wean-to-grow holdings (Table 1).

Of a total of 2,215 outdoor domestic pig holdings whose type (activity, production level) is known in SIGAL, the surveillance programme effectively covered 1,691 (76% surveillance coverage rate), for a total of 15,352 samples.

As a guideline, a mean of nine samples taken per holding and per quarter are sufficient to detect a minimum intra-holding prevalence in the region of 30%, with a confidence level of 95%.

Surveillance in indoor pig production holdings

Despite the lack of mandatory programmed screening, 138 farms underwent screening for Aujeszky's disease (5,359 samples).

Objectives of the surveillance programme

For mainland France and Reunion Island:

- To confirm France's status as Aujeszky's disease-free (AD-free).
- To detect as early as possible any new appearance of the virus in domestic swine.

The population monitored

Domestic swine and farmed wild boars (categories A and B) throughout mainland France and Reunion Island.

Surveillance procedures

Outbreak surveillance

Two levels of suspicion have been defined based on clinical criteria developed in association with the SNGTV: "high" clinical suspicion corresponding to a diagnosis of inclusion and "low" clinical suspicion corresponding to a diagnosis of exclusion (definitions can be found in Memorandum DGAL/SDSPA/N2013-8011 of 15 January 2013). Regardless of the suspicion, the DDecPP must be notified and sampling must be carried out for serological and virological diagnosis.

Programmed surveillance (DGAL/SDSPA/N2013-8010)

Less intense, but targeted serological surveillance in the most at-risk holdings (risk of introduction in outdoor holdings or risk of spread in nucleus and multiplier herds).

For all outdoor holdings, including grow-to-finish holdings: annual serological testing (15 samples from breeding stock and/or 20 samples from slaughter pigs).

In nucleus and multiplier herds: quarterly serological surveillance (15 samples).

Holdings for which AD-free status was revoked or suspended for administrative reasons (in particular due to absence of programmed screening for more than one year) must request and undergo a requalification procedure. Obtaining AD-free status requires two series of negative serological tests performed at a two-month interval, on at least 15 breeding pigs and 30 slaughter pigs.

Health control measures (DGAL/SDSPA/N2013-8011)

In the case of clinical suspicion, regulations stipulate that samples should be taken for serological and virological (PCR) tests. No APMS is issued in the case of low clinical suspicion. An APMS is issued only in the case of high clinical suspicion, low clinical suspicion associated with positive first-line test results (serology or virology), or low clinical suspicion associated with unfavourable epidemiological investigation results.

Serological suspicion is based on non-negative serological results. An animal considered seropositive for AD is one for which two series of tests have been performed at least 15 days apart and show positive results, with each test including two serological analyses using two different assays (gB and gE), because these two methods can rule out the possibility of non-specific reactions.

In the case of positive serological tests, the farm is visited for clinical examination of the animals and to take more samples for additional serological tests (at least 15 days apart). The holding is placed under APMS if an accredited laboratory produces a positive or ambiguous result in any individual test. If only one or two samples are positive or ambiguous, the health control measures can be "relaxed": movements of pigs to slaughter or to authorised terminal holdings are authorised, providing that the clinical and epidemiological investigation of the holding under serological suspicion has given favourable results, that the destination holding or slaughterhouse has agreed in writing that these pigs can be introduced, and that the destination holding is also placed under APMS.

An animal is considered infected by AD when, even in the absence of any suggestive clinical signs of the disease, the results of serological or virological tests confirm the infection.

A site is considered infected when a pig infected with AD is held there or originates from there.

When an outbreak is confirmed, the prefecture declares the pig farm as infected (APDI), which entails depopulation as quickly as possible and cleaning-disinfection operations. Trace-back and trace-forward epidemiological surveys are implemented to determine the source and the conditions under which the infection spread to the holding, and to identify other holdings that are likely to have been infected.

Regulatory References

Council Directive 90/429/EEC of 26 June 1990 (amended) laying down the animal health requirements applicable to intra-Community trade in and imports of semen of domestic animals of the porcine species

Commission Decision 2008/185/EC of 21 February 2008 (amended) on additional guarantees in intra-Community trade of pigs relating to Aujeszky's disease and criteria to provide information on this disease

Ministerial Order of 28 January 2009 laying down the technical and administrative measures in regard to collective prophylactic measures and animal health rules for Aujeszky's disease in départements with Aujeszky's disease-free status

Ministerial Order of 14 August 2001 on the animal health rules required for intra-Community trade of cattle and swine

Ministerial Order of 7 November 2000 laying down the animal health rules required for disseminating swine semen

Ministerial Order of 9 June 1994 on the rules that apply to trade of live animals, semen and embryos and to the organisation of veterinary inspections

Table 1. Testing for Aujeszky's disease in outdoor holdings in 2014 (domestic swine only, holdings having filed a declaration)

Type of outdoor holding	Number of holdings registered*	Number of holdings tested	Proportion of holdings tested (%)	Number of samples	Average number of samples per holding
Farrow-to-grower	198	152	77	1,536	10
Wean-to-grow	7	15	214**	203	14
Grow-to-finish	1,205	794	66	7,609	10
Farrow-to-finish	805	695	86	6,004	9
Total	2,215	1,691	76	15,352	9

* Taken from the BDPORC database in the first quarter of 2015 for mainland France. All départements were included, although five départements did not provide all information on Aujeszky's disease surveillance and départements were not requested to validate their pig population counts, which were taken directly from the SIGAL database. The farrow-to-grower category includes farrowing and wean-to-grow farms; wean-to-finish holdings were included in the grow-to-finish category.

** A failure to update certain holding declarations in BDPORC combined with the lack of corrections by the DDecPPs of pig counts extracted from SIGAL explains why the proportion of wean-to-grow farms tested was greater than 100%.

In all, including all the holdings mentioned previously, 41,995 samples were taken for serological screening of Aujeszky's disease.

Non-negative results

In outdoor production systems, 15 pig holdings presented at least one non-negative result for the first-line ELISA gB test (127 samples). Following these results, 10 sites were placed under APMS. Four sites had to be visited a second time to collect enough serum for confirmatory diagnostics (gE in particular).

In total, regardless of the type of pig holding, 38 sera of pigs or wild boars (relating to 11 suspected cases) underwent second-line testing by the NRL, 15 of them in ELISA gB (a single site) and 23 in ELISA gE.

None of the suspicions in pig holdings were confirmed. Animals from the two wild boar holdings that had been the subject of strong suspicion, following the annual screening, were slaughtered pre-emptively (two wild boars in a holding in the Cher *département* and 61 wild boars in a holding in the Dordogne *département*).

Clinical suspicions

For the entire disease-free territory (mainland France and Reunion Island), one outdoor holding was the subject of a clinical suspicion (Loire-Atlantique *département*): four pigs were tested and returned negative results. Two wild boars (wildlife) were also the subject of a clinical suspicion and underwent testing in the Côtes d'Armor *département*. All these suspicions were overturned.

In the context of clinical suspicions, the NRL received thirteen samples in 2014, from four dogs (from the *départements* of Aisne, Ardennes, Marne and Essonne, all positive), one cow (Haute-Saône, negative), six pigs (two from Corsica and four from Loire-Atlantique, all negative) and two wild boars (Côtes d'Armor, both negative).

The number of clinical suspicions reported by the DDecPPs may be underestimated due to a request for first-line testing made to a laboratory from the network of accredited laboratories in the context of a very low suspicion (exclusion diagnosis).

Costs

In 2014, in the 95 *départements* for which there was usable data, the French government invested around €25,000 for surveillance and control of Aujeszky's disease. Laboratory costs amounted to €11,050 for programmed screening and €380 for health control measures. Veterinary costs were €12,220 for programmed surveillance and €1,210 for enforcing health control measures. In addition, State participation in programmed surveillance in nucleus and multiplier holdings belonging to the French pig breeding agency amounted to approximately €30,400 for sampling and serological analyses (data not consolidated at the date this article was submitted, probably underestimated).

Discussion

No outbreaks of Aujeszky's disease were identified in 2014 in domestic pig holdings in mainland France or Reunion Island. One pig holding showed suspicious clinical signs in mainland France, and one in Corsica. Two wild boar holdings were the subject of serological suspicions. PCR analyses carried out by the network of accredited departmental testing laboratories are not systematically recorded in the central database. Therefore, some differential diagnoses may not have been recorded and the number of tests may be underestimated. Because such data are important for estimating the level of surveillance, this situation needs to change to ensure that the analyses carried out by the network of accredited laboratories can be compiled. The debate around the management of programmed surveillance in pig farming and the computerisation of the results of analyses carried out in laboratories for the swine sector should help, from 2016, better assess

the frequency of these differential diagnoses. And even from 2015, it should be possible to communicate the first results of the programmed surveillance campaign in pig farming, *via* computerised exchanges, to the DDecPPs. However, this work still needs to be consolidated in order to integrate the results of the differential diagnoses in these exchanges of computerised data and to connect the NRL to this scheme.

In contrast, the detection *via* programmed surveillance of two seropositive wild boar holdings, which did not however reveal any active viral circulation, recalls the episode that occurred in 2010 (Rose *et al.*, 2010). These results are a reminder that there is a real risk of recurrence of the disease in domestic pigs, in outdoor holdings in particular. Outdoor holdings are particularly exposed to the disease due to the possible contact with wildlife (Rossi *et al.*, 2008), to the fact that they are less closely monitored compared to indoor holdings and also that clinical signs of infection can be attenuated, especially respiratory symptoms due to generally lower viral shedding at lower pig densities. Combining outbreak and serological surveillance in outdoor holdings, whether of domestic pigs or farmed wild boars (for which outbreak surveillance is limited), is therefore essential (Pol and Le Potier, 2011).

It is difficult to compare the results of the serological surveillance conducted in 2014 with those of 2013 (Marcé *et al.*, 2014), even though the same method (based on the declaration of pig holdings) was used to identify the number of pig holdings in France. While the number of herds tested and the number of samples taken are both on the increase, the proportion of tested herds appears to be in decline. The number of farms registered, whether outdoor holdings alone or all pig farms, increased by 4 to 5% in 2014 compared to 2013. As in 2013, the DDecPPs were not given the opportunity to rectify the results of the extraction regarding the number of holdings (data from the holding declarations and data entered in the BDPORC database) and thus to correct the data for holdings that had not yet filed their declaration. The figures used in 2014 relate to the raw extraction from BDPORC data. Farms that had not yet updated their declarations of activity could not therefore be reclassified *a posteriori* by the DDecPPs. It is also possible that data was updated before the extraction, following the implementation of the dataflow between BDPORC and SIGAL to facilitate consistency checking and enable the DDecPPs to remind farmers who have not yet fulfilled their obligations to declare their activity or notify a change. It should also be noted that the number of registered farmed wild boar holdings was stable between 2013 and 2014. The holding declaration is not yet in effect for farmed wild boar holdings or outdoor holdings, even though these are subject to programmed surveillance for Aujeszky's disease. The standard form for the holding declaration was adapted in 2014, whereas the dataflow between BDPORC and SIGAL was only implemented in 2015. The numbers of farmed wild boars are thus very probably underestimated in the SIGAL database, which was the source of the numbers used for this report. It was not possible for the DDecPPs to correct these population data, whether for domestic pigs or wild boars.

The response rate of the *départements* for the questions on Aujeszky's disease was similar in 2014 to that for 2013 (95 *départements* in 2014, compared with 97 in 2013 and 88 in 2012).

The combination of the lack of any correction to the numbers of pigs on outdoor holdings, which rose between 2013 and 2014, and the stable rate of completion by DDecPPs of the questionnaire on Aujeszky's disease, could explain the observed increase in 2014 in the number of farms tested, and the decrease in the proportion of farms tested. Further encouragement is needed to achieve an increase in the proportion of farms tested. The failure to correct the numbers of pigs also explains why the proportion of wean-to-grow holdings screened is more than 100%.

For all of the outdoor farms registered (domestic pigs), the rate of implementation of programmed surveillance was 76%, lower than 2013, despite the increase in the number of samples and holdings included. Nevertheless, the limitations mentioned in the previous paragraph should be borne in mind. Annual serological surveillance

in outdoor holdings, particularly in farrow-to-grower holdings, should help compensate for the limitations of outbreak surveillance. It is now necessary to ensure that screening is effectively and fully carried out, considering that the nine serological tests performed on average only ensure the detection of a seroprevalence level of 30%, which is too high considering the seroprevalence levels that can occur in outdoor holdings (routine tests involving 15 samples can target a prevalence of 20%, with a 5% error rate).

Stagnation in the number of nucleus and multiplier herds analysed was also observed, which may be related to the stagnation in population counts in this type of holding. For this type of holding, the number of samples per holding and per year rose slightly compared to 2013, as did the average number of samples per holding and per quarter. It appears important to maintain this pressure of analysis at the nucleus-multiplier level in order to maintain the sensitivity of the detection system. If the number of samples per holding decreases, detection will only be effective when intra-herd prevalence is higher than 30%, which is too high a threshold compared to those that may be encountered.

It should also be noted that indoor holdings in some *départements* are still subjected to testing even though this type of holding is not targeted by mandatory screening (because these holdings are considered to be at a lower risk of introduction or spread of the virus). These analyses may nevertheless be appropriate for wean-to-grow holdings, which are the farms that disseminate the animals, even if it is on a lesser scale compared to nucleus and multiplier holdings.

Of the 15 outdoor holdings that showed positive first-line serological results, four required a second round of samples on very short order to obtain sufficient serum to carry out the confirmatory tests. This highlights the importance of taking blood samples (not blots) for serological testing on farms, particularly when an outbreak is suspected, to rapidly confirm or refute the presence of an outbreak of Aujeszky's disease. Repeat tests nevertheless remain infrequent and blots can still be useful, especially when containment is difficult. It would seem appropriate to implement practical training on taking blood samples and on containment of animals for mandated veterinarians who do not work frequently in the swine sector.

In conclusion, all players in the pig sector must remain vigilant in order to ensure early detection of any outbreak. On this subject, two

clinical suspicions, including one in wildlife (a decrease compared to the previous year), were notified in 2014 in mainland France and on Reunion Island, both recognised as Aujeszky's disease-free territories. The two suspicions were dealt with by the NRL. To increase vigilance, the approach involving exclusion tests for suspected cases should be pursued, and all veterinarians should be encouraged to include Aujeszky's disease in their differential diagnoses when flu-like symptoms and abortions cannot be attributed with certainty to any other disease. The exclusion test makes it easier to report suspicions while reducing the consequences for the holding. The current lack of a reliable system for recording these exclusion diagnoses (record of laboratory analyses performed) means that clinical surveillance activities are not clearly or fully described. It is therefore necessary to improve the tools used for monitoring epidemiological information. It is also important to stress that outdoor holdings are the farms most at risk. It is fundamental therefore that programmed surveillance be carried out on all outdoor holdings, and on at least fifteen pigs per holding, as recommended, to detect any infection as close to the source as possible.

References

- Marcé, C., Deblanc, C., Simon, G., Rose, N., Le Potier, M.F., 2013. Bilan de la surveillance de la maladie d'Aujeszky en France en 2012 : maintien du statut indemne de maladie d'Aujeszky en France continentale, *Bul. Epid. Santé Anim. Alim.* 59, 47-50.
- Marcé, C., Deblanc, C., Oger, A., Bourry, O., Simon, G., Rose, N., Le Potier, M.F., 2014. Bilan de la surveillance de la maladie d'Aujeszky en France en 2013 : maintien du statut indemne de maladie d'Aujeszky en France continentale, *Bul. Epid. Santé Anim. Alim.* 64, 45-48.
- Pol, F. et Le Potier, M.F., 2011. Herpès-virose chez le porc : la maladie d'Aujeszky. *Bull. Acad. Vet.* 164,(4) 35-39.
- Rose, N., Bronner, A., Pol, F., Le Potier, M.F., 2010. Point sur la situation épidémiologique de la maladie d'Aujeszky en Aquitaine en 2010 : premières investigations suite à la découverte d'un foyer, *Bull. Epid. Santé Anim. Alim.* 41, 16-17.
- Rossi, S., Hars, J., Garin-Bastuji, B., Le Potier, M.F., Boireau, P., Aubry, P., Hattenberger, A.M., Louguet, Y., Toma, B., Boué, F., 2008. Résultats de l'enquête nationale sérologique menée chez le sanglier sauvage (2000-2004). *Bull. Epid Santé Anim. Alim.* 29, 5-7.