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Focus on regulated and emerging diseases (REDs) – 2012 update

EDITORIAL

This special issue of the *Bulletin Épidémiologique – Animal Health & Nutrition* is the fourth edition of the annual report on surveillance of the main regulated and emerging animal diseases. This publication, made available to all stakeholders in the various networks, contributes to the annual health update prepared as part of the National Epidemiological Surveillance Platform for Animal Health (ESA Platform). This platform brings together the Directorate General for Food (DGAL), ANSES, livestock farming organisations (GDS France and Coop de France), veterinarians (SNGTV), field laboratories (Adilva), as well as hunting and wildlife organisations (FNC, ONCFS), with the aim of constantly improving epidemiological surveillance systems in animal health.

The format of this edition has not changed since last year, with a presentation and analysis of the health setting, a reminder of the surveillance procedures, animal health rules, and the main regulatory texts underlying disease surveillance. At the end of the report, a table summarises the main surveillance data, disease by disease, providing an overall picture of surveillance efforts and the status of animal health. The table also shows data on suspected or confirmed cases of rare or exotic diseases that are not covered by a specific surveillance system. In the future, the relevance of the observations reported in the different articles could be increased through improved quality and completeness of collected data, an issue that is still too often a limiting factor.

Like previous editions, this issue has broadened its scope and now includes an article on Schmallenberg virus, an emerging health risk in 2012. This article reports back on implementation of surveillance set up by the State and then carried forward by professional organisations (GDS France), when this hazard was requalified in national and international monitoring plans. This change in the surveillance system over time, which is indeed unique, was made possible by the existence of the ESA Platform, a body that facilitated the switch from State-run surveillance to surveillance by professionals within the sector. Faced with the emerging risk, the ESA Platform demonstrated its ability to respond quickly to such a situation, and the added value it brings in terms of coordinating members of the Platform, and analysis of surveillance data.

The 2012 edition, which relies on data from a wide range of surveillance systems, shows that the overall animal health status in France is very good. It is nonetheless essential to point out that vigilance must be maintained and encouraged, especially when wildlife is infected. Examples include the slow and difficult eradication of tuberculosis in some at-risk areas, and the presence of brucellosis in wild animals, identified in 2012.

Like all the editions of the *BE*, you will find this issue on the Ministry of Agriculture (www.agriculture.fr) and ANSES (www.anses.fr) websites. It is also referenced on the ESA Platform website (www.plateforme-esa.fr). Moreover, the previous version (BE 54) has now been translated into English to provide international visibility to surveillance methods and results in France. This edition is available solely in electronic format. We hope that the epidemiological surveillance data presented will thus be accessible to the widest possible audience.

Lastly, the Editorial Board and authors would like to thank all those who helped provide the statistics for this publication through their day-to-day commitment: Departmental Directorates for Protection of the Population (DDecPP), veterinarians, livestock farmers, field laboratories, and National Reference Laboratories.

The Editorial Board - BE Regulated and Emerging Diseases

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The *Bulletin Épidémiologique* is a joint publication of the French Agency for Food, Environmental and Occupational Health & Safety, and the Directorate General for Food of the Ministry of Agriculture, Food and Forestry.

Definitions

Outbreak surveillance
Outbreak surveillance, previously known as passive surveillance, refers to any monitoring activity that relies on spontaneous notification of cases or suspected cases of a monitored disease by source data contributors. When this type of system is used, it is therefore not possible to predict the quantity, nature and geographic location of data that will be collected by the network. Outbreak systems are particularly suitable in situations where early warning is needed should a disease emerge or re-emerge. This applies to epidemiological surveillance of exotic diseases, which covers the entire population. In this case, all sources of data for notification of suspected cases need to be mobilised to ensure early and rapid transfer of information.

Programmed surveillance
Planned surveillance, also called active surveillance, involves collection of data through pre-scheduled actions following a methodology that enables extrapolation of the findings to the monitored population. Unlike outbreak surveillance, it is possible to determine in advance the quantity, nature and geographical location of the data that are to be collected by the network. Routine surveillance can be carried out in an exhaustive manner, covering the entire target population, or can focus on a sample of the population. When a specific sample is monitored, it can be considered representative of a group, for example through random selection. The sample surveillance system involves occasional collection of data through surveys or repeated collection using a sentinel population. A risk population may also be chosen as the sample group.

Acronyms

Acersa: French Certification Association for Animal Health
AGID: Agar gel immunodiffusion
ANSES: French Agency for Food, Environmental and Occupational Health and Safety
APDI: Prefectural declaration of infection
APMS: Prefectural monitoring order
BAT: Buffered antigen test
BDNI: National identification database
BNEVP: National division for veterinary and plant health investigations
BPAT: Buffered Plate Agglutination Test
CF: Complement fixation
CIDT: Comparative intradermal tuberculin test
DAAF: French Overseas Directorate for Food, Agriculture and Forestry
DTL: Departmental testing laboratory
DDAAF: Departmental Directorate for Food, Agriculture and Forestry
DDecPP: Departmental Directorate for Protection of the Population
DGAL: Directorate General for Food
DRAAF: Regional Directorate for Food, Agriculture and Forestry
GDS: Animal health protection Farmers' organization
IFN-gamma: Interferon gamma
MAAF: Ministry of Agriculture, Food and Forestry
ND: Notifiable disease
NDCCM: Notifiable disease with compulsory control measures
NRL: National reference laboratory
OIE: World Organisation for Animal Health
ONCFS: National Office for Hunting and Wildlife
SAGIR: French wildlife disease surveillance network
SIDT: Single intradermal tuberculin test
SIRE: Equine information database
SNGTV: French national society for technical veterinary groups
SRAL: Regional Food Authority

Access to legal documentation concerning regulated diseases

- All regulatory texts can be consulted on the Légifrance website (http://www.legifrance.gouv.fr/) consolidated versions with restricted access on the Galatée website (http://galateepro.agriculture.gouv.fr/)
- Memoranda cited as references can be consulted on the website of the French Prime Minister (http://circulaire.legifrance.gouv.fr/index.php?action=accueil) or with restricted access on the Galatée website (http://galatee.national.agri/) or Nocia website (http://nocia.agriculture.gouv.fr/))

Bovine tuberculosis in France in 2012: positive signs but a situation that is still complex in some areas

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Abstract
The situation of bovine tuberculosis in France in 2012 is still contrasting. At the country level, the situation is very good, the herd prevalence is lower than 0.1% and the incidence does not increase. However this situation is not entirely satisfactory since the eradication of the disease in some areas is difficult and slow, specially when wildlife is also infected. Sensitisation measures for the different stakeholders lead to a global increase of the sensitivity of the system which is necessary for early detection of outbreaks but in the meantime a decrease in the confirmation rate can be discouraging for field workers.

Keywords
Regulated disease, bovine tuberculosis, surveillance, cattle

Résumé
Tuberculose bovine en France en 2012 : des signaux favorables mais une situation toujours complexe dans certaines zones
La situation de la tuberculose bovine en 2012 en France continue d'être contrastée. À l'échelle du pays, la situation est globalement très favorable, la prévalence troupeau est inférieure à 0,1 % et l'incidence n'augmente pas. Toutefois cette situation n'est pas totalement satisfaisante, car l'éradication de la maladie dans certaines zones à risque est complexe et lente, d'autant plus lorsque la faune sauvage est également infectée. Les actions de sensibilisation des différents acteurs conduisent à augmenter la sensibilité globale du dispositif nécessaire pour détecter les foyers le plus précocement possible, mais cela s'accompagne d'une baisse du taux de confirmation parfois démotivante pour les opérateurs de terrain.

Mots clés
Maladie réglementée, tuberculose bovine, surveillance, bovins

Important note:

Data collection for 2012 was particularly difficult and a large number of départements provided incomplete responses. When missing data led to a major bias in results, collected data were not taken into account. When this was not the case, variables calculated despite incomplete data were identified with an asterisk (*).

Surveillance of tuberculosis

An overview of surveillance requirements and control measures for bovine tuberculosis is presented in Box 1.

In most *départements*, programmed screening programmes for tuberculosis on livestock farms are scheduled for the wintering period between October and April, and not over the calendar year. Therefore, the results for the 2012 calendar year correspond to the end of the 2011-2012 programme and to the beginning of the 2012/2013 programme, possibly with slight differences in implementation methods.

Wildlife surveillance is also implemented depending on the risk classification of the specific *département* under the Sylvatub scheme (Box 2).

Prophylactic screening intervals for 2012 as reported by Departmental Directorates for Protection of the Population (DDecPP) are shown in Figure 1. Most *départements* discontinued systematic tuberculin testing several years ago (Table 1). An increasing number of *départements* have chosen to determine a tuberculin testing schedule for a specific zone ("zoning"), that is different from the rest of the *département*. Zoning is established by the Departmental Prefect and must be submitted for an opinion to the DGAL. This also applies to changes in intervals for each *département*.

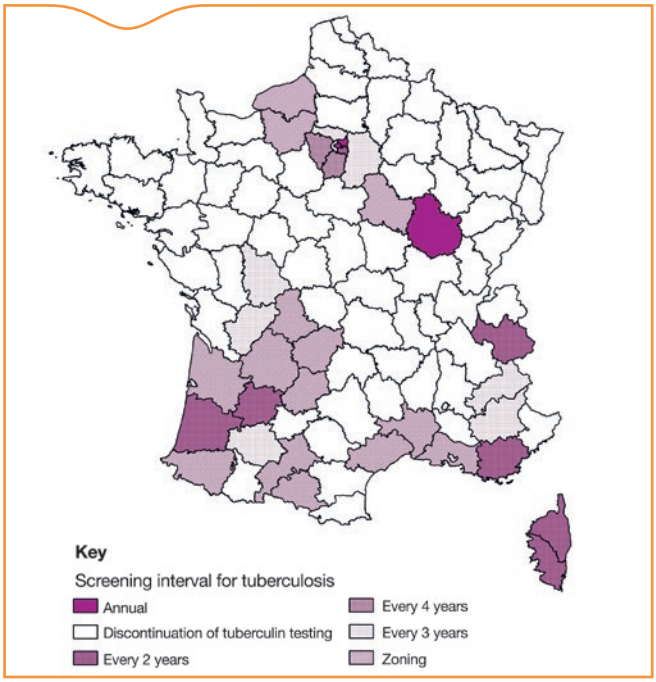

Figure 1. Programmed screening intervals by *département* in 2012

Box 1. Surveillance and control measures for bovine tuberculosis

Objectives

General objective: The motivation underlying control of bovine tuberculosis is the zoonotic nature of the disease. Given the low prevalence of the disease, public health considerations are currently far less important than the economic issues surrounding the officially TB-free status of France.

Specific objectives

- Avoid any increase in the number of outbreaks,
- Keep prevalence below the level required to maintain the officially-free status of France in terms of European Commission requirements,
- Move towards eradication of bovine tuberculosis.

Furthermore, on an individual basis, TB surveillance is integrated in regulatory processes for officially disease-free classification of herds, which is a requirement for free circulation of animals, animal sperm or embryos, and for sale of raw milk within the EU.

Scope of surveillance

Bovine tuberculosis due to *Mycobacterium bovis* or *Mycobacterium tuberculosis*. *Mycobacterium caprae* was recently reclassified and is no longer part of the *Mycobacterium bovis* group, but effectively remains within the scope of the surveillance programme.

Monitored population

All cattle farms across France.

Other susceptible populations undergo routine surveillance through *post-mortem* inspection at the slaughterhouse, particularly goats, sheep, and swine, as well as farmed deer.

Monitoring of wildlife such as deer, wild boars and badgers, follows specific protocols.

Definition of cases

Regulatory definitions were established in Article 12 of Ministerial Order dated 15/09/2003, as amended:

Suspected infection

- Lesions indicative of tuberculosis at the slaughterhouse or on necropsy, or on the basis of a positive histology finding, or a positive PCR result without identification of the bacillus,
- Non-negative tuberculin reactions and/or non-negative results for the interferon gamma assay (IFN-gamma) during a prophylactic procedure or other control, irrespective of the justification for the control.

Confirmed infection

- Identification of *Mycobacterium bovis* or *Mycobacterium tuberculosis*,
- Observation in the same animal of a positive PCR analysis associated with histological lesions indicative of tuberculosis identified by an accredited laboratory, or in an animal from a suspect herd for a reason other than a positive result,
- Histologically suggestive lesions for tuberculosis in an animal that had a positive intradermal tuberculin test.

Regulations provide for other definitions of infected animals, but they are not used in routine practice.

Surveillance methods

Screening

Surveillance of bovine tuberculosis is active and involves several complementary systems.

- Systematic surveillance at the slaughterhouse: inspection of all slaughtered animals for human consumption. Only *post-mortem* inspection is truly relevant for tuberculosis. It involves examination of a certain number of organs including the primary tuberculosis sites such as the lungs and retropharyngeal, tracheobronchial and mediastinal lymph nodes. If suspect lesions are detected, the organs are removed along with associated lymph nodes and examined in an accredited laboratory by PCR/bacteriological testing for mycobacteria.
- Programmed surveillance: testing required to obtain and maintain the officially disease-free status. The general rule is annual screening of all cattle over six weeks through single intradermal tuberculin testing (SIT). Depending on changes in prevalence within a *département*, the screening interval may be extended and the age of screened animals increased, ending with discontinuation of prophylaxis. Rules enabling less stringent surveillance are defined by the Ministerial Order of 15/09/2003, Section III, Article 13. Intradermal tuberculin tests are read 72 hours post-injection. Subjective reading of results with skin palpation (subjective method), a common practice until recently, is accepted under certain conditions. When certain criteria are met, screening can be performed using comparative intradermal tuberculin testing (SICTTSICTT), specifically when there is a strong likelihood of false positive results. In this case, the test is always read as per the objective method with use of a calliper. Under specific conditions, particularly in Camargue, SIT screening can be reinforced using systematic IFN-gamma testing.
- The sensitivity (Se) and specificity (Sp) of these tests are not perfect:
  - > *SIT: Se ~ [80%-91%] and Sp ~ [75% – 99.9%] - depending on zones*
  - > *SICTT: Se ~ [55% - 93%] and Sp ~ [89% -100%]- depending on zones*
  - > *Bovigam IFN-gamma: Se ~ [81% -100%] and Sp ~ [88% -99%]*
  - > *Recombinant IFN-gamma: Se ~ [84% -98%] and Sp ~ [92% -96%]*
- Irrespective of the interval in effect in a *département*, programmed screening can be requested annually for a period of three to five years on production sites that are classified at-risk due to epidemiological links to an infected farm.
- Alongside programmed surveillance, screening can also be implemented when animals are moved. Given that the health system is considered robust and that France is officially TB-free, screening of animals on introduction may be waived, except in certain cases:
  - > *if it takes more than 6 days for the animals to transit between two establishments,*
  - > *if the animals leave a farm classified as at-risk due to proximity to a domestic or wildlife outbreak or because of previous infection,*
  - > *if the animals transit through a farm with a high turnaround and come from a farm located in a département where the cumulative 5-year prevalence of bovine tuberculosis is higher than the national average.*

Animal health rules

Control measures aim to confirm the status of suspect animals and, if necessary, to eliminate infection from the herd. In 2012, testing protocols for suspected cases were harmonised nationally, taking into account the different initial tests (SIT or SICTT). The following principles are universally applicable:

- If non-negative results are found for a farm, a risk analysis is carried out by the DDecPP to assess whether the suspicion is low or high on the basis of epidemiological criteria, and if necessary additional investigations are carried out to test all or part of the herd, as part of control measures, using either SICTT or, when available, IFN-gamma with specific peptides in an experimental setting. In the event of low suspicion, animals are retested six weeks later or are directly slaughtered diagnostically. In this case, damaged organs are sampled and, whether or not lesions are found, retropharyngeal, mediastinal, and tracheobronchial lymph nodes are sampled and tested for mycobacteria by PCR and cell culture. If suspicion is high from the outset, or because reactions to tests performed six weeks after low suspicion confirm the suspected cases, reactors are slaughtered diagnostically and other cattle in the herd are retested after this diagnostic slaughter of confirmed animals.
- If an infection is confirmed, farms to which the disease may have spread or farms that may have been the source of the infection are identified and investigated (farms likely to be infected because of an epidemiological link). Testing is carried out using SIT, SICTT or diagnostic slaughter, and the farms may then be classified at-risk.
- If an infection is confirmed, the infected farm is cleansed. This generally involves complete depopulation of the herd with increased inspection at the slaughterhouse, followed by cleaning-disinfection. In certain specific cases, justified by preservation of local breeds or experimentally in Dordogne and Côte-d'Or, control measures may involve partial depopulation. In this scenario, animals are tested using SICTT or IFN-gamma on several occasions. Reactors are slaughtered for diagnostic purposes. The herd is considered to be cleansed after two favourable tests have been performed at a two-month interval, and is considered reclassified after two further favourable controls at two-month intervals.

Box 1. Surveillance and control measures for bovine tuberculosis

Regulations

All regulations can be consulted on the Légifrance website or with restricted access in the consolidated versions on the Galatée website.

- Council Directive 64/432/EEC of 26 June 1964 on animal health problems affecting intra-Community trade in bovine animals and swine
- French Rural Code, Book 2, Preliminary Title and Title II
- Ministerial Order of 15 September 2003 establishing the technical and administrative framework for collective prophylaxis and control measures for bovine and caprine tuberculosis
- Guidance notes cited in reference can be consulted on the website of the Prime Minister or with restricted access *via* the Galatée and Nocia websites.

Table 1. Programmed surveillance of bovine tuberculosis by tuberculin testing on classified livestock farms in 2012

Cattle herds as of 31/12/2012	224,514
ODF herds as of 31/12/2012 (%)	224,055 (99.87)
Control interval (number of *départements*)	Annual: 10 Discontinued: 56 Every 2 years: 7 Every 4 years: 2 Every 3 years: 8 Zoning: 12
Herds tested by SIT (%)	12,439 (5.5)
Herds tested by SICTT (%)	2,283 (1)
Number of control SITs	497,432
Number of control SICTTs	180,653
Number of SICCT-positive herds (%)	82 (3.6)
Number of non-negative SICTT herds (%)	749 (32.8)
Number of SIT-positive herds (%)	336 (2.7)
Number of non-negative SIT herds (%)	596 (4.8)
Number of non-negative SITs (%)	1,373 (0.28)
Number of positive SITs (%)	1,133 (0.23)
Number of non-negative SICTTs (%)	1,158 (0.64)
Number of positive SICTTs (%)	235 (0.18)
Veterinary professionals involved in controls	1,101
Veterinary professionals reporting non-negative intradermal tuberculin tests (%)	191 (17)
Number of tests on movement	107,435

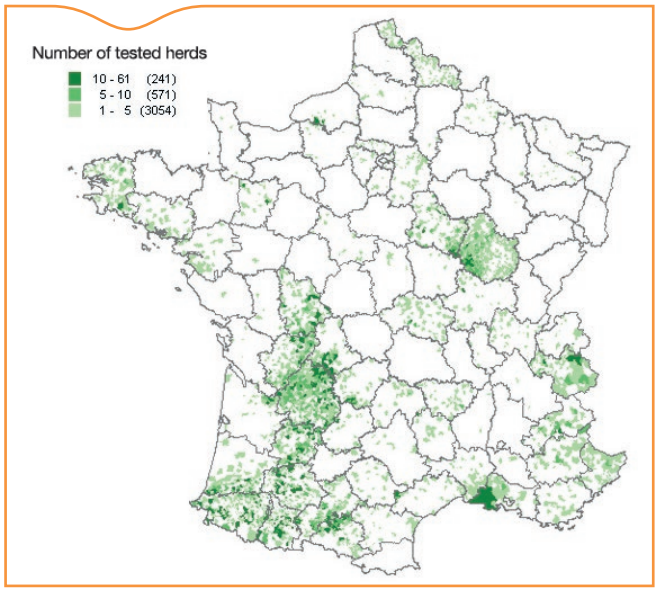

Figure 2. Number of cattle farms undergoing tuberculin testing per municipality in 2012 as part of the annual prophylaxis programmes scheduled in SIGAL (mainland France)

The geographic distribution of tested farms (Figure 2) is consistent with the screening intervals by *département* (Figure 1). Screening was carried out primarily in *départements* that have established zoning, but also in at-risk herds located in *départements* where tuberculosis programmed screening has been discontinued. Overall for 2012, 14,722 cattle farms were tested using single intradermal tuberculin tests (SIT) or single intradermal comparative tuberculin tests (SICTT), accounting for about 6.5% of farms (Table 1).

The choice between SICTT and SIT is related to the risk of infection, on the basis of complex interactions associated with the different ways in which surveillance is organised. The Côte d'Or *département* has used SICTT systematically since 2009, and in 2012, 65% of herds tested with this method were in Côte d'Or *versus* 83% in 2011. The number of *départements* using SICTT increased from 14 in 2011 to 31 in 2012. This significant increase is related to the subsidy scheme implemented by Ministerial Order on 30 October 2011, that covers the cost difference between SIT and SICTT. Moreover, data show that in herds tested using SICTT, the mean number of tested animals was 82, twice the number in herds tested using SIT with a mean of 41. This difference reflects the larger herd size in *départements* that systematically use SICTT, such as Côte d'Or, but also a difference in the minimum age of tested animals, especially when the tests are performed on farms classified at risk.

Tuberculin testing including 180,653 SICTTs and 497,432 SITs was carried out by 1,101 veterinary practices (either veterinarians or veterinary associations or clinics) accounting for 42% of 2,627 veterinary practices. Half of the veterinary practices who carried out intradermal tuberculin testing performed at most 151 tests, but a quarter of them performed 84% of all tests, with a mean of 1,940 intradermal tests per professional.

In *départements* where cattle is raised for bullfighting and where conditions for carrying out intradermal tests are particularly difficult, first-line tuberculosis screening using interferon-gamma (IFN-gamma) was scheduled as an alternative or in addition to intradermal testing. Based on available data, testing involved 101 herds in the Camargue area (in Bouches du Rhône, Gard and Hérault) and 29 herds in Landes, accounting for 7,987 and 2,139 cattle, respectively.

Testing on animal movement involved 107,435 cattle in 10,054 herds by intradermal testing and 1,229 cattle by IFN-gamma. Follow-up of these findings has however been hindered by data processing issues for the corresponding results and a large amount of data is missing.

In order to boost the surveillance system, training and awareness sessions were organised. In 2011 and 2012, the half-day training session on bovine tuberculosis organised by the Ministry of Agriculture, Food, and Forestry and the National society of veterinary technical groups (SNGTV), as part of occupational training for animal health accreditation, was attended by 427 mandated veterinarians from 48 *départements*. In addition, in 2011 and 2012, 67 staff members from Departmental Directorates for Protection of the Population (DDecPP) took part in a 2-day training session on tuberculosis management, organised by the DGAL. In 2012, tuberculosis was addressed in 73 meetings organised by the DDecPP with mandated veterinarians, and in 140 meetings for livestock breeders, in 53 and 37 *départements* respectively. 62 meetings were organised in 36 *départements* for implementation and follow-up of the Sylvatub scheme.

Results of programmed surveillance

Tuberculin testing

The rate of SIT testing by herd was calculated on the basis of the number of herds tested for which at least one test was planned. This rate was determined for 40 *départements*. It was 100% in 19 *départements*, between 90% and 100% in 9 *départements*, and below 90% in 12 *départements*. The mean rate was 92%. For SICTT, the rate was calculated for 15 *départements*, and was 100% in 9 *départements*, between 90% and 100% in 2 *départements*, and below 90% in 4 *départements*. The mean was 95%.

According to available data for 2012, 2531 non-negative reactions (0.4%) were observed on 1,345 farms (10.8%), a figure consistent with 2011 (Fediaesvky *et al.*, 2011). The number of farms with non-negative intradermal testing was slightly higher in 2012.

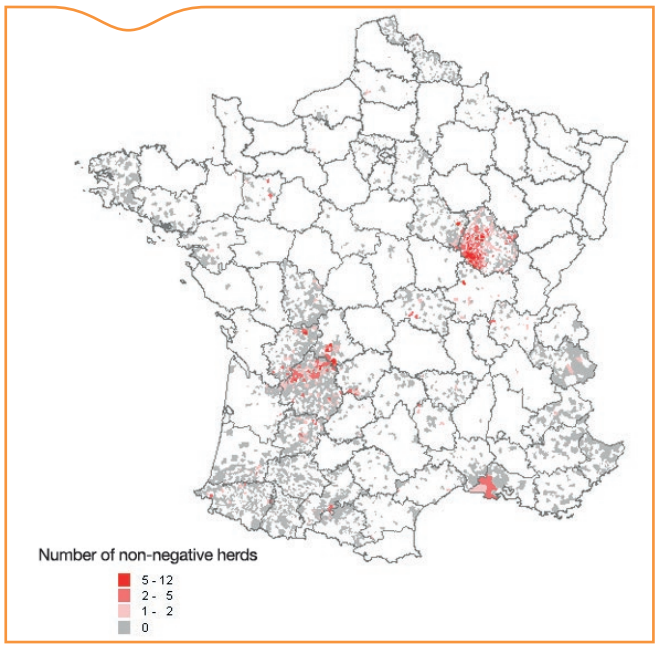

Figure 3. Distribution by municipality of the proportion of farms (%) with a non-negative reaction among all the farms undergoing tuberculin testing in 2012 (mainland France)

The geographic distribution of farms with at least one non-negative test reaction was highly variable (Figure 3) and did not correspond to the distribution of tested farms. The geographic correlation between screening and detection of non-negative results must however not be interpreted hastily, specifically because of the large number of factors affecting the sensitivity and specificity of intradermal tests.

On average, 4.8% of herds tested by SIT had at least one non-negative reaction *versus* 32.8% of herds tested *via* SICTT. This finding may appear surprising given the higher specificity of SICTT compared to SIT. One explanation is that SICTT as programmed screening is recommended for use in areas where non-negative reactions are particularly common, in order to minimise the risk of false-positive reactions, and operators have a high degree of awareness in terms of detecting reactions. In addition, this result is strongly influenced by Côte-d'Or where most SICTTs were performed (44%). Figure 3 shows that reactor farms are concentrated in the western half of the *département*. This proportion is 15% if Côte d'Or is excluded. It is also possible that the results of control measures were inadvertently added to programmed surveillance results when the SIGAL database was used.

The non-negative results were reported by 191 veterinary practices in 41 *départements*, i.e. an increase of 36% *versus* 2011 (Table 1). Veterinary practices who reported non-negative reactions carried out 51% of tuberculin tests country-wide.

Box 2. Sylvatub: Tuberculosis surveillance in wildlife

Since the detection of the first red deer with tuberculosis in the Brotonne forest (Seine-Maritime) in 2001, infected wild animals have been found in several *départements*, chronologically Côte-d'Or, Corsica, Pyrénées-Atlantiques, Dordogne and Charente, then Ariège (ANSES, 2011; Hars and Richomme, 2010). At the end of 2011, on the initiative of the Ministry of Agriculture, a national surveillance scheme known as Sylvatub was established with several outbreak and programmed surveillance systems, as part of the National Epidemiological Surveillance Platform for Animal Health. Its aim was to carry out an integrated assessment of sampling procedures, to harmonise diagnostic methods, and to centralise data from various surveillance systems (Rivière *et al.*, 2013). The results presented correspond to the first year of activity of Sylvatub.

Outbreak surveillance:

A total of 121 suspected cases were reported *via* outbreak surveillance involving examination of carcasses of animals killed during hunting or reports from the SAGIR network. Cases concerned 30 wild boars, 20 red deer, 12 roe deer, and 59 badgers in 24 *départements*. Among these animals, 3 boars, 1 roe deer and 3 badgers were found to be infected and all were found in *départements* known to be infected by bovine tuberculosis (Côte-d'Or, Haute-Corse, Dordogne and Charente) and near bovine outbreaks (Rivière *et al.*, 2013).

Programmed surveillance:

In Côte-d'Or, programmed surveillance in 2011-2012 in the infected area found 17 cases in the wild boar population among 210 tested animals. Only one infected red deer was found among 149 animals. In 2012, results from the infected area show nine infected badgers among 306 tested animals. In addition, an infected badger was detected in a buffer zone, part of a neighbouring area. Surprisingly, the *M. bovis* strain isolated in this animal was the BCG spoligotype which is usually found further north in the *département*.

The 102 wild boars, 12 red deer and 68 badgers tested in the south-east of Yonne, neighbouring Côte-d'Or, were not infected with *M. bovis* (Rivière *et al.*, 2013).

In Dordogne over the same period, only two infected wild boars were discovered in the infected area, of 261 tested animals. Seventeen badgers were found to be infected among 446 animals in 2012 in this same area. Following detection of a roe deer with tuberculosis through outbreak surveillance, programmed surveillance was implemented in a few of the surrounding municipalities, but no other infected roe deer were detected among 41 tested animals.

In Pyrénées-Atlantiques, programmed surveillance identified six infected wild boars in the infected area, among 87 animals. In addition, of 91 badgers, one infected animal was detected for the first time in this *département*, in a municipality where infected wild boars had also been found and where infection had also been detected in cattle.

In the Brotonne forest, no infected red deer were detected among the five animals shot during the 2011-2012 hunting season. However, two young wild boars with tuberculosis were found among 200 tested animals, indicating probable persistence of a source of infection despite the effectiveness of the control strategy involving slaughter of the red deer population. (Rivière *et al.*, 2013).

Importantly, wild animals found to have tuberculosis were all detected in areas showing bovine infection, with exact correlation of isolated spoligotypes in cattle and wildlife, with the exception of one badger in Côte-d'Or in 2012.

Maps showing these results are available on the website of the ESA Platform (www.plateforme-esa.fr).

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Table 2. Distribution of cattle farms based on the type of reaction and status in 2012

		SIT testing		SICTT testing	
	Status of farms	Herds confirmed infected	Herds not confirmed infected	Herds confirmed infected	Herds not confirmed infected
Herds in which at least one positive reaction was found	Mean number of cattle presenting a positive reaction	3.1	3.4	1.5	2.9
	Mean number of cattle presenting another type of reaction	0.7	0.6	3.8	2.5
	Number of tests	72	75	112	109
	Number of herds	30	336	12	82
Herds in which no positive reactions were found	Mean number of cattle presenting a positive reaction	0	0	0	0
	Mean number of cattle presenting another type of reaction	3	4.3	4.1	3.2
	Number of tests	24	57	127	118
	Number of herds	3	201	9	564
	Total number of herds	**33**	**537**	**21**	**646**

On average, the rate of non-negative reactions by veterinarian was 0.9% but among the veterinarians who reported a non-negative reaction, it was 5%. Among veterinarians who reported a non-negative reaction, half detected at least 5 non-negative reactions and tested at least 789 cattle throughout the year. They represented 51% of veterinary practices in Côte d'Or, 35% in Charente, 34% in Dordogne, 26% in Pyrénées-Atlantiques. In areas where cattle is bred for bullfighting and where intradermal testing is particularly difficult to perform, they accounted for 5% of practices in *départements* within the Camargue area, and 7% in Landes. In these areas, first-line screening by IFN-gamma alternating with intradermal testing is highly recommended.

Furthermore, it was reported in 9 *départements* that 656 cattle could not be tested although they were present; 66% of these cases were reported from Mayenne. This is not a specific issue for Mayenne as this indicator was recently included in the information system and was not reported in the same way in all *départements*.

Of the 1,345 farms where non-negative reactions were reported in 2012, 82 reported a positive SICTT reaction. 667 reported reactions that, although they were not positive, were classified mainly as "other", corresponding primarily to ambiguous responses. 336 reported a reaction considered SIT-positive, and 260 an SIT reaction not classified as positive but rather as «other», corresponding primarily to ambiguous responses (Table 1).

In the herds with positive SICTT results, the mean number of animals with a positive SICTT was 2.9 and the mean number of animals with a SICTT reported as "other" was 2.6. In the herds with positive SIT results, the mean number of animals with a positive SIT was 3.4 and the mean number of animals with an SIT reported as "other" was 0.6. In the herds reported to have only «other» SICTT results, the mean number of reactors was 3.3. Among those reporting only "other" SITs, the mean number of reactors was 4.3. The status of the herd appeared to have relatively little influence on the mean number of reactions by herd.

The infection confirmation rate for farms tested using SICTT was 3.1% (21/(21+646)) and 5.8% for those tested using SIT (33/(33+537)). Moreover, the confirmation rate for farms with at least one positive SICTT was found to be 12.7% (12/(12+82)), *versus* 1.6% in the absence of positive SICTT (9/(9+564)). Likewise, in herds tested using SIT, the confirmation rate for farms with at least one positive SIT was found to be 8.2% (30/(30+336)), *versus* 1.5% in the absence of positive SIT (3/(3+201)).

This appears to show a high predictive value of positive reactions but it is important to note that differences in diagnostic methods in herds in which only ambiguous SICTT results were observed hinder comparison of the data. It is however clear that, descriptively, and regardless of potential bias, the extent of reactions appears to be more closely associated with the status of the herd than with the number of reactor animals, and this should be assessed more closely in an analytical study.

Table 3. Surveillance of bovine tuberculosis at the slaughterhouse in 2012 based on reasons for inspection

Routine surveillance	ODF herds with suspected case at slaughterhouse (%)	167 (0.07)
	Cattle from an ODF herd with suspected TB lesions	171
	Cattle from an ODF herd with confirmed TB lesions (confirmation rate)	23 (13.4)
Diagnostic slaughter	Herds with diagnostic slaughter (%)	749
	Herds with confirmation on diagnostic slaughter (confirmation rate in %)	79 (10.5)
	Cattle undergoing diagnostic slaughter	1,355
	Cattle confirmed infected on diagnostic slaughter (confirmation rate in (%)	87 (6.4)
Partial depopulation	Herds undergoing partial depopulation*	45
	Cattle slaughtered under partial depopulation*	3,674
	of which, reactor cattle undergoing partial depopulation*	374
	Cattle confirmed infected under partial depopulation (%)*	23 (0.6)
	of which, reactor cattle confirmed infected (%)*	20 (5.3%)
Complete depopulation	Herds undergoing complete depopulation*	49
	Herds undergoing complete depopulation with lesions (%)*	29 (59.2)
	Cattle slaughtered under complete depopulation*	5 867
	Cattle slaughtered under complete depopulation with lesions (%)*	130 (2.2)

OI : officially disease-free

Programmed screening with IFN-gamma

On the basis of the available data, of the herds tested with IFN-gamma, 34 presented a non-negative reaction in Camargue (of which 4 were confirmed) and none in Landes.

Surveillance on animal movements

Data concerning screening on animal movements were collected at *département* level, so results per herd are not available. Collected data show that non-negative results were obtained for 109 herds across 32 *départements* and these reactions led to confirmed cases in two herds (1.8%).

Surveillance at the slaughterhouse

Based on collected data, 171 cattle from 167 officially disease-free herds in 42 *départements* (Table 3), presented suspect tuberculosis

Table 4. Number of outbreaks and detection circumstances

Incident outbreaks 2012 (herds) (%)	116 (0.051%)
Prevalent outbreaks 2012 (herds) (%)	169 (0.075)
Prevalent herds as of 31/12/12 (%)	90 (0.040)
Infected imported cattle	3
Proportion of herds undergoing complete depopulation (%)*	52.0
Outbreak detected at slaughterhouse (%)*	23.3
Outbreak detected on control screening (%)*	52.2
Outbreak detected on movement testing (%)*	2.2
Outbreak detected by epidemiological survey (%)*	22.2
Outbreak detected in another way (%)*	0.0
Veterinary fees (%)*	0.0
Compensation (%)*	0.7
Laboratory fees (%)*	0.1
Cleaning-disinfection (%)*	0.0
Sundry costs (%)*	0.0
State screening subsidies (%)*	0.0
Surveillance of wildlife (%)*	0.0
- of which laboratory fees (%)*	0.0

* see warning on page 4

lesions at the slaughterhouse. The confirmation rate for these lesions was 13.5% (23/171), a decrease compared to 2011 (22.6%). This confirmation rate, which is only slightly higher than the level of suspected cases on farms, is reassuring since it indicates that not only highly characteristic or extensive lesions would be detected. Gradual computerisation of slaughterhouse cases nationally will provide more specific data on the role of inspection sites on detection of slaughterhouse cases.

Surveillance of herds with likely infection

Available data indicate that 1289 herds with likely infection, i.e. with an epidemiological link to an infected herd, were identified in 2012 in 45 *départements*.

Tuberculin tests were carried out in 80% of these herds (1,032/1,289) and diagnostic slaughter was performed in 90% (1,164/1,280), *versus* 45% in 2011. However, data from certain *départements* appear particularly high and 32% of these herds were classified as a health risk, meaning that they will be included in the next prophylaxis programmes. These investigations led to confirmed infection in 20 farms, yielding an infection confirmation rate in these herds of about 1.5% (20/1,032).

Measures taken in suspect herds

Collected data indicate that 1,227 herds underwent tuberculin testing as a control measure when cases were suspected, and 959 suspect herds or likely infected herds were tested using IFN-gamma, including 432 that had a non-negative result. However, given the complexity of diagnostic protocols, the data need to be analysed at the departmental level.

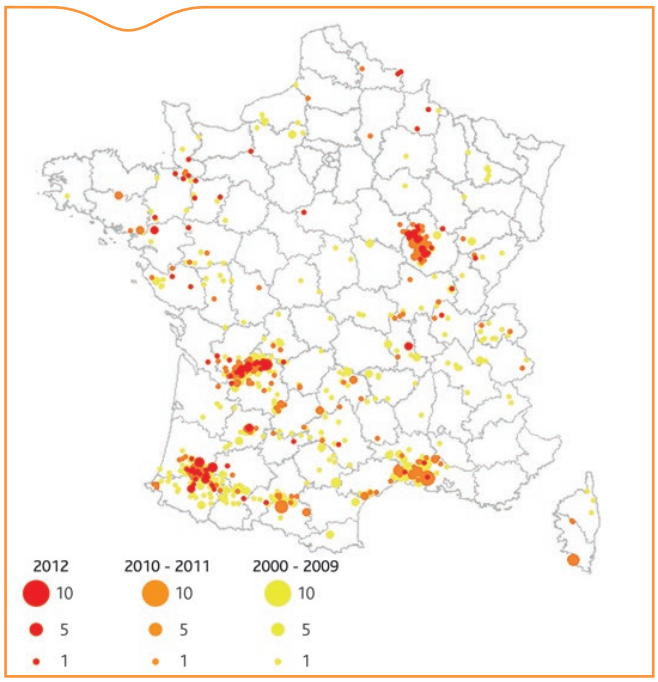

Figure 5. Geographic distribution by municipality of incident outbreaks from 2000 to 2012

Diagnostic slaughter

One or more diagnostic slaughter orders were issued for 749 farms, and a total of 1,355 cattle were slaughtered. The confirmation rate was 10.5% (79/749) at the farm level, and 6.4% (87/1,355) at the animal level (Table 3). These confirmation rates varied between *départements*, depending on the epidemiological context and local policy decisions. In Côte d'Or, the confirmation rate was 8.8% at herd level, it was 27.8% in Dordogne and 8.3% in the other *départements*. These differences are far less pronounced than in 2011, when rates of 4.5% were found in Côte d'Or and 46% in Dordogne, highlighting progress made in harmonisation of procedures.

Outbreaks

Incidence, prevalence and geographic location

In 2012, 116 herds were declared infected with tuberculosis, resulting in 169 prevalent herds for the year (Table 4). The incidence rate for 2012 was therefore 0.05% (116/224,514), *versus* 0.04% in 2011, and prevalence was 0.075% (169/224,514), *versus* 0.077% in 2011 (Figure 3)

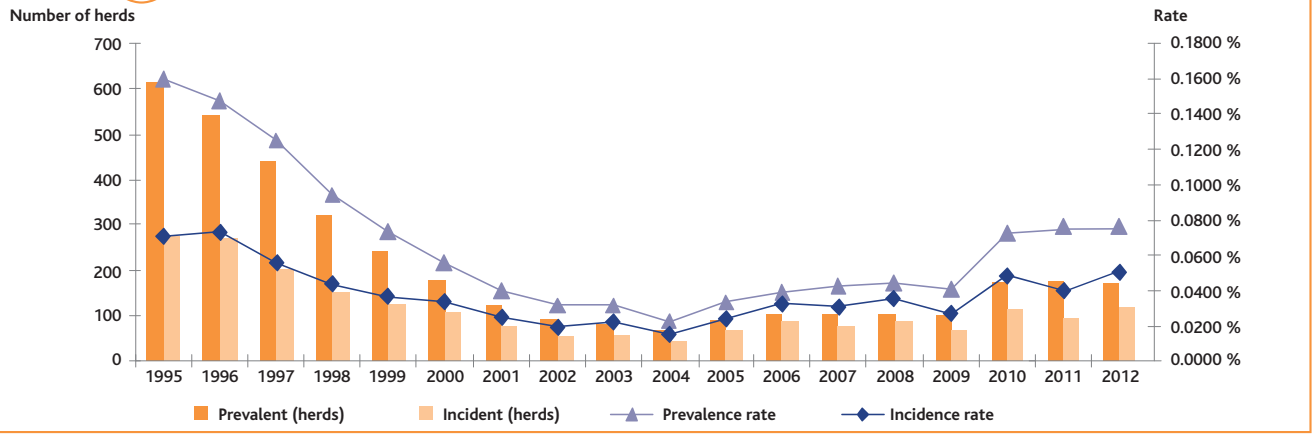

Figure 4. Distribution of detection means (%) for bovine tuberculosis outbreaks between 1995 and 2012

(Fediaesvky *et al.*, 2011).

Three infected cattle were introduced into France from other EU Member States in three *départements* (Bouches-du-Rhône, Gard and Pyrénées-Atlantiques). The origin of infection was considered to be in the country of origin since the genotypes of the isolated strains were characteristic of those in these countries.

The geographic location of the incidence outbreaks was basically the same as that in 2011 and 2010, with most in Côte d'Or (24%), Dordogne (25%) and to a lesser extent in an area between Landes (11%) and Pyrénées-Atlantiques (11%). The decrease observed in 2011 in Camargue appears to have been confirmed. Resuming IFN-gamma screening will partially help to address this question. Like in 2011, a few outbreaks were detected in other *départements* that were placed under close monitoring, particularly Ardennes, where secondary cases were detected in 2013. Investigations carried out following cases detected in 2011 enabled identification of secondary cases, specifically in Basse-Normandie, Brittany and Pays de la Loire.

Means of detection

Data concerning the means of detection of outbreaks in 2012 were recorded overall for each *département*, and certain inconsistencies regarding the number of incidence outbreaks were found. Nonetheless, the trends shown by these data are fairly clear (Table 4, Figure 9).

Overall, more than 70% of incident outbreaks in 2012 were detected on farms through screening programmes (52%), or on farms with epidemiological links with an outbreak (22%). The proportion of outbreaks detected through slaughterhouse screening was lower compared to previous years, which is reassuring given the objective of early case detection on farms.

Control of the disease

Data on the type of control measures were incomplete in nine *départements*. In the others, complete depopulation was carried out for 49 herds with a total of 5,867 cattle being slaughtered: 2.2% (130/5,867) of these animals presented suspect tuberculosis lesions, a proportion comparable to 2011 (2.7%). In all, 59.2% of herds (29/49) that underwent complete depopulation in the concerned *départements* had infected cattle (Table 3).

Partial depopulation was carried out in four *départements*, three on an exemption basis and one on an exceptional basis (Saône et Loire). Overall, for the *départements* providing data, 45 herds were subject to partial depopulation and 3,674 cattle were slaughtered. 1.2% (44/3,674) of these animals had lesions. Seven herds initially managed by partial depopulation were later subject to complete depopulation.

Cost

Cost data provided by the DDecPPs in 2012 was sometimes incomplete for *départements* with expenditure for bovine tuberculosis but no outbreaks, given that analytical spending is not broken down by disease type. On the basis of the information provided, total expenditure was 20.4 million Euros, with 74% accounting for farmer compensation, 12.6% for laboratory fees, and 4.6% for veterinary costs.

Discussion

Errors in collected data are possible due to different conditions of use of the SIGAL database, particularly the risk that programmed screening and control results are confused, and to incomplete or inaccurate data collection by the *départements*, a significant problem in 2012.

The overall health status did not show significant changes between 2012 and 2011. This is to be expected for a disease that evolves slowly and that is difficult to diagnose. Nonetheless, a number of parameters show an improvement in the monitoring system: increase in the proportion of veterinary practices reporting non-negative results, decrease in the number of outbreaks detected at the slaughterhouse

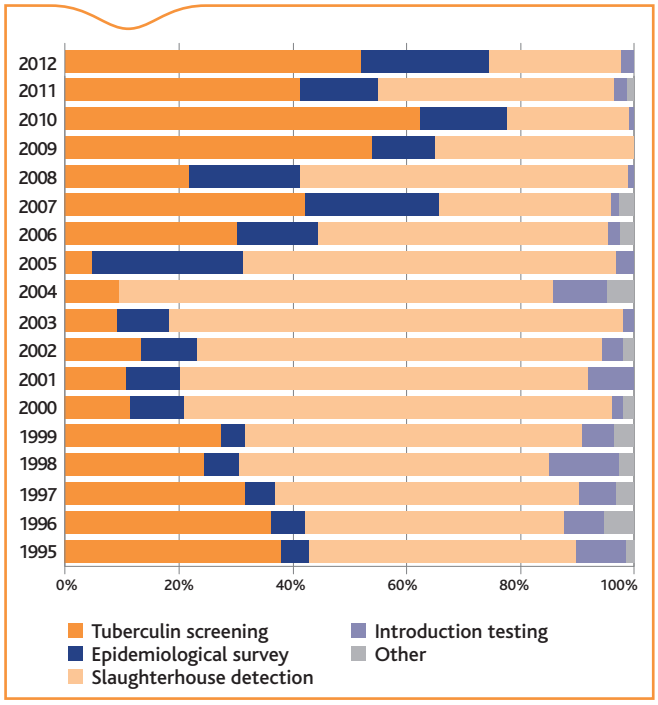

Figure 6. Distribution of detection means (%) for bovine tuberculosis outbreaks between 1995 and 2012

versus detection on farms, decrease in the confirmation rate for suspect cases at the slaughterhouse, and convergence of confirmation rates for diagnostic slaughter. The low confirmation rate for diagnostic slaughter is sometimes discouraging for veterinarians and livestock farmers. An experimental protocol using IFN-gamma was launched in 2013 to study to what extent IFN-gamma testing could replace retesting *via* the intradermal method after a six-week interval.

Moreover, the presence of tuberculosis in wildlife appears to be a lasting phenomenon in risk areas. The Sylvatub scheme has been made permanent and new control measures that take these factors into account need to be developed.

Given the complexity of health situations and monitoring programmes, it appears increasingly necessary to carry out specific local analysis in areas with a high prevalence, in addition to analysis of data at a national level. Epidemiology is being reinforced in 2013 with the creation of Inter-regional veterinary epidemiology groups (CIREV) in Aquitaine and in Burgundy.

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Two cases of **bovine brucellosis** in 2012; vigilance should be maintained

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Abstract

France has been recognised as officially free of bovine brucellosis by the European Commission since 2005. Two outbreaks of bovine brucellosis were confirmed in 2012 in France, while no case had been reported since 2003. The first outbreak, located in the Pas-de-Calais region, was due to an imported animal issued from an infected Belgian herd. The disease did not spread in France since the case was identified very rapidly after introduction. The second French outbreak was confirmed in a dairy farm in Haute-Savoie, through the notification of an abortion. It is highly probable that this outbreak was due to a previously unidentified local wildlife reservoir (Alpine ibex population). These recent cases highlight the importance of maintaining the national surveillance strategy, based on both the annual serological surveillance of all cattle herds as well as on abortion notification. This shows that, despite a generally well-implemented surveillance scheme, and even though abortion notification can still be improved, vigilance should be maintained throughout the country.

Keywords
Bovine brucellosis, surveillance, control

Résumé

Deux cas de brucellose bovine en 2012 appellent à la vigilance

La France est reconnue officiellement indemne de brucellose bovine par la Commission européenne depuis 2005. Deux foyers de brucellose bovine ont été confirmés en 2012 sur le territoire français, alors qu'aucun cas n'avait été rapporté depuis 2003. Le premier foyer situé dans la région Nord-Pas-de-Calais, a eu pour origine l'introduction d'un bovin issu d'un foyer confirmé en Belgique. La maladie ne s'est pas propagée en dehors du foyer, car l'introduction était récente. Le second foyer français a été confirmé dans une exploitation laitière de Haute-Savoie suite à un diagnostic conduit après un avortement. Il est très probable que ce foyer isolé soit dû à un réservoir sauvage local, silencieux jusqu'alors (population de bouquetins). Ces cas récents démontrent l'importance du maintien du dispositif de surveillance, fondé à la fois sur un dépistage sérologique annuel dans les troupeaux et sur la surveillance des avortements. Cela montre que malgré une réalisation globalement satisfaisante, même si la surveillance des avortements peut encore être améliorée, la vigilance reste de mise au plan national.

Mots clés
Brucellose bovine, surveillance, prophylaxie

Brucellosis caused by any *Brucella* other than *Brucella ovis* and *Brucella suis* biovar 2 is classified as a category 1 health hazard (Ministerial Order of 29 July 2013). Some *Brucella* species are found more specifically in certain animal reservoir hosts, for instance, *B. abortus* in cattle and *B. melitensis* in small ruminants. Given the risk to public health, these two species of *Brucella* are currently the *Brucella*e of interest in France for ruminants.

France has been recognised as officially bovine brucellosis-free since 2005 (Commission Decision 2005/764/EC) and had not reported any cases since 2003. The objectives of surveillance are to provide evidence for maintaining this favourable status for France, and to enable early detection of any re-emergence of brucellosis infections.

Surveillance system for bovine brucellosis

Current surveillance and control measures for bovine brucellosis have been in place since 2010 (Rautureau *et al.* 2012) (Box 1). Surveillance is based on notification of abortions and subsequent investigations, and on annual screening of herds.

Brucellosis screening campaigns on farms are organised during the wintering season between October and April, and not over the calendar year. As a result, the data presented here for the calendar year correspond to the end of the screening programme organised in 2011/2012, and to the start of the programme for 2012/2013.

Results of screening

Control screening data analysed for 2012 concern 194,328 tested farms[1] out of the 224,432 cattle farms that are officially free of the disease countrywide. Of these, 65.6% were assessed by serological blood tests, and 34.4% by bulk milk analysis (Table 1).

Concerning surveillance of abortions, a total of 70,853 abortions were recorded in 2012 (+14.8% *versus* 2011) on 36,807 different production sites (+6% *versus* 2011) (Table 1). Despite this increase, the number of reported abortions among cattle is most probably well below the actual number of abortions observed in the field, as demonstrated by recent analyses (Bronner *et al.*, 2013a).

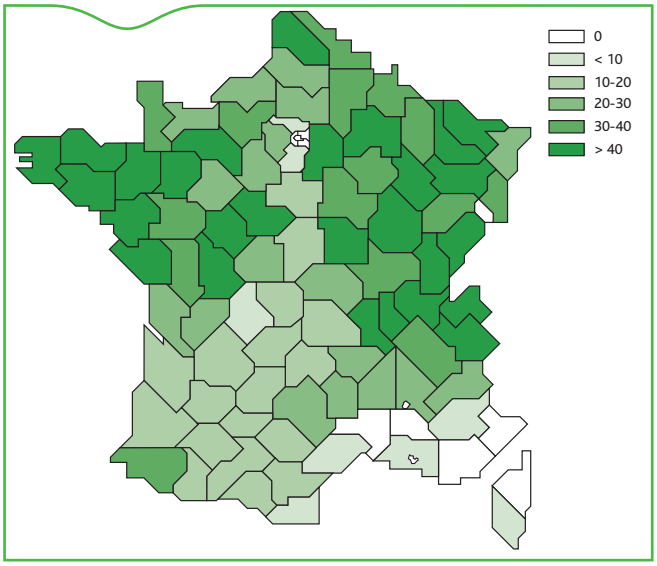

Figure 1. Distribution by *département* of the proportion of farms undergoing control screening and having declared at least one abortion in 2012 (%)

(1) Farms with at least one animal over 24 months, excluding fattening units under exemption.

Table 1. Regional distribution of the number of farms undergoing programmed screening (available data) and suspected serological and clinical cases

Region	Screened herds			Suspended herds following control screening	Reported abortions	Herds identified with a link to outbreak	Infected herds
	Total	Milk screening					
		Number	%				
Alsace	2,151	749	34.82	13	641	0	0
Aquitaine	12,403	1,953	15.75	9	2,995	0	0
Auvergne	15,652	3,980	25.43	12	3,130	0	0
Basse-Normandie	16,257	7,791	47.92	4	6,180	2	0
Bourgogne	9,200	852	9.26	2	3,381	0	0
Bretagne	21,748	12,954	59.56	1	13,842	0	0
Centre	5,533	1,041	18.81	5	1,890	0	0
Champagne-Ardenne	4,527	1,665	36.78	16	1,882	0	0
Corse	945	0	0.00	0	1	0	0
Franche-Comté	6,171	3,880	62.87	5	2,604	2	0
Haute-Normandie	5,916	2,323	39.27	2	1,738	1	0
Île-de-France	212	28	13.21	0	34	0	0
Languedoc-Roussillon	2,518	417	16.56	17	423	0	0
Limousin	9,046	525	5.80	10	1,237	0	0
Lorraine	7,915	3,336	42.15	19	3,585	0	0
Midi-Pyrénées	17,319	2,957	17.07	8	3,149	0	0
Nord-Pas-de-Calais	7,360	3,689	50.12	10	2,891	0	1
Pays de la Loire	21,296	8,897	41.78	7	10,572	3	0
Picardie	5,217	2,345	44.95	8	1,510	0	0
Poitou-Charentes	6,896	1,468	21.29	3	2,162	0	0
Provence-Alpes-Côte d'Azur	1,093	151	13.82	1	163	10	0
Rhône-Alpes	14,953	5,828	38.98	28	6,845	35	1
Total	**194,328**	**66,829**	**34.39**	**180**	**70,855**	**53**	**2**

The distribution of abortion reports per number of farms controlled this year (Figure 1) indicates that the reporting rate seems to be higher in specialised regions (Bretagne, Rhône-Alpes, Pays de la Loire), like in 2011.

Suspected and confirmed cases

Suspected cases from programmed screening

At the herd level, 0.16% of tested farms (205/127,343) presented at least one positive result on pooled serum ELISA test or on Rose Bengal test (RBT).

Moreover, 0.69% of farms tested using bulk milk had an initial positive ELISA result (461/66,829).

In total, 180 farms (130 tested through blood sampling and 50 through milk sampling) had their status suspended due to positive results confirmed as part of control screening by repeat of positive results using milk testing or individual positive blood testing. The investigations carried out as part of control measures in these herds included serological analyses (n=1,686) and/or diagnostic slaughter (n=40), with no subsequent confirmation of brucellosis.

All of these non-confirmed positive results can potentially be attributed to lack of specificity (cross reactions) and/or to the quality of practical testing.

Suspect abortions

Only 32 of the 70,853 reported abortions, i.e. 0.045%, were associated with a positive serological result both *via* RBT and complement fixation (CFT) testing, the regulatory definition of suspect animals.

Among these suspect animals, a case of *Brucella melitensis* biovar 3 was confirmed in April 2012 in Haute-Savoie in 21-head dairy herd (Box 2).

Concerning this case, 52 herds (45 cattle farms and 7 small ruminant farms) were identified (upstream/downstream, vicinity, equipment, etc.) as having a link to the outbreak in ten different *départements*. Investigations carried out included serological screening of 1905 cattle and 932 small ruminants from 39 herds. All of the findings were entirely favourable.

Moreover, reinforced screening was carried out in the area considered at risk during autumn 2012 on return from summer grazing. This involved testing of 8,522 cattle in 175 herds and 5,214 small ruminants in 53 herds. These investigations resulted in eleven diagnostic slaughter procedures for cattle and three for sheep. All subsequent bacteriological tests were found to be favourable (data not included in Table 1).

Other suspected cases

The first case of bovine brucellosis in France identified in 2012 was located in Pas-de-Calais and confirmed in March 2012. It was related to the recent introduction of an animal from the first Belgian outbreak confirmed in 2012 (Bronner *et al.*, 2013b). This *Brucella abortus* biovar 3 outbreak was detected during epidemiological surveys carried out following confirmation of the Belgian case (Box 2).

The Belgian authorities also reported previous transfers of animals from the infected farm to Seine-Maritime. These animals for slaughter had been housed for several days on a cattle farm before being sent for slaughter. The animals present at that time in this cattle farm were monitored by two serological tests, both negative. No other infected site was detected in France in relation to the outbreaks in Belgium.

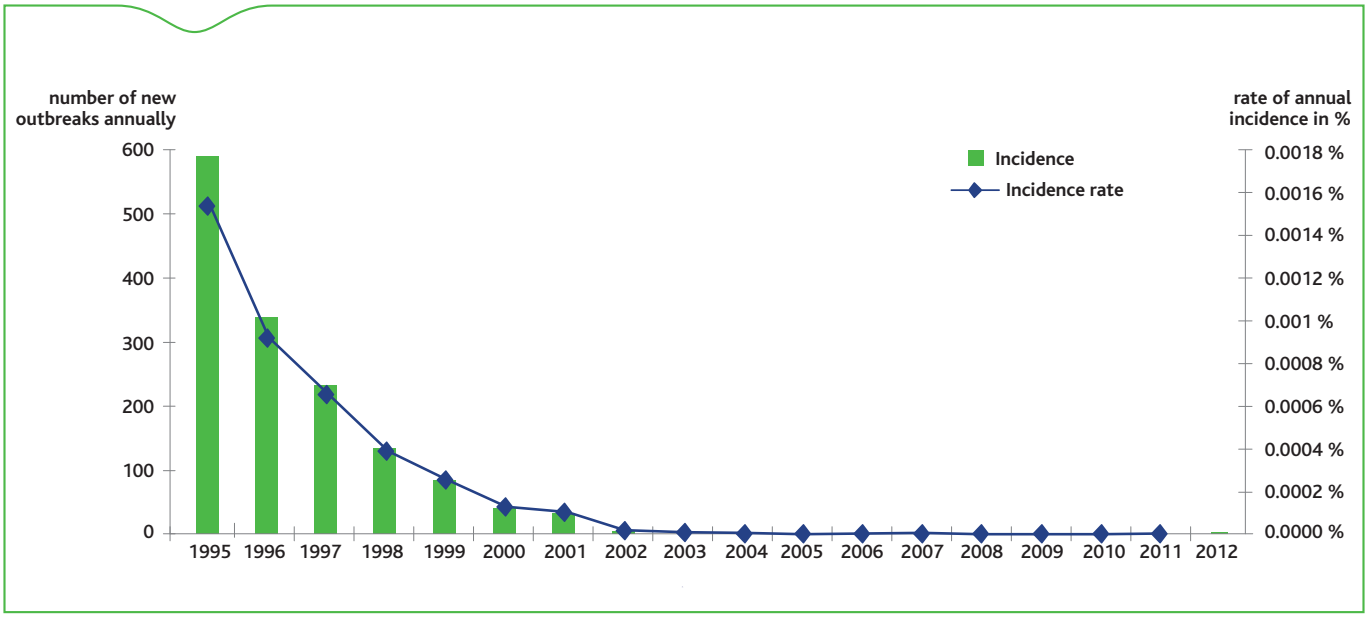

Figure 2. Changes in incidence (number and rate) of herds infected with bovine brucellosis in France between 1995 and 2012.

In total for 2012, 255 farms were placed under Prefectural Monitoring Orders (APMS) (farms considered suspect) *versus* 88 in 2011. Two cases of brucellosis were found in 2012, the first in the country since 2003 (Figure 2).

Cost

In 2012, the French government allocated nearly 5.6 million Euros (3.6 million in 2011) to surveillance and control measures for bovine brucellosis. The difference is mainly related to management of the two outbreaks and subsequent surveillance (farmer compensation, analyses and veterinary interventions).

Conclusion

The two cases of bovine brucellosis detected in 2012 underline the importance of maintaining a high level of vigilance because of potential re-emergence.

Highly responsive surveillance is required to identify cases of reappearance as quickly as possible, to prevent spread within herds and to prevent potential contamination of other farms. Caution is however needed to avoid «false alerts» mainly due to non-specific serological reactions.

As a result, improving the abortion reporting system has been identified as an important means for optimising brucellosis surveillance, and has been included in a priority work topic for the ESA Platform, "Surveillance of abortion-related diseases in livestock farming" (Bronner *et al.*, in press). Improving this system could involve revising the protocol for targeted surveillance and/or better interaction with differential diagnostic procedures.

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Box 1. Surveillance and control measures for bovine brucellosis

Objectives of surveillance

- Provide evidence of the country's officially bovine brucellosis-free status.
- Early detection of any emergence in domestic cattle.

Monitored population

Domestic cattle across the country.

Surveillance procedures

- Programmed surveillance:

Annual serological screening based on blood sampling from at least 20% of animals over 2 years of age, or on bulk milk.

- Outbreak surveillance:

Reporting of abortions and investigation; serological screening of each abortion and swab-sample of the uterine cervix in the aborting cow. If serological results are positive, bacteriological testing is performed on the swab.

Animal health rules

Suspected cases of infection arise from either two rounds of unfavourable controls following control screening, or a positive serological result following abortion.

When suspected cases result from programmed screening, animals with positive results are tested individually. If the test is once again positive, the positive animals are slaughtered and detection tests for Brucella are performed on lymph nodes through bacteriological culture.

The herd is considered infected when Brucella is detected on culture, or when the suspected farm has a direct epidemiological link to an infected farm, through animal movements, for example. The herd is then placed under Prefectural declaration of infection (APDI).

The whole herd is slaughtered if Brucella abortus or melitensis is isolated, or if Brucella-related abortion has occurred.

Regulations

- Council Directive 64/432/EEC of 26 June 1964, as amended, on animal health problems affecting intra-Community trade in bovine animals and swine, establishing requirements for control measures applicable to intra-Community trade and import of animal sperm from the swine species
- Ministerial Order of 22 April 2008 establishing the technical and administrative framework for collective prophylaxis and control measures for bovine brucellosis

Box 2. Two outbreaks of brucellosis detected in France in 2012

2012 saw the re-appearance of cases of bovine brucellosis in France. Two outbreaks were detected in cattle while no cases had been identified for nearly ten years (Rautureau *et al*. 2012).

First outbreak discovered following cases in Belgium

The first case, located in Pas-de-Calais, was related to introduction of an animal from a Belgian outbreak. The outbreak in France was identified in March 2012 following an alert reported by Belgium to the French authorities indicating the recent sale of animals to France from the first holding identified as infected in Belgium in 2012, a farm in Namur province (Bronner *et al*. 2013). No other farms with an epidemiological link to this outbreak were found in France, since the infection was limited to one of two farms receiving animals from Belgium. Confirmation of the case in France with detection of *Brucella abortus* biovar 3 led to slaughter of all the cattle on the production site. Infection had not yet spread within the farm on the basis of serological findings.

However, in Belgium, six outbreaks were detected in 2012-2013 and all had an epidemiological link. The same biovar had been isolated in another province (Liège) in 2010, but its origin could not be determined at that time. *Brucella abortus* biovar 3 was historically the most common biovar isolated in cattle in Belgium and in France, and is still present in southern Europe. The exact origin of the Belgian outbreaks in 2012-2013 involving *B. abortus* bv. 3 has not been identified to date.

Second outbreak involving an indigenous case

The second outbreak in France was confirmed in a dairy farm in Haute-Savoie following a diagnosis related to an abortion. This case led to human contamination through consumption of raw milk cheese produced on the farm (Mailles *et al,* 2012).

Following abortion at seven months of gestation, serological testing was performed on the nine-year-old cow and results were found to be positive both on RBT and CFT. Bacterial sampling in the form of a vaginal swab was not performed before administration of antibiotics *via* the intrauterine route. On serological retesting several weeks later, the animal still had a positive result, prompting the DDecPP to request bacteriological testing on milk samples. In April 2012, these tests revealed *Brucella melitensis* biovar 3 in milk from this animal.

This bovine case has rather unique characteristics, indicating the high variability in pathogenesis and immune response to brucellosis. Surprisingly, all the other animals in the herd were found to have negative serological test results (RBT, CFT and ELISA). In-depth bacteriological tests performed on the entire herd after slaughter revealed *B. melitensis* bv. 3 in the aborting animal and in only one other animal. Four other animals showed positive results for *Brucella*-specific PCR on lymph nodes (Brucellosis NRL data), although no serological reaction was found. Intra-herd spread therefore appears to have started, but to a very limited extent without causing serological reactions or clinical repercussions in the other animals in the herd. These findings may suggest low-grade circulation and therefore most probably recent infection of the herd (Jay *et al.* in press).

Surveys conducted since summer 2012 have not identified any other infected domestic herd in the area surrounding the farm, while several cases have been detected in wildlife (chamois and especially Alpine ibex), with identified *Brucella* strains belonging to the same genetic group (Brucellosis NRL data). Surveillance and control measures have been implemented to limit or even eliminate the risk of recontamination of domestic ruminant herds from this reservoir in wildlife.

Farms with a link to the outbreak, through purchase or sale of animals or geographic proximity, were identified very rapidly and were all found to be seronegative. Likewise, all the herds in the same mountain area (summer grazing or local herds) had favourable results on extensive serological testing carried out in the autumn after return from summer grazing.

Regulations

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No brucellosis outbreaks detected in **sheep and goats** in France in 2012, but vigilance has to be maintained

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Abstract

No outbreak of sheep and goat brucellosis has been reported in France since 2003. Vaccination was stopped in the country in early 2008. In 2012, 64 departments were officially recognized as free by the European Commission. The national surveillance programme aims at detecting any reintroduction and extending this disease-free status throughout the whole country. It consists of annual serological surveillance within flocks as well as abortion notification. The implementation of this surveillance is satisfactory as regards serology but not for abortion notification. Positive serological reactions were regularly notified, but none were confirmed after specific investigations. Cross-reactions, well known in brucellosis serology, explain these false positive results which need appropriate management. Despite Brucella melitensis (the main Brucella species in small ruminants) being isolated from a bovine outbreak in 2012, no infection was detected in small ruminants during this episode.

Keywords
Sheep and goat brucellosis, surveillance, control

Résumé

Aucun foyer de brucellose ovine et caprine détecté en France en 2012 mais une vigilance à maintenir

La France n'a connu aucun foyer de brucellose ovine et caprine depuis 2003 et la vaccination contre la maladie n'est plus pratiquée sur le territoire depuis début 2008. En 2012, 64 départements étaient reconnus officiellement indemnes par la Commission européenne. La surveillance, fondée sur un dépistage sérologique régulier dans les troupeaux (surveillance active) et sur la surveillance des avortements (surveillance évènementielle), vise à détecter une réintroduction de l'infection. Elle contribue, avec la police sanitaire, à maintenir le statut indemne (pour les départements reconnus comme tels) et à l'étendre à l'ensemble du territoire national. La réalisation de la surveillance sérologique est satisfaisante mais la surveillance des avortements est insuffisante et mérite d'être réactivée. Dans le cadre de la surveillance active, des réactions sérologiques positives sont régulièrement constatées mais, après investigations, ne sont pas confirmées comme d'origine brucellique. Bien que Brucella melitensis (souche prédominante chez les petits ruminants) ait été isolée dans un foyer bovin en 2012, aucun élevage de petits ruminants n'a été atteint en lien avec cet épisode.

Mots clés
brucellose ovine et caprine, surveillance, prophylaxie

Brucellosis caused by any *Brucella* other than *Brucella ovis* and *Brucella suis* biovar 2 is classified as a category 1 health hazard (Ministerial Order of 29 July 2013). *Brucella melitensis* is found specifically in small ruminants and represents the main reservoir for this bacterium.

Surveillance system

Screening procedures

Since 2006, 64 of the 101 *départements* in France have been recognised as officially ovine and caprine brucellosis-free (Commission Decision 2006/169/EC).

Based on available data, 87% of small ruminant herds had a health qualification for brucellosis as of 31 December 2012. Nearly 13% lacked qualification, the same proportion as in 2011. It appears that these herds almost exclusively involve small-scale owners with no commercial activity.

Given the difficulties in consolidating data from the national information system, some information concerning herd surveillance is incomplete. Caution should therefore be exercised when interpreting the calculated effective screening rates.

Programmed screening is carried out at different intervals depending on the *département* (ranging from annually to every 10 years) and in regulatory terms, only a proportion of sheep in each herd need to be tested (Box). Data collected for 95 *départements* (Table 1) indicate that 47,970 farms underwent serological controls in 2012 involving about 1,600,000 animals out of 114,031 production sites accounting in total for 6.7 million animals over 6 months of age.

Abortion surveillance

Data concerning screening of abortions were available for 86 *départements* (Table 2). In these *départements*, a total of 4,643 abortions were reported for 2912 herds.

Overall, there was an improvement in data reporting in the national information system. In 2011, data on abortions were available for only 58 *départements* (Rautureau *et al.*, 2012). The number of reported abortions therefore increased by 55% *versus* 2011 (1,538 herds and 2,576 abortions). The number of farms in these *départements* is 106,918, yielding a mean rate of abortion notification by herd of 2.7% (+ 1.9% *versus* 2011), with significant geographic differences. These differences should be interpreted with caution, given the risks of partial transmission of data to the national information system.The veterinarian is called on when there are on average 1.6 abortions in a herd. Analysis of the available data shows that of the 4643 reported abortions, 15 showed a positive result for the Rose Bengal Test (RBT), a seropositive rate of 0.32%.

Even in *départements* where abortions are notified to the authorities, the proportion of herds for which abortions are reported is extremely low, as mentioned in previous years (Rautureau *et al.*, 2012). There is concern that the system may lack sensitivity and responsiveness in terms of early detection of brucellosis in the event of re-appearance.

Suspected and confirmed cases

On the basis of available data for 2012, of the 1.6 million individual tests performed using the RBT method during control screening, 1,730 (0.11%) were positive. Of these 1,730 samples that yielded a

Table 1. Results of programmed screening and management procedures following suspicion of brucellosis in small ruminants in 95 *départements* in France in 2012

Region	Number of *départements*	Number of herds	Screened herds		Herds undergoing serological tests following suspected cases		Total number of diagnostic slaughter and cultures
			Number	Proportion (%)	Number of initial tests	Number of follow-up tests	
Alsace	2	1,351	517	38.3	3	1	0
Aquitaine	5	9,401	5,516	58.7	50	6	0
Auvergne	4	6,739	1,950	28.9	22	11	5
Basse-Normandie	3	9,417	4,814	51.1	33	10	5
Bourgogne	4	5,075	1,562	30.8	4	0	0
Bretagne	4	8,836	4,315	48.8	1	0	1
Centre	6	5,787	1,589	27.5	67	2	7
Champagne-Ardenne	3	1,070	259	24.2	3	2	0
Corse	2	925	684	73.9	4	3	1
Franche-Comté	4	2,580	373	14.5	1	4	2
Haute-Normandie	2	5,505	948	17.2	0	4	0
Ile-de-France	8	584	128	21.9	1	0	0
Languedoc-Roussillon	5	3,265	1,742	53.4	2	1	0
Limousin	3	5,027	1,183	23.5	3	0	0
Lorraine	4	2,926	599	20.5	0	14	4
Midi-Pyrénées	8	11,987	9,155	76.4	215	7	9
Nord - Pas-de-Calais	2	2,249	728	32.4	15	6	4
Pays de la Loire	5	8,677	1,459	16.8	0	11	2
Picardie	3	2,130	883	41.5	33	2	0
Poitou-Charentes	4	7,750	820	10.6	105	0	28
Provence-Alpes-Côte d'Azur	6	3,946	3,246	82.3	47	4	55
Rhône-Alpes	8	8,078	5,500	68.1	11	2	2
Total	**95**	**113,305**	**47,970**	**42.3**	**620**	**90**	**125**

Box. Surveillance and control measures for ovine and caprine brucellosis in 2012*

Objectives of surveillance

- Early detection of any emergence in domestic sheep and goats.
- To provide evidence on the status of the 64 *départements* considered officially ovine and caprine brucellosis-free, and to extend this status to the whole country.

Monitored population

Domestic sheep and goats across France.

Surveillance procedures

- Programmed surveillance:

Mandatory serological screening at variable intervals depending on the *département* and type of livestock farm.

In sheep, programmed screening generally covers a sample of animals in each herd (25% of animals over six months of age), while in goats, 100% of animals over six months of age are tested. Irrespective of their status, the *départements* implement longer intervals for control screening, with testing every 2 to 10 years, by pooling farms by municipality or by canton.

Annual screening is carried out on farms that produce raw milk. Most *départements* with transhumance involving border areas have also retained annual screening, whether animals pass through the *département*, or it is a departure or arrival point.

First-line serological testing involves RBT with additional complement fixation testing (CFT) if results are positive.

- Outbreak surveillance:

Notification of abortions and investigations with serological screening of each abortion, as well as vaginal swab sampling for aborting female animals. If serology is positive, bacteriological testing is carried out on the swab.

Animal health rules

Suspected cases of infection arise from an unfavourable initial test following control screening or an abortion.

In suspected cases following programmed screening, all adult animals within the herd are tested individually. Subsequently, there is serological follow-up of positive animals or diagnostic slaughter to detect *Brucella* in lymph nodes by bacteriological culture, in order to determine the status of the herd.

In cases of abortion, serological and bacteriological testing is performed.

The herd is considered to be infected when *Brucella* is detected on culture or when the suspected herd has a direct epidemiological link to an infected herd, through animal movements for example. The herd is then placed under Prefectural declaration of infection (APDI).

Complete depopulation takes place when *B. abortus* or *B. melitensis* are isolated, or if *Brucella*-related abortion occurred.

Regulations

– Council Directive 91/68/EEC of 28 January 1991, as amended, on animal health conditions governing intra-Community trade in ovine and caprine animals

– Ministerial Order of 13 October 1998 establishing the technical and administrative framework for collective prophylaxis and control measures for ovine and caprine brucellosis

* New Ministerial Order of 10 October 2013 establishing the technical and administrative framework for collective prophylaxis and control measures for ovine and caprine brucellosis, amending the provisions of 13 October 1998.

Table 2. Abortion notification for small ruminants in 86 *départements* in 2012

Département	Herds (number)	Abortions (number)	Interventions (number)	Reporting herds	
				Number	Proportion (%)
01	947	34	22	21	2.2
02	738	29	31	15	2.0
03	1,904	61	61	52	2.7
04	1,010	17	17	27	2.7
05	1,24	407	137	89	7.9
06	417	7	7	6	1.4
07	1,672	126	148	48	2.9
08	511	44	24	21	4.1
09	1,031	41	26	22	2.1
10	250	11	7	5	2.0
11	483	47	27	24	5.0
12	3,134	577	299	250	8.0
13	613	6	6	6	1.0
14	2,364	5	5	2	0.1
15	1,252	33	33	13	1.0
16	2,009	81	64	37	1.8
17	942	37	26	13	1.4
18	994	66	31	28	2.8
19	1,161	21	22	11	0.9
2A	360	71	17	12	3.3
2B	565	99	14	13	2.3
21	642	29	28	16	2.5
23	1,275	25	14	11	0.9
24	1,806	9	9	9	0.5
25	582	8	7	7	1.2
26	1,019	120	120	43	4.2
27	1,604	17	16	10	0.6
28	655	5	5	5	0.8
29	1,926	7	7	1	0.1
30	1,032	14	8	8	0.8
31	2,087	29	16	15	0.7
32	854	6	2	2	0.2
33	2,275	17	17	9	0.4
34	623	14	3	3	0.5
35	3,266	15	13	12	0.4
36	1,295	268	201	135	10.4
37	882	138	138	57	6.5
38	1,262	65	65	38	3.0
39	648	11	7	7	1.1
41	1,498	51	51	20	1.3
42	962	95	98	46	4.8
43	1,780	97	53	47	2.6
45	463	24	24	7	1.5
46	1,285	255	255	108	8.4
47	1,106	2	2	1	0.1
48	832	53	52	41	4.9
49	1,387	36	36	18	1.3
50	5,111	43	43	32	0.6
51	309	1	1	1	0.3
52	726	28	22	13	1.8
53	1,770	22	22	9	0.5
54	869	24	16	16	1.8
55	790	24	10	4	0.5
56	1,416	57	23	13	0.9
57	533	15	5	5	0.9
58	1,125	46	46	24	2.1
59	1,369	1	1	1	0.1
60	655	5	4	4	0.6
61	1,942	13	11	5	0.3
62	880	2	2	1	0.1
63	1,803	33	23	19	1.1
64	3,550	1,566	657	462	13.0
65	1,207	100	55	48	4.0
66	295	1	1	1	0.3
67	784	9	9	4	0.5
68	567	14	12	8	1.4
69	642	152	96	36	5.6
70	1,183	13	2	2	0.2
71	2,470	123	123	47	1.9
73	784	122	90	36	4.6
74	790	83	70	27	3.4
76	3,901	36	36	20	0.5
78	380	24	24	5	1.3
79	2,630	908	271	187	7.1
80	737	5	5	5	0.7
81	1,940	217	217	83	4.3
82	449	25	9	8	1.8
83	466	9	9	8	1.7
84	316	20	10	7	2.2
85	1,417	103	107	41	2.9
86	2,169	491	409	271	12.5
87	2,591	30	30	27	1.0
88	734	10	6	4	0.5
89	838	59	59	34	4.1
90	167	1	1	1	0.6
91	86	11	11	2	2.3
Total	**106,918**	**4,643**	**2,937**	**2,912**	**2.7**

positive RBT result on initial testing, 179 (10.3%) were positive on complement fixation testing (CFT).

Following detection of these positive results (RBT+ and/or CFT+), which correspond to the regulatory definition of suspect animals, additional investigations were carried out on more than 620 farms (Table 1).

To determine the status of these farms, serological testing and diagnostic slaughter (206 cases) were carried out, either directly or after a second serological testing. A total of 90 farms required two serological tests. No cases of brucellosis were detected through these procedures.

Following the *B. melitensis* bovine outbreak in Haute-Savoie (see article concerning bovine brucellosis in this issue), investigations were carried out in seven herds of small ruminants with a link to the infected bovine production site, with a total of 932 screened animals. Moreover, reinforced screening was performed on return from summer grazing in autumn 2012 for herds in the vicinity of the bovine outbreak. This involved screening of 5,214 small ruminants in 40 herds. Three diagnostic slaughter procedures were ordered. The results of all these investigations were found to be favourable.

Cost

The French government allocated nearly 960,000 Euros to surveillance and control of brucellosis in small ruminants in 2012.

About 43% of this amount was used to pay veterinary fees, and 55% for laboratory tests. In addition, subsidies were paid for control screening in 25 *départements* for raw-milk producing or transhumant herds, for which the control rhythm is still annual. This accounted for nearly 35% of the total amount spent by the government.

Discussion

The health status of France concerning ovine and caprine brucellosis for 2012 appears to be satisfactory. No outbreaks in small ruminants were detected.

However, the two cases of bovine brucellosis in 2012 are a reminder of the importance of maintaining high levels of vigilance. Like the system in place for cattle farming, brucellosis surveillance in small ruminants is theoretically ensured by two complementary systems: periodic large-scale surveillance by control screening, and clinical surveillance based on abortion notification. However, the abortion surveillance system is not really functional, given the very low number of abortions reported.

The conclusions of ongoing efforts to improve the abortion notification system for cattle farming (Bronner *et al.* 2012) should subsequently be taken into account for small ruminants, specifically *via* the "Surveillance of abortion-related diseases in livestock farming" group of the ESA platform (Bronner *et al.*, *in press*).

Until these conclusions are available, recent changes to the surveillance system for brucellosis in small ruminants (Ministerial Order of 10 October 2013) should enable optimisation of programmed surveillance and help to put forward a more realistic, functional outbreak surveillance system. In this way, although all abortions will still need to be recorded by breeders, only episodes indicative of infectious disease, i.e. three abortions within seven days, will require samples to be taken to investigate brucellosis. In parallel, this reporting system for abortions will be enhanced by gradual implementation of differential diagnosis for other abortion-related diseases on the initiative of professionals.

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Report on surveillance and control of **enzootic bovine leukosis** in France in 2012

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Abstract

France has been officially disease-free with regard to Enzootic Bovine Leukosis in cattle, sheep and goats since 1999. Annual prevalence is below 0.01%. The aim of surveillance is to assess the officially disease-free status and to detect any recurrence of cases of enzootic bovine leukosis. All the cases detected in 2012 presented only serological reactions, which is consistent with the disease's pathogenicity, with less than 10% of infected animals developing tumoral forms.

Keywords

Enzootic bovine leukosis, surveillance, disease control

Résumé

Bilan de la surveillance et du contrôle de la leucose bovine enzootique en France en 2012

La France est officiellement indemne de leucose bovine enzootique chez les bovins, ovins et caprins depuis 1999. L'incidence annuelle est inférieure à 0,01 %. La surveillance vise à attester le statut officiellement indemne et à détecter une recrudescence éventuelle des cas. Tous les cas détectés en 2012 ne présentaient que des réactions sérologiques, ce qui est cohérent avec l'évolution de la maladie pour laquelle moins de 10 % des animaux infectés développent des formes tumorales.

Mots clés

Leucose bovine enzootique, surveillance, prophylaxie

No changes were made to the surveillance and control system for enzootic bovine leukosis (EBL) in 2012 (Rautureau *et al.*, 2012) (Box).

Results

France has been recognised as officially EBL-free since 1999 (Commission Decision 1999/465/EC).

In 2012, serological screening involved 46,638 farms, of which 70.1% were tested using blood analysis and 29.9% by bulk milk analysis.

Suspected and confirmed cases

Outbreak surveillance identified three animals in 2012 with suggestive lesions at the slaughterhouse, but the lesions were not confirmed as EBL virus-related lymphosarcoma.

In terms of programmed screening, 517 animals were tested using individual serological analysis (ELISA) following suspected infection as part of serological screening showing a positive blood or milk result. When control analyses are carried out on bulk milk samples, all animals included in the pool must be tested to determine which ones were positive. Among the tested animals, 21 had a positive result but ultimately only two cases were confirmed through a second individual control [ELISA or agar gel immunodiffusion (AGID)]. In comparison to 2011, the proportion of animals testing positive on individual control following suspected infection was lower (21/517 = 4.1% *versus* 25/422 = 5.9%) (Rautureau *et al.*, 2012).

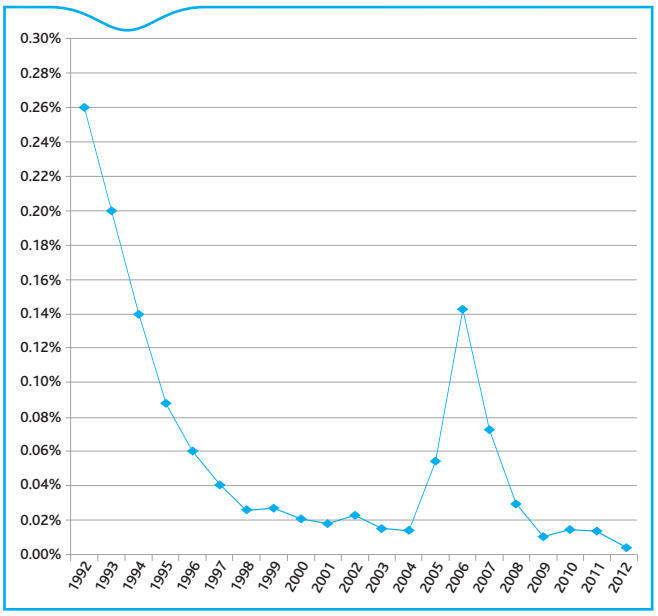

Figure 1. Change in incidence of enzootic bovine leukosis in France between 1995 and 2012 (percentage of infected herds)

In collaboration with accredited laboratories, the National Reference Laboratory (NRL) handles some of the first- and second-line control analyses. The NRL thus examined 105 samples by AGID from 25 production sites as first or second individual controls.

The two outbreaks were therefore latent forms that were detected through control testing of milk.

The annual incidence, as the percentage of of herds with at least one case per total number of tested herds in 2012, was therefore 0.0043% (95% CI [0.0005-0.0155]) in 2012 *versus* 0.0077% ([0.0024-0.013]) in 2011 (Rautureau *et al.*, 2012), a value that is not statistically significant.

The peak in incidence observed in 2006 (Figure 1) that was not confirmed subsequently, was related to false-positive serological reactions obtained with an ELISA kit withdrawn from the market in 2006.

Cost

The total amount spent by the French government in 2012 for the management of EBL, including control measures and slaughter procedures, was about 12,800 Euros, with 60% going towards laboratory analyses.

Overall, maintenance of the officially EBL-free status of France is relatively inexpensive for the government, particularly as a result of the low number of suspected cases to investigate, unlike surveillance of brucellosis.

Discussion

The health status in France concerning EBL therefore appears to be excellent and the country can be considered completely disease-free, even though a few sporadic cases are still observed.

Reports of suspected clinical cases in slaughterhouses or on farms suggest that outbreak surveillance is operational, but it is difficult to evaluate the system's effectiveness given the lack of data on the actual incidence of suggestive lesions of all causes.

An overview of these latent forms was presented in the 40th issue of the *Bulletin Épidémiologique – Animal health and nutrition* of November 2010 (Fediaevsky *et al.*, 2010).

On the whole, data suggest that programmed surveillance and outbreak surveillance work well together. Moreover, it is not surprising that no cases are detected by clinical surveillance, given the low levels of infection and the long disease course.

In this favourable context and the classification of the disease as a category 2 health hazard (Ministerial Order of 29 July 2013), it may be possible in the future to revise the surveillance system.

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Box. Surveillance and control measures for bovine leukosis

Objectives of surveillance
- Follow-up of the country's officially EBL-free status.
- Detection of any recurrence of cases in domestic cattle.

Monitored population
Domestic cattle across the country.

Surveillance procedures
- Programmed surveillance

Surveillance by serological screening every five years using blood samples from at least 20% of animals over two years of age, or on bulk milk.

- Outbreak surveillance

Surveillance of suspected enzootic bovine leukosis lesions at the slaughterhouse.

Animal health rules
Suspected cases of infection arise either from a positive result for a test performed on pooled blood samples or on bulk milk, or from suspect lesions identified histologically.

In this case, individual serological testing is performed on all animals over 12 months within the herd. If positive animals are detected, the herd is placed under Prefectural declaration of infection (APDI).

Cattle recognised as infected are isolated and slaughtered within 30 days.

Disease-free status can only be regained after two rounds of serological testing on all animals over 12 months, with a three to six month interval.

Regulations

– Council Directive 64/432/EEC of 26 June 1964, as amended, on animal health problems affecting intra-Community trade in bovine animals and swine, establishing requirements for control measures applicable to intra-Community trade and import of animal sperm from the swine species.

– Ministerial Order of 31 December 1990 establishing the technical and administrative framework for collective prophylaxis and control measures for enzootic bovine leukosis.

Bovine spongiform encephalopathy in 2012: for the first time since the beginning of programmed surveillance in France, no cases of classical BSE were identified

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Abstract

In 2012, a single case of atypical bovine spongiform encephalopathy (BSE) was detected among the 313,216 animals sampled during the rendering process, bringing the total number of atypical BSE cases identified in France since 1990 up to 28 (14 cases of L-BSE, 14 cases of H-BSE). The 937,725 animals tested in the context of the slaughterhouse programme all screened negative for BSE. For the first time since programmed surveillance began in 2000, no cases of classical BSE were identified during the year.

Keywords
BSE, epidemiological surveillance, control measures, cattle, France

Résumé

Encéphalopathie spongiforme bovine en 2012 : aucun cas d'ESB classique identifié pour la première fois depuis le début de la surveillance active en France

En 2012, un seul cas d'encéphalopathie spongiforme bovine (ESB) atypique a été détecté parmi les 313 216 animaux prélevés à l'équarrissage, portant à 28 le nombre total de cas d'ESB atypiques identifiés en France depuis 1990 (14 cas d'ESB atypique de type L, 14 cas d'ESB atypique de type H). Les 937 725 animaux testés dans le cadre du programme en abattoir étaient tous négatifs pour la recherche d'ESB. Pour la première fois depuis le début de la surveillance active en 2000, aucun cas d'ESB classique n'a été identifié au cours de l'année.

Mots clés
ESB, épidémiosurveillance, police sanitaire, bovins, France

An overview of the BSE surveillance system and control measures is presented in Box 1 below

Change in the number of cases

Testing of 937,725 samples taken at slaughterhouses and 313,216 at rendering plants in 2012 revealed five non-negative results including a single positive confirmed case of atypical L-BSE (Box 2). This brings to 14 the number of L-BSE cases identified between 1 January 1990 and 31 December 2012.

For the second consecutive year, no cases of BSE were detected at the slaughterhouse, and for the fifth consecutive year, no suspected clinical cases were reported (Sala *et al.*, 2012).

The total number of classical BSE cases identified since implementation of surveillance in 1990 has thus remained unchanged since 2011 at 1003 reports, while the number of atypical BSE cases is 28, i.e. 14 H-type BSE cases, and 14 L-type BSE cases.

In 2012, control measures involved slaughter procedures for 17 animals, including 16 extended cohort animals[1] and one descendant.

Cost

The tests carried out at the slaughterhouse are funded by the cattle production sector, for the additional amount exceeding European co-funding of €8 per test. The French government covers 100% of the costs related to rendering plant testing, with reimbursement of €8 per test from European co-funding.

For 2012, the French government made 19.7 million Euros available for BSE surveillance, with about 6.5 million for the slaughterhouse programme (€8 per test on slaughtered animals) and 13.1 million Euros for the rendering plant programme (€51 on average per test at the rendering plant). Increasing the screening age for animals at the slaughterhouse has resulted in a saving of about 13 million Euros since January 2009.

The cost of control measures in 2012 was €27,664.

European Union co-funding scheduled to be allocated to France in 2012 for the transmissible spongiform encephalopathy (TSE) control programme, that includes BSE and scrapie, amounts to 10 million Euros.

Discussion

Since 1 January 2002, the date at which programmed surveillance became 100% operational for the target animal categories, the number of tested animals as part of BSE surveillance on rendering has remained relatively stable, between 250,000 and 300,000 animals per year. In parallel, the gradual increase in the minimum age of sampling for animals at the slaughterhouse has led to a two-thirds reduction in the number of tests carried out at the slaughterhouse between 2002 and 2012 (Figure 1).

The prevalence of classical BSE was zero in 2012, confirming control of this anazooty. For the first time since BSE programmed surveillance was set up, no case of classical BSE was identified in France (Figure 1).

Concerning atypical forms of BSE, the L-type BSE case identified on rendering remains within the epidemiological profile of L-BSE cases diagnosed to date, i.e. animals aged over eight years and mainly reared for meat production (Sala *et al,* 2012).

For the second consecutive year, no cases of BSE were detected at the slaughterhouse. This finding, along with risk analyses carried out at the European level (EFSA, 2012), should make it possible to further reduce surveillance at the slaughterhouse.

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(1) Animals born up to 12 months before or after birth of the case animal and congeners reared with the case animal during the first year of life, while the case animal was under 12 or 24 months

Box 1. Surveillance and control measures for BSE in cattle

Objectifs

- To determine the prevalence of BSE in cattle.
- To detect, when applicable, any re-emergence of the BSE epizootic.

Monitored population

- **Programmed surveillance:** cattle aged over 24 months (living or rendered), cattle over 48 months between 1 January 2009 and 30 June 2011, and cattle over 72 months from 1 July 2011, intended for human consumption.
- **Outbreak surveillance:** the entire cattle population.

Surveillance procedures

- **Outbreak surveillance**

Carried out through the national BSE epidemiological surveillance network. Based on clinical surveillance of animals on the farm and at the slaughterhouse (ante-mortem inspection). Any suspected case detected on the farm by the attending veterinarian is confirmed or ruled out by the veterinarian coordinating the departmental network.

- **Programmed surveillance**

Since 2001, two surveillance programmes have been in place:

> *Slaughterhouse programme: systematic screening of all cattle over 72 months (48 months between 1 January 2009 and 30 June 2011, 30 months before January 2009, and 24 months between July 2001 and July 2004), intended for human consumption.*

> *Rendering programme: screening of all cattle over 24 months that died on the farm or were euthanised for disease or accidents.*

Definition of suspected animals and cases

- Any animal with the following characteristics is considered suspect for BSE:

> *Living, slaughtered or dead animal presenting or having presented progressive neurological and/or behavioural disorders and/or deterioration of the general state that cannot be attributed to a disease other than BSE;*

> *Animal with a non-negative or suspect result on a rapid specific BSE test (ELISA, western blot or immunochromatographic methods).*

- Any suspect animal with a positive result for a confirmation test recognised by the Ministry of Agriculture (immunohistochemistry, western blot) is considered to be infected with BSE.

Animal health rules

In suspected cases of BSE, the farms that held the animal during its first two years of life, and possibly the site currently holding the suspect animal, are placed under Prefectural monitoring order (APMS). If the case involves clinical suspicion, the suspect animal is then euthanised and diagnostic samples are taken.

If the case is confirmed, the farm or farms concerned are placed under Prefectural declaration of infection (APDI), the cattle belonging to the same birth cohort as the confirmed case are slaughtered (animals born up to 12 months before or after birth of the case animal) along with cattle reared with the case animal during the first year of their life, while the case animal was under 12 or 24 months at the site of birth or of rearing, respectively. On these farms, if the affected BSE animal is female, calves born to the case animal in the two years preceding death, or showing clinical signs, or born during the clinical phase, are slaughtered.

Regulations

– Regulation (EC) No 999/2001 of the European Parliament and of the Council of 22 May 2001 laying down rules for the prevention, control and eradication of certain transmissible spongiform encephalopathies.

– Ministerial Order of 3 December 1990 establishing control measures for bovine spongiform encephalopathy.

Box 2. BSE strains

A single BSE strain was recognised until 2003, when two new strains were identified. The atypical biochemical profile of these new strains compared to the «classical» BSE strain gave rise to the names used for the three known BSE strains:

- C-type BSE (Classical BSE) for the form of the disease causing the anazooty related to contamination of animals via feed,
- Atypical L-type BSE (L-BSE) for the strain characterised molecularly by a low level of the biglycosylated proteinase K-resistant prion protein (PrPres) form and an apparent molecular mass of PrPres that is slightly lower than in C-type BSE on western blot,
- Atypical H-type BSE (H-BSE) characterised by an apparent molecular mass of PrPres that is higher than in C-type BSE on western blot.

The two atypical BSE strains also differ from the classic strain in terms of epidemiological characteristics (Sala *et al.*, 2012):

- Low prevalence of less than 1 case/million that is relatively constant over time and consistent geographically with its presence in countries apparently free from C-type BSE, suggesting that these forms are not contagious and not caused by simultaneous exposure of groups of animals, unlike in the case of C-type BSE,
- A mean age at diagnosis of 12.5 years on average, which is higher than that of animals with C-type BSE: 7 years on average for the cases detected in France.

Figure 1. Results of BSE surveillance since launch of monitoring in France: number of tests performed per surveillance programme, and number of detected cases by BSE type and by surveillance programme

Surveillance of spongiform encephalopathies in small ruminants in 2012: prevalence of classical and atypical scrapie remains very low

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Abstract

In 2012, 66,379 goats and 52,911 sheep were screened at the slaughterhouse and during rendering for transmissible spongiform encephalopathies: 22 cases of atypical scrapie were detected in sheep versus five in goats. Only two cases of classical scrapie in sheep and two cases in goats were detected. An overview of surveillance since 2002 shows a drop in prevalence of classical scrapie in sheep and goats, as well as an unexpected drop in prevalence of atypical scrapie in sheep.

Keywords

TSE, small ruminants, programmed surveillance, outbreak surveillance, prevalence

Résumé

Surveillance des encéphalopathies spongiformes des petits ruminants en 2012 : la prévalence des tremblantes classique et atypique se maintient à un niveau très faible

En 2012, 66 379 caprins et 52 911 ovins ont été testés à l'abattoir et à l'équarrissage pour la recherche d'encéphalopathies spongiformes transmissibles : 22 cas de tremblante atypique ovine ont été détectés contre cinq chez les caprins. Seuls deux cas de tremblante classique ovine et deux cas de tremblante classique caprine ont été détectés. Le bilan de cette surveillance depuis 2002 montre une diminution de la prévalence de la tremblante classique chez les ovins et les caprins, et de manière inattendue une diminution de la prévalence de la tremblante atypique chez les ovins.

Mots clés

EST, petits ruminants, surveillance active, surveillance événementielle, prévalence

An overview of the surveillance system, its objectives and implementation methods is presented in the Box. No changes were made between 2011 and 2012.

Results

Number of tests carried out

A total of 119,290 samples were taken in 2012. The objectives of the surveillance programme were achieved for sheep at the slaughterhouse (11,923 samples) and at the rendering plant (40,988 samples), and for goats at the slaughterhouse (55,909 samples). Concerning goats at the rendering plant, the programme provides for systematic sampling, but data are not available to assess whether all animals were screened (10,470 samples taken).

Sampling rates

Estimates indicate that nearly 35% of farms had at least one tested animal in 2012. Like in previous years, it is apparent that inconsistency in sampling rates tends to correspond to zones linked to different production areas (Figures 1 and 2).

Changes in prevalence for classical and atypical scrapie

The prevalence of atypical and classical scrapie (Figure 3) is calculated as the number of atypical or classical cases relative to the number of tests performed (like in 2011, all the tests used in 2012 were able to detect atypical scrapie).

Prevalence rate for classical scrapie in 2002 at the rendering plant was deliberately omitted. Indeed, a previous analysis carried out during the 2002 programme (Morignat *et al.*, 2006), highlighted classification errors for animals according to the programme, with some samples from control measures on affected farms being classified in the rendering plant programme.

In 2012, a single case of classical scrapie was found in sheep at the slaughterhouse, like at the rendering plant. The prevalence of classical scrapie in sheep has consistently decreased year on year since 2002, both at the slaughterhouse (Chi-square trend analysis $p = 6.5*10^{-11}$) and at the rendering plant (Chi-square trend analysis $p < 2.2*10^{-16}$).

Like for classical scrapie, the prevalence of atypical ovine scrapie has decreased significantly since 2002, both at the slaughterhouse with four cases detected in 2012 (Chi-square trend analysis $p = 4.8*10^{-4}$) and at the rendering plant with 18 cases detected in 2012, Chi-square trend analysis $p = 3.9*10^{-4}$).

Since 2008, no cases of classical scrapie have been found in goats at the slaughterhouse, and only two cases of classical scrapie have been found at the rendering plant. The prevalence of classical scrapie in goats remains very low, whether at the slaughterhouse or the rendering plant.

Similarly, the prevalence of atypical scrapie in goats remains very low at the slaughterhouse with one case detected in 2012, and was stable at the rendering plant with four cases detected this year.

Genotyping in sheep

In sheep, there are differences in genetic susceptibility to atypical and to classical scrapie. Homozygous ARR sheep are almost completely resistant to classical scrapie, and alleles VRQ, ARQ and AHQ have decreasing susceptibilities. Susceptibility to atypical scrapie is related to the presence of the AHQ and $AF_{141}RQ$ alleles.

In 2012, of the 767 genotype tests performed in negative sheep at the slaughterhouse or rendering plant, 743 provided interpretable results. For all breeds combined, the following frequencies were found in the tested animals: ARR allele 57%, ARQ allele 35%, VRQ allele 5%, and AHQ allele 2%.

Since 2002, a slight increase in the frequency of the ARR allele has been found in all breeds, with a corresponding decrease in the ARQ allele. The proportions of sheep harbouring the VRQ and AHQ alleles appear to be relatively stable (Cazeau *et al.*, 2005).

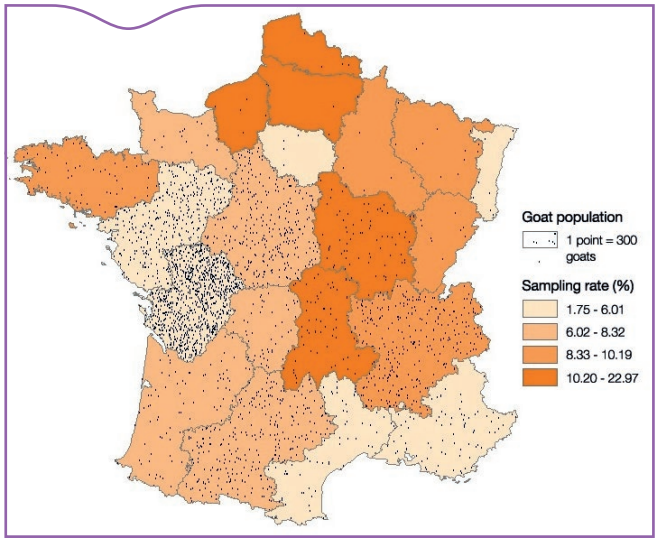

Figure 1. Sampling rate for goats in 2012 (slaughterhouse and rendering plant). To indicate the population, one point represents 300 goats and is placed randomly within the region. The denominator is the goat population by region for 2010 (source: Agreste (Office for statistics, evaluation and forecasting in agriculture))

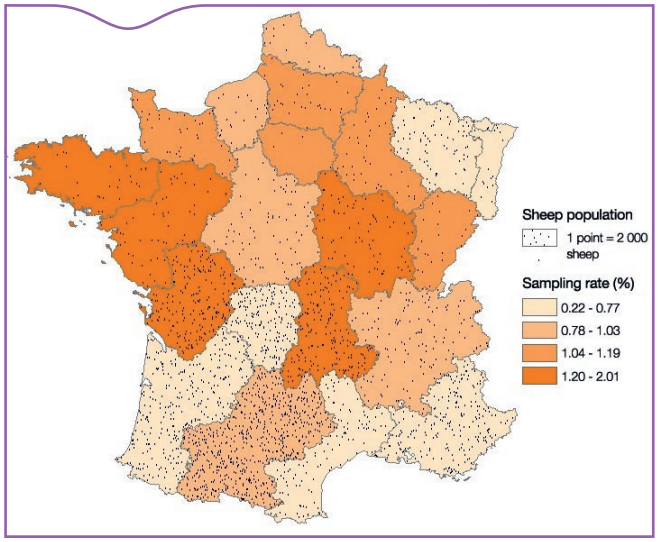

Figure 2. Sampling rate for sheep in 2012 (slaughterhouse and rendering plant). To indicate the population, one point represents 2000 sheep and is placed randomly within the region. The denominator is the sheep population by region for 2010 (source: Agreste (Office for statistics, evaluation and forecasting in agriculture))

Cost

Surveillance of small ruminants at the slaughterhouse had a cost of about 626,000,00 euros in 2012. Surveillance at the rendering plant, which includes brain sectioning and sampling, had a cost of 4.4 million Euros. The total cost of surveillance for France was therefore 5 million Euros. About 35% of this expenditure should be covered by the European Union (subject to a favourable decision being made), as part of co-funding for TSE surveillance and eradication programmes, with the remaining cost to be paid by France, i.e. 3.2 million Euros.

In addition, the cost of control measures is estimated to be 103,000 euros and genotype testing (including control measures) to be 995,350 Euros. After allocation of European funds (21% of expenditure), the cost for the French state will be 960,000 Euros.

Discussion

Concerning classical scrapie, a significant decrease in prevalence has been observed since 2002, both in sheep and goats. This lower prevalence is possibly multifactorial and could be explained by the control measures implemented for the disease in affected herds, and by selection of genetically resistant animals. Moreover, a case-control epidemiological survey found that certain concentrated feeds may have been a source of classical scrapie contamination (Philippe *et al.*, 2005). Measures for withdrawal of specified risk materials (SRMs) and then banning of meat-and-bone meal appear to have contributed to reduced exposure to classical scrapie *via* feed.

For the atypical forms, no change was observed for prevalence in goats, while the rate has decreased in sheep, although this lower rate does not appear to be consistent with a sporadic disease, with no risk factor identified, as it has been proposed (Fediaevsky *et al.*, 2010).

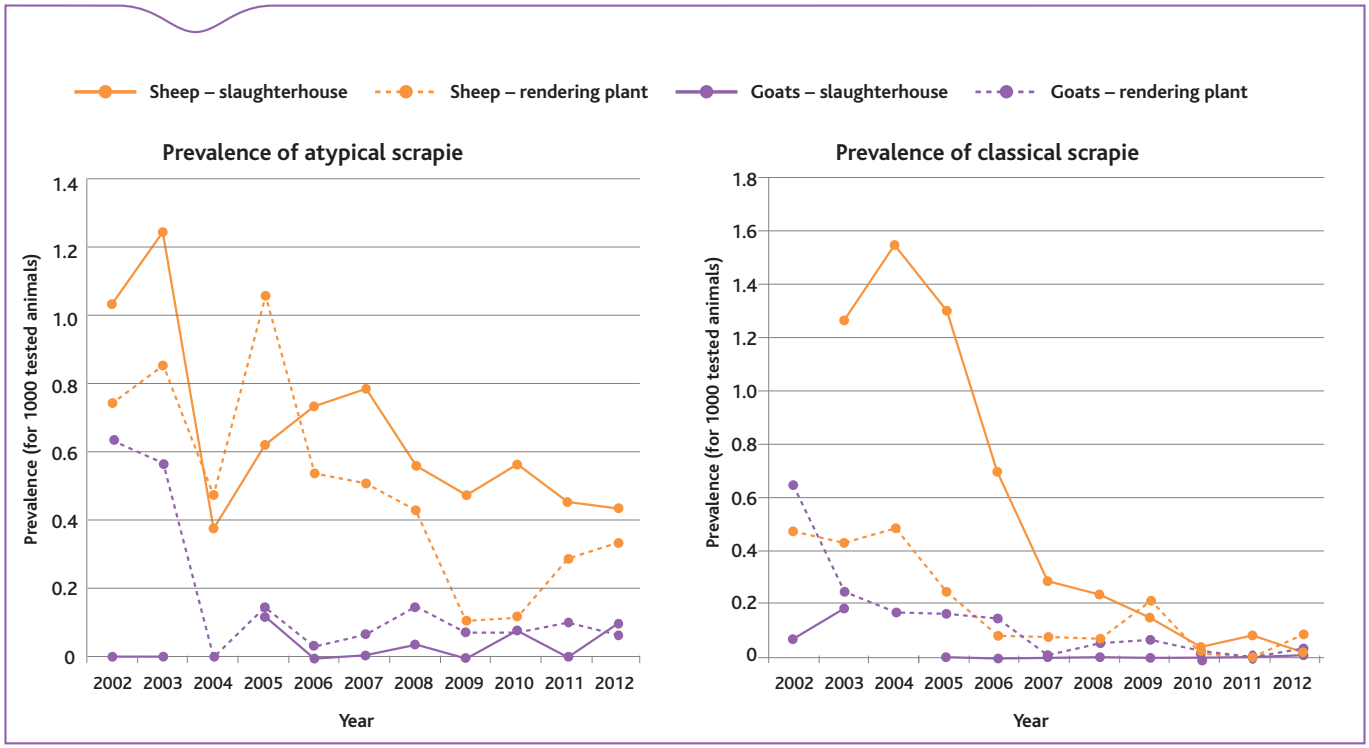

Figure 3. Changes in prevalence of classical and atypical scrapie in sheep and goats at the slaughterhouse and rendering plant

Like in the case of BSE surveillance, implementation of active national surveillance programmes has had an impact on the functioning of outbreak surveillance for scrapie. The number of suspected and clinical cases has decreased considerably each year since 2002, and no suspected cases were reported in 2012.

Overall, both forms of scrapie are rare and remain at very low levels. No suspected cases of BSE were detected in small ruminants in 2012.

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Box. Surveillance and control measures for scrapie

Objectives
- To determine estimated prevalence of scrapie in small ruminants.
- To detect, as the case may be, any presence of BSE in small ruminants.

Monitored population
Active surveillance: live sheep and goats, animals at rendering plants or intended for human consumption in mainland France.

Outbreak surveillance: the entire sheep and goat population.

Surveillance methods
- Outbreak surveillance

On the basis of clinical signs on production sites or on ante-mortem inspection at the slaughterhouse.

If a clinical case is suspected on a production site, the livestock producer must inform the farm's veterinarian and the suspected case must be reported to the veterinary authorities.

- Programmed surveillance

Annual surveillance since 2002 at the European level.

Random sampling of adult sheep and goats slaughtered for human consumption (3% and 9.1% of slaughtered animals, respectively) and of rendered adult sheep (40,000). Systematic screening of rendered adult goats.

Animal health rules
If a case is reported on clinical suspicion or if a non-negative result is obtained on a rapid test, the farms where the suspect animal was born, lived for more than nine months during its first year, or where it gave birth, are considered at risk. These herds are placed under Prefectural monitoring order (APMS), which specifically prohibits sale of small ruminants, as well as their milk and any derived dairy products.

If the case is confirmed, the herds are subject to health control measures that vary depending on the TSE strain identified:

> *BSE: total depopulation of the herd of birth and any herds in which the case animal may have given birth;*
> *Classical ovine scrapie: elimination of genetically susceptible animals from the herd of birth. Animals can be sold only to the slaughterhouse and the milk of genetically susceptible animals must be destroyed. These measures are replaced by reinforced follow-up for three years if the affected animal transited through several farms;*
> *Classical caprine scrapie: total depopulation of the herd of birth;*
> *Atypical scrapie: very close monitoring of the risk herd for two years. The animals can be sold only to the slaughterhouse or to another establishment with the same status.*

Regulations

– Guidance note DGAL/SDSPA/N2012-8042 of 22 February 2012: Surveillance of transmissible spongiform encephalopathies (TSE) in small ruminants
– Regulation (EC) No 999/2001 of the European Parliament and of the Council of 22 May 2001 laying down rules for the prevention, control and eradication of certain transmissible spongiform encephalopathies

IBR in France in 2012: certification procedure is in progress

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Abstract

The 2011/2012 surveillance campaign for infectious bovine rhinotracheitis (IBR) has ended with a national prevalence rate of 10.7%, and an incidence rate of 1.95%, an increase compared to previous years. The situation is highly variable, depending on the *département*, the production type, regional specificities with regard to farming practices, and the historical interest in IBR in certain regions. The national certification rate is 62% up by two points compared to the last campaign. Since IBR leads to requests for additional guarantees in the context of trade within the European Union, measures have been taken to obtain recognition for the French IBR certification programme at EU level.

Keywords

Infectious bovine rhinotracheitis, IBR, cattle, epidemiological surveillance

Résumé

L'IBR en France en 2012 : une démarche de qualification toujours en progression

La campagne 2011/2012 de surveillance de la rhinotrachéite infectieuse bovine (IBR) se termine sur un taux de prévalence national de 10,7 %, et un taux d'incidence de 1,95 %, en augmentation par rapport aux années précédentes. Les situations sont très variables selon les départements, les types de production, les particularités régionales sur le plan des pratiques d'élevage et selon l'intérêt historique porté à l'IBR dans certaines régions. Le taux national de qualification s'élève à 62 %, en hausse de deux points par rapport à la campagne précédente. L'IBR donnant lieu à des demandes de garanties additionnelles dans le cadre des échanges au sein de la Communauté européenne, des démarches ont été entreprises pour faire reconnaître au niveau européen le programme français de certification IBR.

Mots clés

Rhinotrachéite infectieuse bovine, IBR, bovins, épidémiosurveillance

Infectious bovine rhinotracheitis (IBR), initially recognised in its genital form, appeared as a respiratory condition in the United States in the 1950s and then in Europe in the 1960s. Today, the disease is considered important primarily for its repercussions on trade. IBR is included in the Terrestrial Animal Health Code of the World Organisation for Animal Health (OIE) and is often associated with trade requirements that are common obstacles to animal trade. At present, the clinical incidence of IBR appears to be low, irrespective of the prevalence of infection in a given *département*. However, although clinical episodes are rare, they can have significant economic consequences for the livestock farm.

IBR is caused by bovine herpesvirus 1 (BoHV-1). One of the specific characteristics of the virus is that it can persist in latent form in asymptomatic carrier animals and, during a period of stress or drug treatment (particularly with corticosteroids), can be reactivated leading to recurrence of viral shedding and contamination of other animals in the herd. Reactivation followed by renewed shedding in apparently healthy cows represents a major risk of introduction or spread of infection within the herd. Vaccination of seropositive cattle is one way of limiting recurrence of shedding.

There are currently two surveillance and control schemes, one voluntary and the other mandatory (Bronner *et al.*, 2010) (Box):

- The voluntary scheme is underpinned by a national certification protocol, managed by the French Certification Association for Animal Health (Acersa). Implementation of the scheme relies on a network of local certification units, called STCs. These bodies can grant two different certifications for herds: IBR-free (A) and IBR-controlled (B). Certification as IBR-controlled means that there is no viral circulation in the herd, but it may have seropositive animals aged more than 48 months at the time of receiving this certification. The health requirements underlying herd certification are stipulated in a statement of requirements approved by the Ministry of Agriculture (National Statement of Requirements CC IBR 01, 2010);
- The mandatory scheme is based on the Ministerial Order of 27 November 2006 and relies on screening of tank milk in dairy farms (every 6 months) or on annual blood sampling of cattle over 24 months, on screening of cattle following transfer, and on vaccination of seropositive animals.

This article presents the results obtained through these certification and control schemes for the 2011-2012 programme, for the period from 1 June 2011 to 31 May 2012. The results given below are based on data collected specifically from local farmers' animal health protection organisations (GDSs) through an annual update questionnaire. This article also discusses changes in control measures that have occurred over the last few months.

Box. Surveillance and control measures for infectious bovine rhinotracheitis (IBR)

Objectives
- To determine the estimated prevalence of IBR in cattle.
- To contribute to certification of the health status of herds in France.

Monitored population
Domestic cattle across mainland France.

Surveillance procedures
- Mandatory surveillance
  - > *Serological screening on transfer for all animals, irrespective of their age, with ad hoc exemptions granted in some cases;*
  - > *Serological screening of cattle herds: every 6 months, of tank milk on dairy farms, and annually, through blood sampling of cattle over 24 months of age on beef cattle farms.*
- Voluntary certification of herds

Since 1996, through officially recognised certification of herds, cattle buyers can be given health guarantees for IBR. The certification scheme is managed by Acersa, with implementation in the field by local certification units (STCs). The health requirements underlying herd certification are stipulated in a statement of requirements approved by the Ministry of Agriculture.

Animal health rules
Any animal that is not seronegative must be vaccinated within two months of notification of results, unless the animal is slaughtered.

Regulations

Ministerial Order of 27 November 2006 establishing collective control measures for infectious bovine rhinotracheitis.

Results from the mandatory scheme

Prevalence and incidence rates

As of 31 May 2012, mandatory IBR screening of herds revealed that 10.7% of tested herds on average had at least one seropositive animal (data from 84 *départements*). This figure is higher than rates found in previous years (on 31 May 2010 and 2011, the prevalence was 8.1% and 8.9%, respectively).

The prevalence varied between 0.10% and 84.4% depending on the *département*. Lowest prevalence rates were found in *départements* with primarily dairy production (Figure 1). This variability can be partially explained by the initial situation and previous local control plans. It may also be related to certain regional practices, such as summer grazing or particularly dense trade networks, more common in beef farm areas, that complicate control efforts (Mémeteau *et al.*, 2011). Moreover, certain beef farming regions have farm layouts that are highly fragmented, increasing the risk of transmission due to greater contact between herds grazing in the same area. In contrast, dairy farming practices are often more conducive to control, specifically with a higher cull rate, making it possible to eliminate positive animals more quickly. Grazing areas are also less spread out, implying less contact with other herds and thereby a lower risk of contamination. In addition, specific local conditions may be found, for example in northern France along the Belgian border since Belgium had high prevalence rates for many years (the herd prevalence rate was 67% in 1998) (Boelaert *et al.*, 2000). The control programme implemented in Belgium in 2006 (Royal Order of 22 November 2006), which became mandatory in 2012, is likely to improve the situation.

The IBR incidence rate for the 2011-2012 programme was 1.95% (data for 84 *départements*) with values ranging from 0% to 13.4% depending on the *département* (Figure 2). Like for prevalence rates, an increase was observed compared to previous years (on 31 May 2010 and 2011, the incidences rates were 0.9% and 0.6% respectively).

This observed increase in incidence rates was also found for certified livestock farms (see below "Incidence of IBR in certified herds").

For the 2011-2012 programme, the effective national rate of control screening reached 90.6% (data for 81 *départements*). As a comparison, this rate was 97% for the 2010/2011 programme and 89% for 2009/2010.

Results of testing on transfer of animals to a herd

The second measure provided for in the Ministerial Order is IBR screening on arrival of any animal in a herd, regardless of its age. However, this requirement may not apply to some animals: transfer to an exempt fattening farm (if the farm has entirely closed facilities), cattle that have undergone vaccination certified by a veterinarian, and cattle from herds certified IBR-free if transported in controlled conditions. Data collected for 81 *départements* indicate a proportion of 1.4% seropositive cattle on purchase for all transferred animals, whether certified or not, excluding exempt establishments.

Results from the voluntary scheme

Herd certification rates

As of 31 May 2012, 62.0% of herds in mainland France (excluding exempt farms) had an IBR-free or an IBR-controlled certification (data for 86 *départements*). Here again, the picture is not consistent country-wide with herd certification rates varying from less than 0.4% to almost 98.3% (Figure 3). This difference can be explained by the same factors underlying the variable prevalence observed for mandatory screening. The certification rate also depends on past interest in this disease, with some *départements* being quicker than others to implement control measures and certification systems, before mandatory controls were established.

The number of certified herds has increased steadily since the certification system was introduced as part of Acersa, rapidly between 2001 and 2007, then more slowly in recent years (Figure 4).

Overall, as of 31 May 2012, 121,703 herds were certified. IBR-A certified herds, corresponding to the IBR-free status, were the most common, accounting for 99.3% of certified herds (120,839 herds), *versus* 0.7% with IBR-B certification for IBR-controlled herds (864 herds). This low percentage can be explained by the fact that IBR-controlled status is often no more than a transitional step for a herd in the process of eradicating the disease.

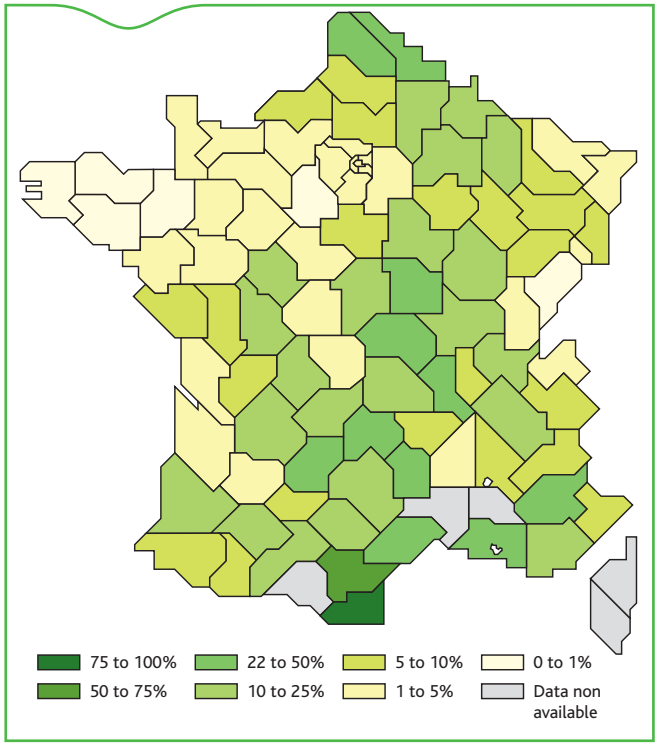

Figure 1. **Prevalence rate (herds) by *département* as of 31 May 2012 (GDS France data)**

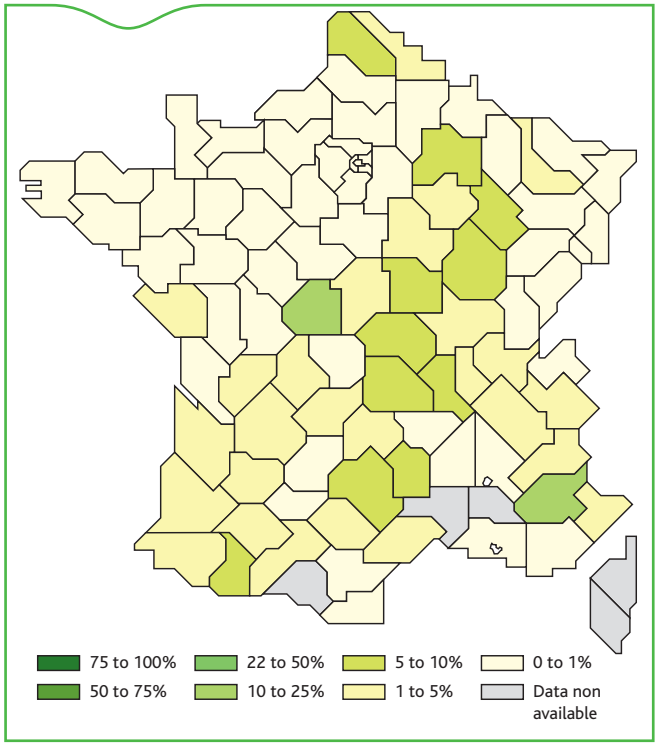

Figure 2. **Incidence rate (herds) by *département* as of 31 May 2012 (GDS France data)**

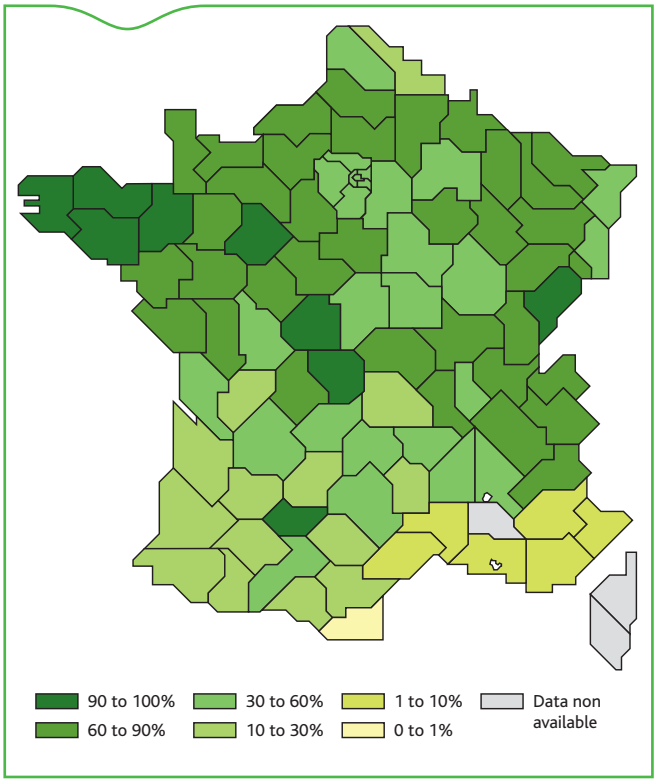

Figure 3. Proportion of certified herds by *département* as of 31 May 2012 (Acersa data)

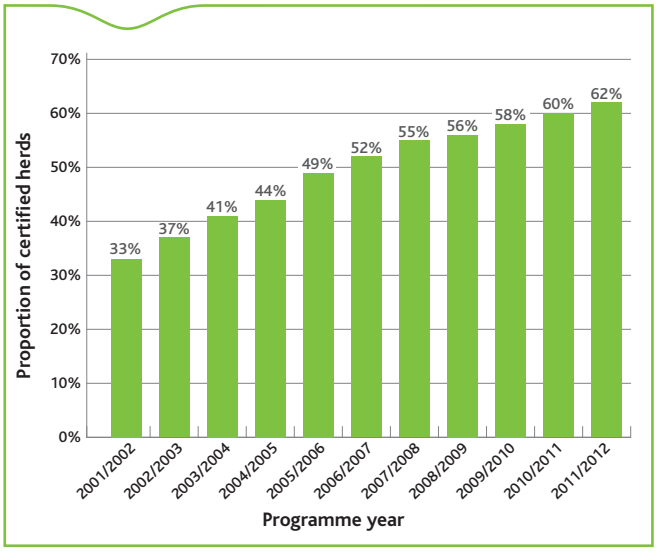

Figure 4. Change in the proportion of IBR certified herds since 2001

Incidence of IBR in certified herds

IBR-A certified herds

During the 2011/2012 programme, positive animals were found in 1,026 herds that had been certified with an IBR-A status on 1 June 2011, representing 0.8% of herds with this certification at the start of the programme.

In 82% of cases, the herds had one or two positive animals (Figure 5). This proportion is similar to that observed for the previous programme (84% of herds with one or two positive animals in the 2010/2011 programme).

IBR-B certified herds

New positive cases were found during the 2011/2012 programme in 55 herds that had been certified with an IBR-B status on 1 June 2011, representing 3.6% of herds with this certification at the start of the programme.

80% of these herds also had one or two new positive cases (Figure 6).

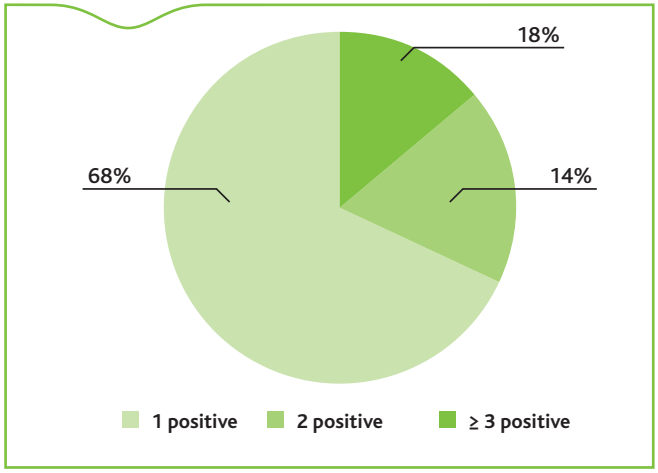

Figure 5. Distribution of IBR-A certified herds in which positive animals were detected during the 2011/2012 programme, based on the number of positive cases

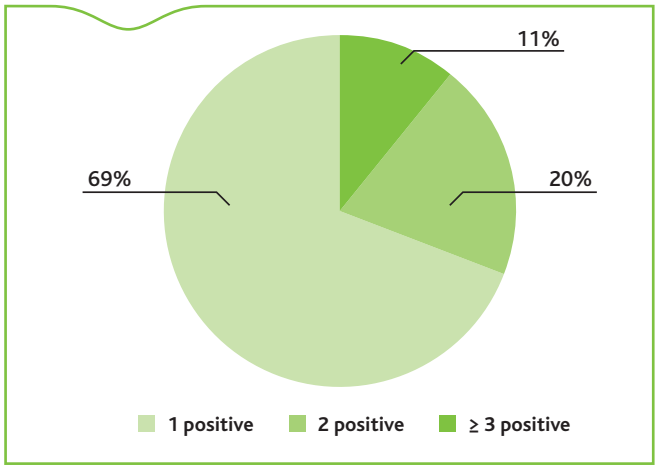

Figure 6. Distribution of IBR-B certified herds in which new positive animals were detected during the 2011/2012 programme, based on the number of new positive cases

These findings indicate that the distribution in IBR-B herds, based on new positive cases, is very similar to that in IBR-A certified herds. It could therefore be assumed that the presence of infected vaccinated animals within a herd (case for IBR-B herds) is not a risk factor for the occurrence of new positive cases, reflecting the efficacy of vaccination in controlling viral circulation.

Discussion

These results, along with those observed through mandatory control screening, that show an increase in incidence and prevalence by herd, were obtained following implementation of new ELISA serological testing kits with higher detectability and sensitivity during the 2010/2011 programme. The testing procedures for these kits were also adjusted for 2011/2012: change in the order of testing using various kit types for individual analysis on pooled serum with a non-negative result. This change came about because acceleration of the IBR control programme increased the need for higher sensitivity analytical kits to detect infected animals. As a result, it is possible that the programme identified infected animals that would not have been found with earlier-generation kits, animals that would have been eliminated in this way. However, the large number of detected animals could be indicative of a possible slight shift in method specificity that may result in false detection of healthy animals.

Given this situation, an Acersa working group including scientists, risk managers and laboratories, was established in order to put forward a proposal on changes to management rules. The conclusions of their assessment were implemented starting from the 2011/2012 programme and continued in 2012/2013. They include more suitable

management rules for isolated positive cases found on certified farms and establishment of a database enabling STCs to record seropositive cases on certified farms. Follow-up of these farms, and specifically information on changes in their status, will in time provide epidemiological data that will help to optimise control rules, both from the point of view of livestock farmers and risk managers, by maintaining a suitable safety level given the importance of the disease.

Conclusion and future trends

A nation-wide analysis of results shows that there are differences between *départements*, and more generally a slight increase in prevalence and incidence rates. This change raises questions since many positive results concern isolated single reactors, and this has led to a number of assessments of control strategies, analytical testing and epidemiological aspects.

Although current control measures are able to reduce the impact of infection for livestock farmers who have isolated positive animals, the situation must be assessed more closely to develop a better understanding of some of the results observed (positive cases detected in certified herds, inconsistent results between methods, etc.). Recording of specific cases by STCs and regular contact with local risk managers enable Acersa to document the various situations. The IBR NRL for its part, is working on the development of a confirmation tool, and in collaboration with Acersa on the implementation of a collection protocol for analytical and epidemiological data concerning specific cases. The aim initially is to collect comparable usable data on these cases, and subsequently to propose suitable solutions.

The picture across the European continent is also variable since Denmark, Austria, Finland, Sweden, Bolzano province in Italy and certain regions in Germany (Upper Palatinate and Upper Franconia Lands of Bavaria) are disease-free, while the Netherlands and Belgium have high prevalence rates. IBR control can result in additional guarantees in the area of trade within the European Union since certain countries or regions have obtained recognition of their control system or the status of disease-free area. This enables these Member-States to insist on additional guarantees when cattle are transferred to farms within their national territory, guarantees that French livestock farmers are subject to. France has begun the process of applying for recognition of its national IBR control scheme at the European level, in particular its IBR-free certification, which would help lift certain trade restrictions.

The growing demand for guaranteed cattle provides excellent motivation for farmers, who could benefit from certification but who have not yet made an application, to do so without delay.

Acknowledgements

We would like to thank all accredited laboratories for diagnosis of IBR in serum or milk, the GDS units that implement IBR controls, and the coordinators of local certification units, who provided us with the data that made it possible to draft this article.

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Bovine hypodermosis in France in 2012: second consecutive year with no outbreaks

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Abstract

During the 2011-2012 campaign, 9,517 herds underwent random and targeted screening checks for bovine hypodermosis (serological analyses and sight checks); 67% of the herds checked were chosen by random draw, while 33% underwent targeted screening. For the second year in a row, no outbreaks were detected. The epidemiological situation in France is therefore very good. The difficult problem of border areas still needs to be dealt with, since the warble fly (*Hypoderma*) can be found in these areas, and outbreaks are possible due to an absence of organised control plans in bordering countries, an absence of natural barriers, and the proximity of French and foreign herds on summer pasture lands. Because of this, reinforced monitoring of these risk areas, disease introduction surveillance, and targeted screening are being maintained in order to avoid undermining the efforts that have been made over the last few years.

Keywords

Bovine hypodermosis, warble fly, cattle, epidemiological surveillance

Résumé

Hypodermose bovine en France en 2012 : deuxième année consécutive sans foyer détecté

Durant la campagne 2011-2012, les dispositifs de surveillance aléatoire et orientée de l'hypodermose bovine (analyses sérologiques et contrôles visuels) ont porté sur 9 513 cheptels : 67 % des cheptels surveillés ont été tirés au sort et 33 % ont fait l'objet de contrôles orientés. Aucun foyer n'a été mis en évidence, pour la seconde année consécutive. La situation épidémiologique de la France est donc très favorable. Il reste cependant encore aujourd'hui à gérer le problème délicat des zones frontalières, où la présence du varron et donc de foyers est toujours possible du fait de l'absence de plans de lutte organisée dans les pays limitrophes, de l'absence de barrières naturelles, et de la proximité entre troupeaux français et étrangers en zone d'estive. Dans ce contexte, la surveillance renforcée des zones à risque, la surveillance des introductions et les contrôles orientés sont maintenus pour ne pas compromettre les efforts entrepris depuis plusieurs années.

Mots clés

Hypodermose bovine, varron, bovins, épidémiosurveillance

Hypodermosis (warble) is an internal myiasis in cattle characterised by infestation of subcutaneous connective tissue in the dorsolumbar region by larvae of flies in the *Hypoderma* genus, following a period of migration and larval transformation. The larvae develop in bovine tissue over the winter and emerge in the spring after forming a nodule on the animal's back and perforating the skin.

In the past, this disease had substantial economic consequences: reduced milk production, slowed growth of young animals, immunosuppression caused by larvae, and damage to the hide when the larvae exit through the skin in the spring. For these reasons, farmers came together at the end of the 1980s to implement an organised control plan, region by region. Each regional control scheme had two components: a systematic treatment phase at the beginning of the plan, followed by a serological testing phase for several years. Serological testing became mandatory for all herds in France in July 1998 and was reinforced by the Ministerial Order of 6 March 2002. A rapid decrease in the country-wide prevalence of hypodermosis was then observed in herds between 1998 and 2001, from 5.7% to 0.4% (Mémeteau *et al.*, 2011). Given the rate of eradication, in February 2006 bovine hypodermosis in its clinical form became a notifiable disease with compulsory control measures (Decree No. 2006-178, 17 February 2006). It is now considered to be a category 2 health hazard (Ministerial Order of 29 July 2013).

There are currently two surveillance schemes, one voluntary and one mandatory (see Box):

- The mandatory scheme is underpinned by the Ministerial Order of 21 January 2009 and relies on:
  - A random surveillance plan conducted annually that aims to determine whether the prevalence of infestation in a zone is below a certain level (5%). Implementation of this scheme is entrusted to the GDSs.

    This surveillance scheme entails serological analysis of pooled sera or bulk milk (sampled between 1 December of the previous year and 31 March of the current year for blood samples, and between 1 January and 31 March of the current year for milk samples). Sampling takes places as part of bovine control screening procedures, in a randomly selected group of herds. Animals in herds found to be positive then undergo a sight check in the spring to confirm or rule out the presence of hypodermosis.

    Serological surveillance can also be supplemented by random sight checks[1]. These inspections take place during the period when the larvae emerge between 1 April and 30 June each year.

    At the end of the random surveillance programme, and on the basis of an annual report forwarded by the national coordinator (GDS), the DGAL determines which zones are hypodermosis-controlled or hypodermosis-free. An area is considered to be a hypodermosis-controlled zone when the rate of infestation of herds, demonstrated by the random scheme through serology and sight checks, has been less than 5% for two consecutive years. Hypodermosis-free zones have had an infestation rate, demonstrated by random serological testing, of less than 1% for two consecutive years.
  - A targeted screening programme carried out to detect outbreaks of hypodermosis. This programme increases the probability that infested herds will be detected, but also aims to raise awareness among breeders for whom the risk of infestation is related to farming methods. Targeted screening focuses on potentially at-risk herds, specifically when there is an epidemiological link to an infested herd, when they are located in a zone where infestation may recur (particularly border areas, i.e. any municipality located less than 5 km from the border), and based on farming practices (trade, summer grazing) or non-negative test results obtained through random surveillance.

(1) On 31 March each year, if less than 80% of randomly selected herds have been analysed serologically, herds that have not been analysed are inspected visually to reach at least 80% of herds monitored in the area. Serological testing is given preference because sight checks are far less sensitive.

- The second surveillance scheme involves issuing health status qualification, to complement the mandatory measures. It serves to guarantee the status of the herd of origin when animals are sold. This scheme is coordinated by the French Certification Association for Animal Health (Acersa) and implemented by local qualification units (STCs) authorised to grant the following qualifications to herds within their areas: hypodermosis-controlled herd, or hypodermosis-free herd, depending on the zone's status, and guaranteeing the status of the herd of origin when animals are sold. Livestock farmers can apply for either of the qualifications if their herds are located in controlled or disease-free zones and fulfil the conditions in the national statement of requirements (Acersa Statement of requirements - CC VAR 01), and are reported as being in a zone where there is an accredited STC for issuing hypodermosis qualifications.

This article presents descriptive results for bovine hypodermosis obtained through the random and targeted surveillance schemes for the 2011-2012 programme that took place between 1 July 2011 and 30 June 2012. The results presented below are based on data collected specifically from FRGDS groups (Regional animal health protection groups) and forwarded by GDS units that implement the bovine hypodermosis surveillance plan.

Results

During the 2011-2012 programme, 9,513 herds were tested as part of the random and targeted surveillance schemes for bovine hypodermosis through serological analyses and sight checks. 67% of monitored herds were chosen randomly, 33% were identified for targeted surveillance.

Random surveillance of herds

Evaluation of the rate of herd infestation is based on a random sampling plan involving random computerised selection from among all herds in a region, excluding finishing herds that are exempt and housed entirely in closed facilities.

Given the qualitative approach, the sample size is determined on the basis of a threshold prevalence level (5% for the controlled status) and the number of herds present. As such, for a given number of herds present, a maximum acceptable number of positive herds is defined indicating the infestation rate is effectively below 5%, with a probability of 95%. If this maximum number is exceeded, the zone cannot then be considered controlled. As an example, for a zone with 2,000 herds, if the control plan includes 150 herds, the maximum acceptable number of positive herds is three, and if the plan includes 58 herds, none should be positive.

For the 2011-2012 programme, 7,440 herds were selected randomly, including 7,096 that still had cattle at the time of control. Overall, 6,396 herds were tested randomly (6,288 herds by serological control and 108 by sight checks), yielding an effective national testing rate of 90% on average. This rate of effective testing ranged between 81% and 99% depending on the region. The requirement concerning the level of tests to perform, i.e. more than 80% of the random sample, was therefore fulfilled for all regions. This testing level of below 100% is primarily related to the limited time available for carrying out serological controls. A certain number of randomly selected herds could not be controlled, particularly for blood testing, because control screening took place before 1 December or after 31 March. It is also interesting to note that this level of testing is stable compared to the previous year, 90% for the 2010-2011 programme.

Random serological testing

In total, 6,288 herds underwent serological testing: 3,820 were analysed only through blood sampling, 2,058 only through milk, and 409 *via* both blood and milk samples (mixed herds).

Any non-negative result for pooled blood samples leads to individual testing. A non-negative result for one or more animals entails loss of the negative status of the herd. A positive result for a large quantity of pooled milk (tank milk) leads to a positive status for the herd. If the result is uncertain, a second sample is taken before 31 March in order to determine the status of the herd.

During the 2011-2012 programme, six herds were identified as seropositive through the random surveillance scheme (positive blood samples). These six herds were located in five different regions: Provence-Alpes-Côte d'Azur, Nord-Pas de Calais, Midi-Pyrénées, Auvergne and Alsace. Sight checks for infestation intended to detect any cutaneous lesion suggestive of the presence of at least one hypodermosis larva were carried out on the six herds and were negative. These seropositive herds were therefore not registered as bovine hypodermosis outbreaks, but were considered to be the result of residual antibodies or false-positive test results, given the test specificity of 99.8% according to the supplier's validation dossier (Institut Pourquier, 2001). These herds will be included in targeted serological controls next year.

Random sight checks

6,595 animals in 108 herds were inspected visually. No outbreak of clinical hypodermosis was detected.

Box. Surveillance and control measures for bovine hypodermosis

Objectives

- Mandatory surveillance
  - > *Confirmation of the controlled or disease-free status of the various regions of mainland France (an infestation rate of below 5% or 1%, respectively).*
  - > *Early detection of any outbreak of hypodermosis.*
- Voluntary qualification scheme
  - > *Guarantee the status of the herd of origin for animal sales.*

Monitored population

Domestic cattle across mainland France.

Surveillance procedures

- Outbreak surveillance

Any cutaneous lesion suggestive of bovine hypodermosis must be notified to the Departmental Directorate for Protection of the Population (DDecPP) in the *département* where the suspect animals are located.

- Mandatory programmed surveillance
  - > *Screening of a random sample of herds by serological analysis of pooled sera or bulk milk in a random sample of herds, along with possible random sight checks;*
  - > *Targeted screening of herds or animals considered to be at-risk (epidemiological link with an affected herd, geographic location of the herd in an area at risk of re-infestation, farming practices, non-negative results obtained during serological screening plans).*
- Voluntary scheme

This scheme is managed by Acersa and results in qualification of production sites. It is implemented in the field by local qualification units (STCs) authorised to grant herds within their area the qualification hypodermosis-controlled or hypodermosis-free, guaranteeing the status of the herd of origin when animals are sold. Herds qualify if they are in a controlled or disease-free zone respectively and fulfil the conditions of the national statement of requirements.

Animal health rules

Bovine hypodermosis has been a notifiable disease with compulsory control measures in its clinical form since 2006, and is now a category 2 health hazard.

If a farm is found to have clinical cases of bovine hypodermosis, the clinically affected animal or animals, as well as those suspected of being infested, must be treated.

Regulations

– Ministerial Order of 29 July 2013 concerning the definition of category 1 and category 2 health hazards for animal species.

– Ministerial Order of 21 January 2009 establishing collective prophylaxis and control measures for bovine hypodermosis.

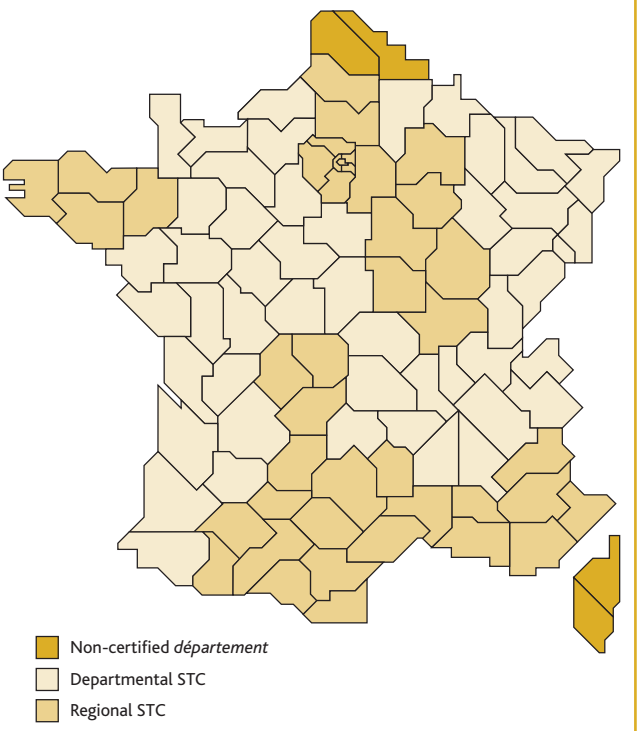

Figure 1. Local accredited qualification structure (Acersa data)

Targeted surveillance of herds

3,117 herds were tested as part of targeted surveillance *via* serological analyses and sight checks.

Targeted serological testing

Serological analyses were carried out in 2,804 herds, with blood samples in 1,585 herds and milk samples in 1,219. These tests detected two seropositive herds in two regions: Nord-Pas de Calais and Midi-Pyrénées. Sight checks performed on both herds were however negative. These seropositive herds were therefore not registered as outbreaks of bovine hypodermosis, but were considered to be the result of residual antibodies or false-positive results.

Targeted sight checks

In all, 313 herds were inspected visually, including 195 (62%) in border areas, and no outbreak was detected.

Control of transfers and treatment measures

Any animal arriving on a farm must undergo hypodermosis treatment, unless exempted (cattle from qualified controlled herds in compliance with the Acersa statement of requirements, transfer to a finishing herd under exemption with cattle only in closed facilities, or cattle born after 31 October and transferred before 1 March of the following year).

In all, 6,147 animals were transferred and treated in mainland France for the 2011-2012 programme, of the 6,627 cattle that should have been treated, providing a treatment rate of 93%. If animals are not treated, this results in the implementation of targeted control of the animal and/or herd of origin.

Tactical treatment in at-risk herds (preventive treatment) was administered for a total of 29,305 cattle in 444 herds. To a very large extent, these treatments were administered in border areas.

Report on implementation of local qualification units

The national control scheme includes 21 regions or zones, including six that have borders with Belgium, Luxembourg, Spain or Italy (14 *départements* in all). Most *départements* and some regions are organised into local qualification units (STCs), accredited by Acersa to manage the control plan for bovine hypodermosis (Figure 1).

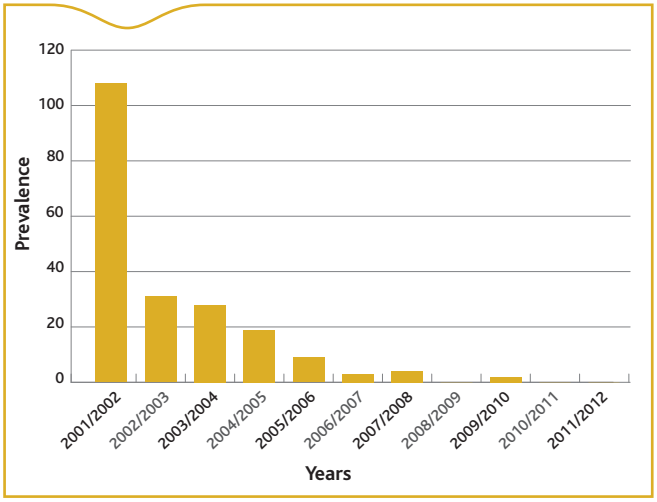

Figure 2. Changes in prevalence of bovine hypodermosis since 2002 in France

At this time, only two *départements* on the mainland have not submitted an application for approval of a local STC. These are Pas de Calais (62) and Nord (59). The epidemiological situation in the border area with Belgium has improved. This should facilitate the management programme and thereby enable these *départements* to initiate applications.

Discussion

Results obtained for the 2011-2012 programme indicate that all regions have an infestation rate below 5% (according to serological testing and/or sight checks) and fulfil requirements concerning the number of tests to perform (more than 80% of the random sample). Therefore, as per the criteria stipulated in the Ministerial Order of 21 January 2009, all the regions of mainland France have a controlled zone status. In addition, the vast majority of *départements* and regions on the mainland have STCs, with regional or departmental organisations accredited by Acersa, and can issue herds with the hypodermosis-controlled qualification.

During the 2011-2012 programme, no outbreaks of bovine hypodermosis were detected, for the second consecutive year. The epidemiological picture is therefore highly favourable. As a reminder, the two most recent outbreaks in France occurred during the 2009-2010 programme. They involved a localised outbreak in the Marne *département* related to transfer of an affected animal from Belgium, and an outbreak near the Italian border in the Hautes-Alpes *département*.

In view of the very low prevalence levels observed over the recent programmes (Figure 2), all or part of the zones could work towards the disease-free qualification. To achieve this, stricter sampling requirements would be needed and could only be accepted if neighbouring regions were grouped together. However, recognition as a disease-free zone is not currently planned since there are no economic benefits compared to the controlled status.

Cost

The 14 *départements* bordering Spain, Italy, Belgium or Luxembourg spent 145,722 Euros on control of hypodermosis (with 60,000 Euros from the State). The measures taken included awareness raising initiatives for breeders, administrative and technical follow-up (targeted screening of herds), and tactical treatment of animals. This expenditure enabled protection of 4,287 herds; these can be considered a buffer zone for the rest of the French territory.

Conclusion

During the 2011-2012 programme, no outbreak of bovine hypodermosis was detected.

The results obtained mean that the disease-controlled status can be maintained for all regions, since hypodermosis can be considered absent at a prevalence rate of 5%.

However, border areas remain susceptible. The presence of warble fly and therefore of outbreaks is still possible in the absence of organised control plans in neighbouring countries, absence of natural barriers, and proximity between French and foreign herds in summer grazing areas (the range of *Hypoderma* flies is about 5 km).

Given this context, surveillance of at-risk zones, tactical treatment, monitoring on transfer, and targeted controls, remain important, with the *départements* most exposed due to their borders playing the role of buffer zone.

Additionally, nation-wide vigilance particularly through control on transfer of animals must be maintained to avoid undermining the efforts made over the last few years.

Acknowledgements

We would like to thank all the accredited laboratories for diagnosis of hypodermosis in serum or milk, all GDS personnel, responsible for implementation of hypodermosis control, and coordinators of local qualification units (STCs), who provided the data essential for drafting this article.

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Bluetongue surveillance in 2012: Recovery of disease-free status in mainland France

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Abstract

Bluetongue disease (BT) first appeared in northern France in late 2006 and then rapidly spread over the entire country, infecting over 30,000 French farms in 2008. Since then, all of mainland France has been viewed as a single protection zone with regard to serotypes 1 and 8, considered as endemic. After several mandatory vaccination campaigns (2008-2010) followed by voluntary campaigns (2010-2012), the number of outbreaks fell drastically. In 2012, thanks to clinical and programmed surveillance systems, the absence of BT virus circulation was shown for the second year in a row throughout all of mainland France. Since the last bluetongue outbreak detected dated back to June 2010, the French authorities were able to declare on 14 December 2012, in compliance with the EU regulations, that mainland France was officially BT-free.

Keywords

Bluetongue disease, surveillance, ruminants, disease-free status

Résumé

Surveillance de la fièvre catarrhale ovine en 2012 : recouvrement du statut indemne en France continentale

La fièvre catarrhale ovine (FCO) est apparue dans le Nord de la France à la fin d'année 2006 puis s'est rapidement propagée sur l'ensemble du territoire jusqu'à infecter plus de 30 000 élevages français en 2008. Depuis, l'ensemble de la France continentale était considérée comme une zone unique de protection vis-à-vis des sérotypes 1 et 8 considérés comme endémiques. Après plusieurs campagnes de vaccination obligatoires (2008-2010) puis volontaires (2010-2012), le nombre de foyers a été drastiquement réduit. En 2012, les dispositifs de surveillance évènementielle et programmée ont permis de démontrer pour la deuxième année consécutive l'absence de circulation virale sur l'ensemble du territoire continental. Le dernier foyer de FCO ayant été détecté en juin 2010, les autorités françaises ont pu déclarer, conformément à la réglementation européenne, le territoire continental indemne de FCO le 14 décembre 2012.

Mots clés

Fièvre catarrhale ovine, surveillance, ruminants, statut indemne

Box 1. Surveillance and control measures for bluetongue in mainland France in 2012

Objectives of surveillance

- To detect any recurrence of viral circulation for endemic serotypes (1 and 8).
- To identify any introduction of an exotic serotype.
- To regain the official bluetongue-free status of mainland France.

Monitored population

Domestic ruminants.

- Outbreak surveillance

Clinical surveillance requires that all holders of animals from susceptible species and all state veterinarians notify the administrative authorities of any clinical signs suggestive of BT. This notification is followed by monitoring of the suspect farm and a nationally harmonised diagnostic protocol.

- Programmed surveillance

Commission Regulation (EC) No 1266/2007 defines the minimum prevalence that national bluetongue surveillance systems must be able to detect to demonstrate absence of viral circulation. EU requirements concerning this target prevalence level were changed in 2012. In the first half year, target prevalence detection was 2% (with a 95% confidence interval) by geographic unit, corresponding to 150 samples per *département* per month. In the second semester, the official target prevalence detection level was increased to 5%, corresponding to 60 samples per *département* per month.

The preferred target group for sampling was to be young non-vaccinated cattle. Collection of samples was coordinated by DDecPPs with three different scenarios: i) slaughterhouse sampling; ii) sentinel farm sampling; iii) random sampling during veterinary visits for other reasons.

- Diagnostic protocol

The same diagnostic protocol was applied across the country, whether sampling was part of programmed surveillance or clinical suspicion (each step described below is carried out only if the preceding step provided a non-negative result):

> *1/ Group RT-PCR (without serotype targeting) by the Departmental testing laboratory*

> *2/ Typing RT-PCR for serotypes 1 and 8 (to rule out any exotic serotype) by the Departmental testing laboratory*

> *3/ Group RT-PCR and typing RT-PCR by the NRL (on the same sample tested by the Departmental testing laboratory)*

> *4/ Viral isolation in embryonated eggs and/or on Culicoides cell lines (KC cells) by the NRL.*

The reference technique for confirming or ruling out an outbreak was viral isolation. In parallel to this method, RT-PCR was carried out at D+15 for suspect animals, making it possible to rule out suspicions without waiting for the viral isolation result, when the result was negative.

Control measures in place for 2012

In the event of clinical or analytical suspicion, the farm of origin is placed under Prefectural monitoring order (APMS) while results of tests performed by the NRL are pending.

If an outbreak is confirmed with an exotic viral serotype (i.e. other than 1 and 8), the national emergency health intervention plan is implemented by the Prefect.

If an outbreak is confirmed with an endemic viral serotype (1 or 8), the farm is placed under Prefectural declaration of infection (APDI). The vaccine status of the animals is assessed and, if necessary, animals with non-valid vaccination are vaccinated by the health service veterinarian.

No euthanasia or slaughter procedures apply.

Regulations

- Council Directive 2000/75/EC laying down specific provisions for the control and eradication of bluetongue
- Commission Regulation (EC) No 1266/2007 on implementing rules for Council Directive 2000/75/EC as regards the control, monitoring, surveillance and restrictions on movements of certain animals of susceptible species in relation to bluetongue
- Ministerial Order of 22 July 2011 establishing the technical and administrative framework applicable to bluetongue control in mainland France

In 2012, three schemes were in place for surveillance of bluetongue: clinical surveillance, active laboratory-based (programmed) surveillance (Box 1) and entomological surveillance (See paper Balenghien *et al.* in this issue). Surveillance in Corsica involves a specific procedure (Box 2).

Results of viral circulation surveillance

Outbreak surveillance

In 2012, investigations concerning clinical suspicions were conducted in 37 *départements* (Figure 1). In total, 151 cattle from 96 different farms and 20 sheep from 5 different farms underwent virological testing recorded in the SIGAL database as reference interventions for clinical suspicion. Overall, 64% of suspected cases were recorded between April and August (Figure 2).

Virological analyses made it possible to rule out all suspected cases. No clinical cases were therefore detected in 2012.

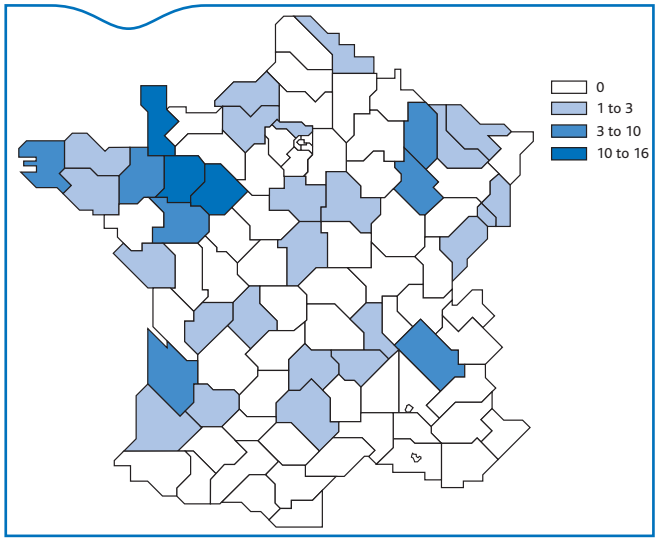

Figure 1. Number of farms (all species) with clinical suspect cases for bluetongue in 2012 by *département*

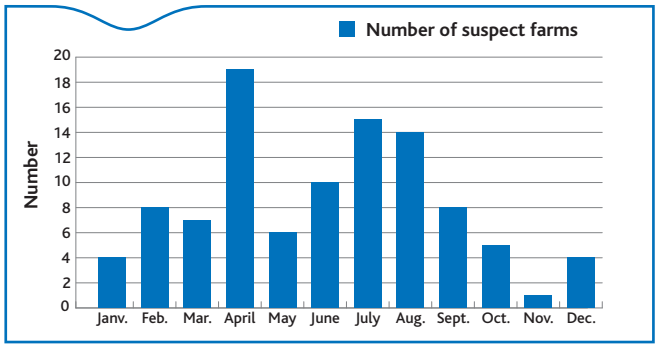

Figure 2. Number of farms with clinical suspicion in 2012 by month

Programmed surveillance

For the full year, 1,260 animals were to be tested virologically by PCR in each *département* (except those with very low populations), for a national objective of 109,320 tests. Ultimately, 99,401 analyses were performed, yielding a national effective rate of testing of 91% (Figure 3).

In line with instructions, 92% of recorded analyses in SIGAL corresponded to cattle, 7% to sheep and 1% to goats. Of the 67 DDecPPs responding to the question on programmed surveillance methods during the annual survey of the Animal Health Office, 45 (67%) used a sentinel model, 20 (30%) a slaughterhouse model, and 14 (21%) a random model. Each of these models accounted for 74%, 21% and 5% of all samples taken, respectively. These proportions were similar in cattle and in small ruminants.

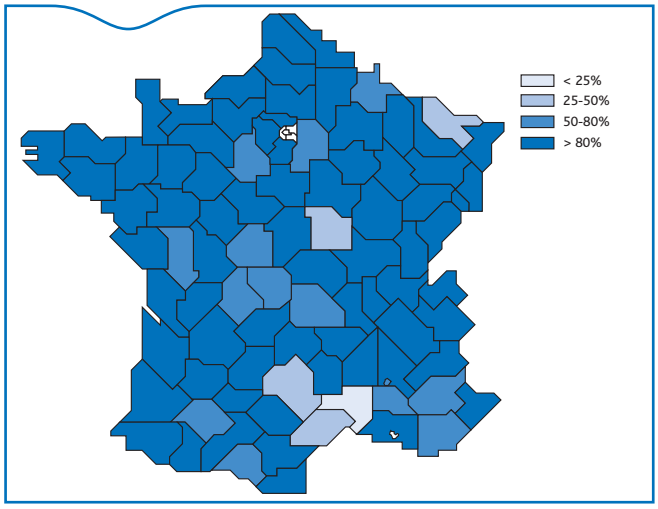

Figure 3. Rate of bluetongue sampling in 2012 by *département*

Like in 2011, no viral circulation was detected on the mainland through programmed surveillance. Non-negative results were obtained by some Departmental testing laboratories (Table 1), but all these suspect cases were ruled out following investigation by the NRL (negative viral isolation and negative re-rest PCR at D+15).

Table 1. Number of first-line non-negative results for BT recorded in SIGAL in cattle by sampling model and suspect serotype in 2012 (all these suspect cases were ruled out following investigation by the NRL)

	Sentinel sampling	Slaughterhouse sampling	Random sampling
Serotypes 1 and 8	44	38	28
Serotype 1 only	12	27	3
Serotype 8 only	9	2	1
Total	**65**	**67**	**32**

In all, 173 non-negative samples on PCR (167 for cattle and 6 for sheep), representing cases of analytical suspicion, were investigated by the NRL. Forty of these cases were immediately ruled out by initial PCR (performed on the sample taken on D0) and 133 were ruled out on PCR with negative results carried out on D+15 and on negative viral isolation (Table 2 and Figure 4).

It is difficult to determine the origin of the non-negative PCR findings. Several hypotheses can be made, including long-term persistence of viral RNA after prior infection, very low-level circulation of a non-infectious virus, or detection of the BTV genome in the inactivated vaccine. Overrepresentation of samples collected at the slaughterhouse or randomly among those showing non-negative results (Table 1) could point to the vaccine hypothesis. Clearly, the vaccine status of sampled animals (not recorded in the databases) was more difficult to check when the samples were taken using these models, compared to those from the sentinel model.

However, all these samples with a Ct below 35 were inoculated on eggs and/or KC cells and all these isolation tests were found to be negative, confirming the absence of infectious virus.

Cost

In 2012, the French government spent about 4.3 million Euros on bluetongue surveillance, including about 3.9 million on programmed surveillance (blood sampling and laboratory analyses), and 400,000 Euros for entomological surveillance (trapping, diagnosis and laboratory analyses). However, the State discontinued funding for vaccination in 2011 with a switch to voluntary vaccination paid by livestock breeders. The 2012 surveillance and control programme for bluetongue organised by the authorities was co-funded by the European Union for an amount of 1.2 million Euros.

Table 2. Results obtained by the NRL on investigation of suspect samples

Species (number of samples)	Results for PCR carried out by the NRL on D0 samples				Result of viral isolation
	RNA not detected	RNA detected Serotypes 1 and 8	RNA detected Serotype 1 only	RNA detected Serotype 8 only	Presence of virus
Cattle (167)	40	109	7	11	0
Sheep (6)	0	6	0	0	0

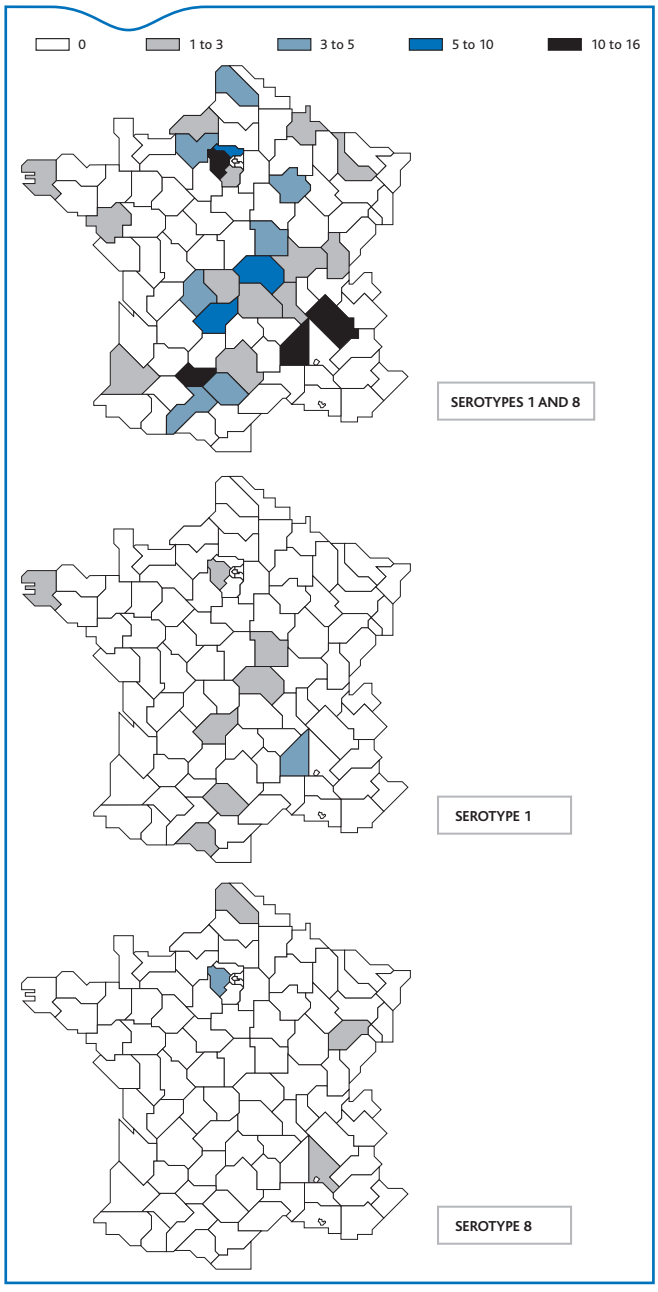

Figure 4. Number of suspect samples (all species) for serotypes 1 and 8 (left), serotype 1 (centre) and serotype 8 (right) by *département*. All these analytical suspected cases were ruled out by the NRL

Discussion and outlook

After its introduction in 2006, bluetongue rapidly spread across the country, affecting more than 30,000 farms in 2008. However, following two mandatory vaccination campaigns (2008-2010) and two voluntary vaccination campaigns (2010-2012), and thanks to combined efforts by the authorities and professionals in the sector, the number of outbreaks of bluetongue declined rapidly until eradication of the disease in 2010 (Table 3). After two full seasons of vector activity during which the surveillance programme demonstrated the absence of viral circulation, mainland France was declared disease-free on 14 December 2012, in accordance with European regulations.

The Epidemiological Surveillance Platform for Animal Health (ESA Platform) was asked to put forward a proposal on changes to the surveillance programme for bluetongue to adapt procedures to this new epidemiological picture, while remaining compliant with European regulations. The follow-up group proposed that the 2013 surveillance programme should be based on serological screening in the autumn on young non-vaccinated cattle exposed to *Culicoides* bites.

Table 3. Change in the number of outbreaks between 2006 and 2012

Number of outbreaks	2006	2007	2008	2009	2010	2011	2012
Serotype 1	0	3	4,932	9	1	0	9
Serotype 8	6	15,527	27,510	77	0	0	0

Box 2. Surveillance of bluetongue in Corsica in 2012

Corsica has had a specific surveillance programme for bluetongue in place since 2007 that is different from procedures in mainland France. Until 2012, the system was based on first-line serological testing.

Background of the disease in Corsica
Serotypes 2, 4 and 16 of bluetongue emerged on the island in 2000, 2003 and 2004, respectively, with an epizootic peak in 2001 when 326 outbreaks of serotype 2 were confirmed. No outbreaks have been confirmed since March 2005.

Outbreak surveillance
Same procedures as in mainland France.

Programmed surveillance
In 2012, programmed surveillance of bluetongue in Corsica was based on detection of antibodies in young non-vaccinated cattle. In all, 3000 calves were tested annually at the slaughterhouse. Animals underwent first-line testing by accredited laboratories using an ELISA method. The CIRAD NRL performed confirmation tests using ELISA and seroneutralisation on seropositive samples. In parallel, suspect animals were analysed by RT-PCR at the ANSES NRL.

Control measures
Corsica is a regulated zone for serotypes 1, 2, 4, 8 and 16. Suspected outbreaks with these serotypes do not prompt implementation of an emergency health intervention plan. Voluntary vaccine campaigns were considered for sheep (serotypes 1, 2, 4 and 8), and cattle (serotypes 1 and 8) but vaccination was not implemented because the bivalent 2/4 vaccine was not available.

Results and outlook
Although no clinical cases of the disease have been detected in Corsica since 2005, the results of programmed surveillance obtained in 2012 were difficult to interpret. Serological analyses (seroneutralisation) performed by the CIRAD NRL found non-negative results for various serotypes (1, 4, 8, and 16), which could be caused by very low-level viral circulation, a serological vaccine marker, or possible non-specific reactions. These results were not able to demonstrate clear absence of viral circulation on the island. As a result, the surveillance system was adapted in 2013 to enable Corsica to work towards recovery of the disease-free status. This process was however interrupted by the occurrence of serotype 1 clinical outbreaks in September 2013 in both *départements* of the island.

Culicoides population activity in 2012 and four-year review of the surveillance system

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Abstract

Wide-scale monitoring (160 traps) of the activity of *Culicoides* populations took place from 2009 to 2012 throughout both mainland France and Corsica following bluetongue virus epizootics. We compared the diversity and dynamics of *Culicoides* throughout France in 2012 with previous years. We focused on maps of vector inactivity, summarising results from 2009 to 2012.

Keywords

Entomological surveillance, *Culicoides*, period of vector inactivity, bluetongue

Résumé

L'activité des populations de* Culicoides *en 2012 et bilan des quatre années du dispositif de surveillance

Un réseau de surveillance entomologique (160 pièges) des populations de Culicoides *a été en activité entre 2009 et 2012 en France continentale suite à la transmission du virus de la fièvre catarrhale ovine (FCO). Nous présentons ici la diversité et la dynamique des* Culicoides *sur l'ensemble du territoire en 2012 en regard des années précédentes. L'accent est mis sur les cartes de période d'inactivité vectorielle synthétisant les résultats 2009-2012.*

Mots clés

Surveillance entomologique,* Culicoides*, période d'inactivité vectorielle, fièvre catarrhale ovine

Between 2009 and 2012, *Culicoides* populations were monitored in accordance with European regulations, using 160 traps located across the country, with one or two traps per *département*. Trapping took place once a week in the spring and autumn, and once a month the rest of the year, under the responsibility of the Departmental Directorate for Protection of the Population (DDecPP). Collected specimens were sent to three sorting centres for identification and counting: French Agricultural Research Centre for International Development (Cirad) in Montpellier, Interdepartmental Alliance for Mosquito Control on the Mediterranean Coast (EID-Med) in Montpellier, and to the Strasbourg Institute for parasitology and tropical diseases (IPPST). Mainland France recovered its bluetongue-free status in December 2012, and trapping has been restricted to Corsica since January 2013. The 2012 update is intended as an overview of four years of surveillance of *Culicoides* populations.

In 2012, 3,365 collections were performed with identification already completed at the time of drafting this article, corresponding to 85% of the theoretical collection target. In all, 1,235,452 *Culicoides* were captured, belonging to at least 69 species *versus* 77 in 2009, 69 in 2010 and 79 in 2011. This major collection effort organised by the network identified several new species in France between 2009 and 2012: *Culicoides abchazicus*, *Culicoides manchuriensis* and *Culicoides ibericus*. The *Culicoides* populations found in farms in France were, like in previous years, dominated by species of the Obsoletus group (93% of specimens), primarily *C. obsoletus* and *C. scoticus* (morphologically indistinguishable and accounting for 80% of collected insects), *C. dewulfi* (10%) and *C. chiopterus* (3%). Population abundances in 2012 were particularly high in January because of warm temperatures, much like the situation observed at the end of 2011, making it impossible to declare a period of vector inactivity during winter 2011-

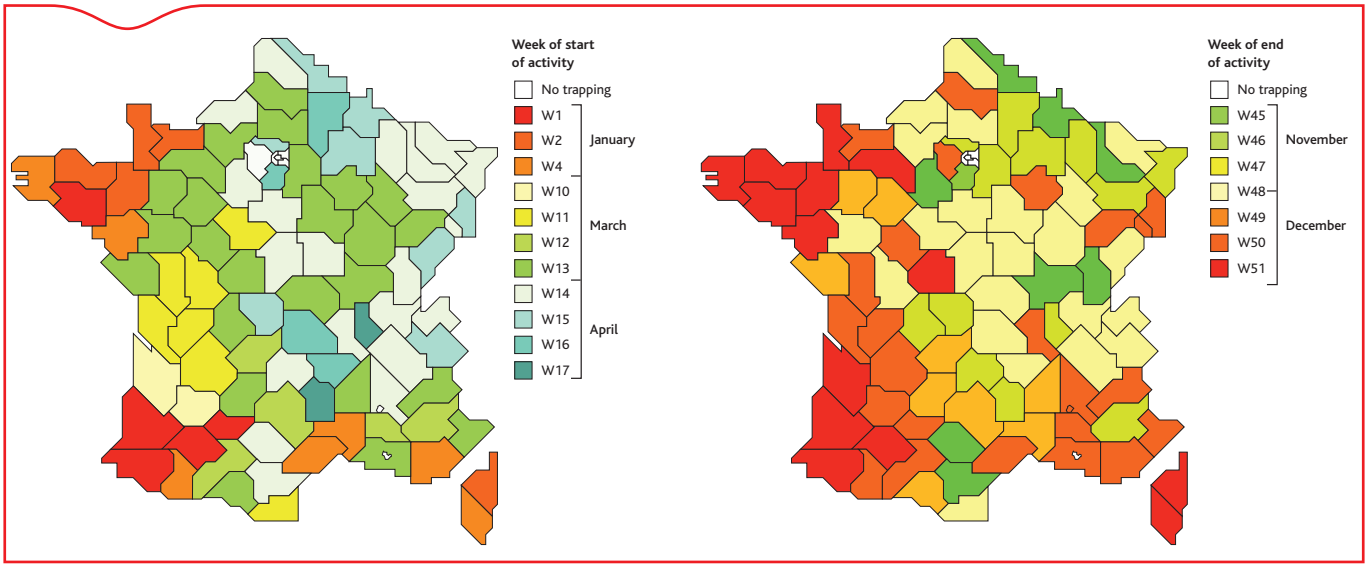

Figure 1. Activity of *Culicoides* populations in France between 2009 and 2012

The map on the left shows the earliest week of activity observed between 2009 and 2012, and on the right, the latest week of activity. According to the regulatory definition, the population is considered active when there are more than five parous females/trap/day.

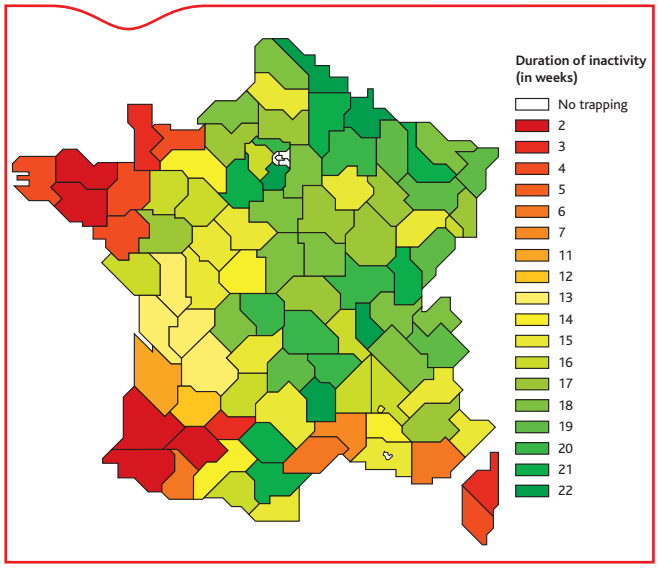

Figure 2. Minimum theoretical duration of *Culicoides* population inactivity observed in France between 2009 and 2012

This duration was calculated by selecting the earliest date of activity by *département* between 2009 and 2012, and the latest data of activity, and by determining the interval between these two dates: (52 - [latest date - earliest date of activity]).

2012. Measured abundances between March and September 2012 do not appear to be different from those found in previous years, except in April when numbers were lower. However, abundances were lower in October compared to previous years, related to lower temperatures around the middle of the month, and in November despite mean temperatures slightly higher than the normal range, but nonetheless inconsistent between regions. Temperatures were cooler in the North-West and generally higher than normal in the South and East. Colder averages at the start of December were followed by a mild year end, with maintenance of *Culicoides* population activity on the Atlantic seaboard.

Determining the period of inactivity of *Culicoides* was one of the main objectives and tasks for this entomological surveillance network between 2009 and 2012. Pooled activity data for the four years provides a geographical representation of the start and end of *Culicoides* activities (Figure 1).

This map shows that the start of activity can occur very early, beginning in January in Corsica, the far South-West and Bretagne, and in some *départements* on the Mediterranean coast (Figure 1). By early March, activity is found around these areas: southern part of the Atlantic seaboard and *départements* in the far South-East. Activity is recorded for the rest of the country between the end of March and the end of April, with a general gradient from the South-West to the North-East and a late start in the Massif Central. Activity then stops in early November in the far North-East, and by the end of November, activity has stopped across two-thirds of the country, extending from the North-East. The Atlantic seaboard, the far South-East and Corsica then follow in December. Certain *départements* present an earlier or later start to activity than neighbouring *départements*. This can be explained by differences in local weather conditions, environments and farming practices, such as the dates animals are moved from grazing to indoor facilities, with relocation of traps from outside to inside buildings. As a parallel to these two maps, we have produced a map showing the minimum duration of vector inactivity (Figure 2). The period of inactivity of *Culicoides* populations, from a regulatory perspective, is very short to non-existent in Corsica, in the far South-West, and secondarily in Bretagne and certain *départements* on the Mediterranean coast. This period of inactivity lasts two to four months in the Western third of the country and the far South-East, and up to five months in the Massif Central and North-East.

Acknowledgements

We warmly thank all those involved in trapping activities over the past four years for their essential contribution to the functioning of this network: DDecPP, GDS, and EID-Med personnel, as well as livestock farmers.

Overview of the surveillance of congenital infection by the **Schmallenberg virus** in domestic ruminants in 2012

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Abstract
Schmallenberg virus (SBV) was first detected in France in January 2012 with the birth of ruminants displaying congenital birth defects (arthrogryposis-hydranencephaly syndrome (AHS)). A national surveillance system for congenital SBV was launched by the government and then pursued through farmers' organisations. About 4,000 affected farms were detected in 2012 throughout France. SBV spread quickly and extensively in 2012 due to the absence of control and prevention measures.

Keywords
Schmallenberg, orthobunyavirus, ruminants, emergence, National Platform for Epidemiological Surveillance

Résumé
Bilan de la surveillance de l'infection congénitale par le virus Schmallenberg chez les ruminants domestiques en 2012
L'émergence du virus Schmallenberg (SBV) a été révélée en France à partir de janvier 2012 par la détection de formes congénitales de l'infection chez les ruminants, caractérisées par un syndrome arthrogrypose-hydranencéphalie (« SBV congénital »). Une surveillance du SBV congénital a été déployée par l'Etat puis, dans un second temps, relayée par les organisations professionnelles (GDS France). Au total, la surveillance mise en œuvre a permis d'identifier près de 4 000 exploitations atteintes en 2012 sur la quasi-totalité du territoire, révélant une diffusion rapide et massive de ce nouveau virus contre lequel les moyens de prévention ou de lutte étaient inexistants.

Mots clés
Virus Schmallenberg, orthobunyavirus, ruminants, émergence, Plateforme d'épidémiosurveillance en santé animale

Emergence of the Schmallenberg virus

The Schmallenberg virus (SBV) was first identified in November 2011. It had been observed in the summer of 2011 in association with a febrile diarrhoea syndrome in cattle in the Netherlands and Germany (Hoffmann *et al.*, 2012). In mid-December 2011, congenital forms of SBV infection were identified for the first time in lambs in the Netherlands. These forms of the disease, characterised by an arthrogryposis-hydranencephaly syndrome, were subsequently identified in calves and kids (Box 1). By analogy with genetically comparable viruses that cause similar lesions (the Akabane virus), it has been estimated that the congenital deformities resulted from infection during the second month of gestation in small ruminants (i.e. three or four months before parturition), and the third to the sixth month of gestation in cattle (i.e. three to seven months before birth) (Kirkland *et al.*, 1988). SBV is principally transmitted by *Culicoides* (midges – although the role in transmission of mosquitoes and other arthropods has not been ruled out) (Scholte *et al.*, 2013).

Box 1. The two clinical manifestations of the Schmallenberg disease

Congenital SBV: Deferred manifestation of foetal infection by SBV leading to the birth of offspring in cattle, sheep and goats that are most frequently non-viable, through abortions, premature births or stillbirths, associated with various congenital malformations (arthrogryposis, shortened hock tendons, twisted neck, deformed sternum and spine, deformed jawbones and skull). Nervous disorders may also be observed.

Acute SBV: Acute manifestation of infection by SBV characterised in cattle by episodes of diarrhoea, drop in milk production and hyperthermia, possibly associated with the return to oestrus and abortions at the start of gestation. This clinical form has not been reported in small ruminants (Wernike *et al.*, 2013). It should be noted that there has been as yet no organised surveillance of acute SBV in France.

Methods used to monitor congenital SBV

Surveillance of the "first wave"

In response to the European alert concerning the emergence of congenital forms of SBV, the DGAL and the French Epidemiological Surveillance Platform for Animal Health (ESA platform) have jointly deployed a congenital SBV surveillance system since January 2012 (Memorandum DGAL/SDSPA/N2012-8007 of 4 January 2012).

This was a clinical outbreak surveillance system for arthrogryposis-hydranencephaly syndromes in newborn small ruminants (Dominguez *et al.*, 2012a). The goal was first to detect the presence of the virus in France, and then to document the geographic distribution of the disease in relation to the dissemination of the virus during the period of midge activity in 2011 (i.e. the "first wave" of the spread of the virus). The probability of contamination related to this first wave was considered low after the drop in vector activity (i.e. after January 2012); for this reason, surveillance of the first wave of congenital SBV was interrupted after four months for small ruminants (i.e. at the end of May, after already being scaled down mid-April in the most heavily affected *départements*) and after seven months in cattle (i.e. end of August) (Dominguez *et al.*, 2012b).

From January to April 2012, SBV diagnosis was carried out by virological analysis (rtRT-PCR) on a brain sample from clinically suspect newborns, or, if this was not possible, on a blood sample from the mother. ELISA kits were available from April 2012 (Bréard *et al.*, 2013) and the

biological diagnosis of congenital SBV was performed by serological analysis of sera from clinically suspect newborns (preferably collected before colostrum intake), or otherwise of serum from the mother (Memorandum DGAL/SDSPA/N2012-8087 of 18 April 2012).

Surveillance of the first wave of congenital SBV was funded by the State (visit by veterinarian on suspicion and biological analyses).

Surveillance of the "second wave"

From May 2012 onwards, cases of acute SBV (Box 1) were confirmed in adult cattle in the Pyrénées-Atlantiques *département* (Sailleau *et al.*, 2013) indicating that the SBV virus had survived the winter. It was also demonstrated that SBV continued to spread during the period that the vectors were active in 2012 by the positive results of the virological analyses carried out during the summer under the testing of animals for export. Cases of congenital SBV resulting from the spread of the virus during the vectors' period of activity in 2012 (the "second wave") were therefore expected, four months (at the earliest) after recommencement of the spread of the virus, i.e. from September onwards.

The initial data collected on congenital SBV seemed to indicate only a generally moderate effect on livestock production (Dominguez *et al.*, 2012c). This led the French authorities, in agreement with the professional bodies, to consider Schmallenberg as a non-priority disease, in compliance with the position of the international organisations (the disease has not been added to the list of animal diseases drawn up by the OIE (OIE, 2012), nor to the Annex to the European Commission's Decision 90/424/EEC). As such, the disease ceased to be subject to mandatory surveillance by the State. Surveillance of the second wave was carried out by the network of the Farmers' animal health protection organisation (GDS) as a part of the ESA Platform, with a view to plotting the geographical distribution of congenital SBV from 1 September 2012.

Biological diagnosis was performed by serological analysis on sera from the newborns, preferably collected before colostrum intake. In cases where sampling under these conditions was impossible, in areas considered to have been heavily affected by the first wave[1], the diagnosis was performed by viral analysis on a brain sample from the suspect newborn (photographs showing the malformations were also considered as sufficient evidence, given the very specific nature of the malformations observed), and in areas considered to have been only slightly affected by the first wave, diagnosis was carried out by serological analysis on a serum sample from the mother.

The veterinarian's visit and, where appropriate, the sampling were funded by the State under the terms of the mandatory reporting of abortions for brucellosis surveillance. Analyses, however, were paid for by the farmers, except where specific local measures had been taken (e.g. grants from the GDS or from local authorities).

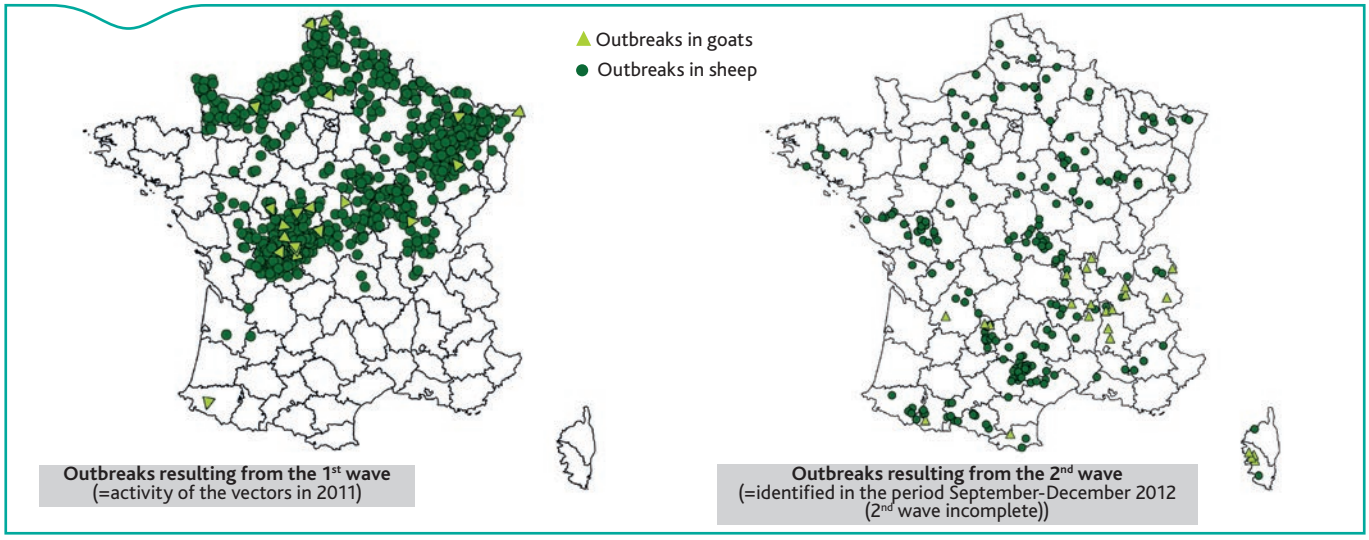

Figure 1. Location of outbreaks of congenital SBV identified in small ruminants in 2012

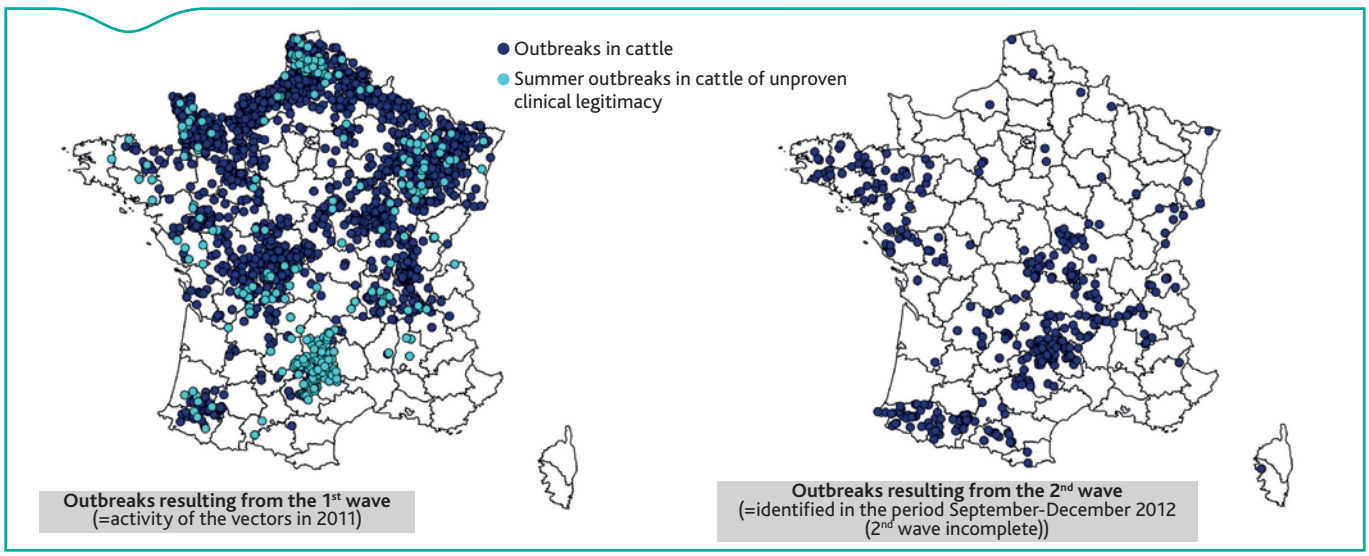

Figure 2. Location of outbreaks of congenital SBV identified in cattle in 2012

(1) *Départements* in which more than 20 outbreaks of congenital SBV had been identified by 15 June 2012 [http://www.plateforme-esa.fr/images/documents/surveillance%20sbv%20congnital_document%20technique.pdf

Results

In France, the first cases of congenital SBV in domestic ruminants were confirmed at the end of January 2012. They related to births at the start of January (with the delay resulting from the need to obtain biological confirmation of clinical suspicions). These first outbreaks detected may have corresponded to infection of ewes dating from September 2011 and infection of cows between May and September 2011. It can therefore be estimated that SBV probably started to circulate among domestic ruminants no later than September 2011 (Dominguez *et al.*, 2012d; Dominguez *et al.*, 2012e). Furthermore, retrospective serological studies carried out in the Meurthe-et-Moselle and Manche *départements* confirmed that the virus was already circulating in October 2011, but it had not been detected in serum collected in August and September 2011 (Zanella *et al.*, 2013).

Surveillance of the first wave of congenital SBV (January-August 2012) identified 3,276 affected farms: 2,117 cattle farms, 1,139 sheep farms and 20 goat farms. The density of the outbreaks of congenital SBV resulting from the first wave was higher in *départements* in the northern half of France (except for western France) (Figures 1 and 2). Northern France had been exposed for longer, because SBV may have arrived in France *via* the north-east.

The peak of the epidemic of congenital SBV resulting from the first wave of viral circulation was reached first in small ruminants (Figure 3)

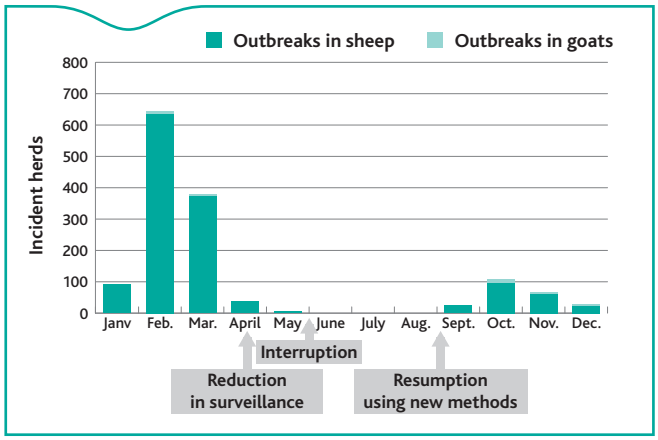

Figure 3. Monthly incidence of congenital SBV in herds of small ruminants in France in 2012

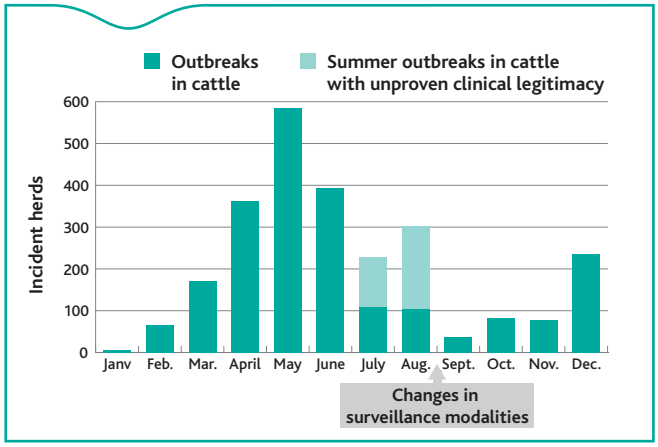

Figure 3. Monthly incidence of congenital SBV in herds of cattle in France in 2012[3]

3. The fall-off in the summer (July-August) of the incidence of congenital SBV was only slight in cattle. An inspection of the corresponding suspicion sheets showed that for 316 outbreaks in cattle reported during the summer, clinical legitimacy for the suspicion of congenital SBV could not be established by analysis of these records; it cannot be ruled out that some of these outbreaks could concern acute infection with SBV wrongly reported as outbreaks of congenital SBV ("summer outbreaks in cattle with unproven clinical legitimacy") and then, later because of the longer interval between foetal infection and birth, in May in cattle (Figure 4).

The resumption of the epizootic episode in September (outbreaks resulting from the second wave) was more intense in areas that had not been heavily affected by the first wave (Figures 1 and 2).

At the end of December 2012, surveillance of the second wave[2] had led to the identification of 621 affected farms (393 outbreaks with cattle, 205 with sheep, 23 with goats).

Discussion

Congenital infection by SBV was confirmed in about 4,000 farms in France in 2012. This is only a fraction of the farms where this emergent virus had circulated (gravid females possibly absent during the window of exposure to risk; rate of clinical expression of the congenital infection by the SBV; probable under-reporting of clinical suspicion; variability of the sensitivity of the surveillance between the first and the second waves etc.). The surveillance system set up demonstrated that the new virus had spread across almost the entire territory of France. There was still no possibility for prevention and control of SBV in 2012 (no vaccine, ineffective measures for protection against the vectors, no regulatory measures restricting the movements of animals).

Cattle and sheep are the two most severely affected species by the circulation of SBV, while the incidence of congenital SBV in goats seems to have remained low. Regarding wild animals, specific serological surveys showed that the SBV had circulated fairly widely among wild ruminants in 2012. Red deer, for instance, were found to be seropositive in different regions of France (Laloy *et al.*, 2013).

The ESA Platform demonstrated its capacity to mobilise rapidly in response to an emergency as well as its added value in terms of coordination. It facilitated the process of passing responsibility for surveillance from the State to professionals on the ground. It also contributed to carrying out the surveys designed to raise awareness of this new disease (impact on farming (Dominguez *et al.*, 2012c; Touratier *et al.*, 2012); circulation of the virus within herds (Gache *et al.*, 2013); clinical presentation of acute SBV (Collin *et al.*, 2013)).

The considerable spread of SBV in France in 2011 was not detected clinically. In neighbouring countries with non-specific national surveillance schemes, disorders related to the acute infection of cattle with SBV had attracted attention (especially in the Netherlands, although these observations did not lead immediately to an etiological description) (Bartels *et al.*, 2011). The effort to reinforce the State's ability to detect unusual animal health events in France must be continued, as well as the effort to develop the capacity to respond rapidly to emerging animal health issues (raising awareness among networks of partners in the field, reflex procedures, flexible generic tools for managing data, etc.).

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Porcine brucellosis in France in 2012: three outbreaks including one in a local breed

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Abstract
As in previous years, surveillance of porcine brucellosis in 2102 was based primarily on outbreak surveillance. While the outbreaks discovered in 2010 had shown for the first time since 1993 that holdings of local breeds could also be affected by brucellosis, in the same way as other outdoor holdings, this trend continued in 2011 and 2012 with three outbreaks affecting pigs from both local and industrial breeds in 2012. No outbreaks were detected in intensive farming despite five serological suspicions.

Keywords
Regulated disease, porcine brucellosis, epidemiological surveillance, swine

Résumé
Brucellose porcine en France en 2012 : trois foyers dont un en race locale
Comme pour les années précédentes, la surveillance de la brucellose porcine en 2012 a reposé principalement sur une surveillance événementielle. Alors que les foyers découverts en 2010 avaient révélé pour la première fois depuis 1993 que les élevages de races locales pouvaient également être concernés par la brucellose, au même titre que les autres élevages porcins de plein air, cette tendance se confirme en 2011 et 2012 avec trois foyers portant à la fois sur des porcs de races locales et de race industrielle en 2012. Cinq suspicions sérologiques ont été infirmées en élevage hors-sol.

Mots clés
Maladie réglementée, brucellose porcine, épidémio-surveillance, suidés

Here, we present the results of porcine brucellosis surveillance in 2012 (see Box below for details on the surveillance programme).

Results

A total of 5,303 analyses were carried out in quarantine and artificial insemination (AI) centres (101 holdings tested). Of these analyses, 235 were positive (4%). In 2012, positive results involved 40 holdings, with 1 to 102 positive tests per holding (i.e. an average of 6 positive tests per holding). The proportion of positive results per holding varied between 0.8% (1 positive out of 1,026 tests) and 14% (102 positive out of 717 tests).

In 2012, there were seven suspected cases involving outdoor holdings: three following observation of clinical signs (abortion, infertility), three following serological tests and one in a holding having an epidemiological connection with an infected holding. Routine serological tests were set up in 2011 for certain local breeds in which outbreaks had previously been observed, particularly in pig breeds shown at the Paris International Agricultural Show (Bronner *et al.*, 2011). Of the seven suspected cases, three were confirmed.

Five cases were also suspected in an indoor holding after positive serological tests. None were confirmed.

Three suspected cases involved holdings with farmed wild boars. No outbreaks were confirmed.

In 2012, three outbreaks of porcine brucellosis — all in outdoor pig holdings — were notified in three French *départements* (Bouches du Rhône, Dordogne and Eure; Figure 1). For the three outbreaks, the pathogen was confirmed after isolation and identification of *Brucella suis* biovar 2 by the National Reference Laboratory (NRL). For one of these outbreaks, the suspected case began in late 2012 and was confirmed in early 2013.

For these three suspected outbreaks, 146 pigs were serologically tested, of which 42 were seropositive (BAT+ and CF+) and 12 underwent bacteriological testing, with isolation of *Brucella* for three of them, all from different outbreaks. The proportion of seropositive pigs per outbreak varied between 11% (n = 4 out of 36 pigs) and 100% (n = 7 pigs).

These outbreaks involved intensive pig farms as well as one small, family-run outdoor holding with a local breed (Noir de Bigorre - Gascon: a breed with a small population size, in which artificial insemination is not practised, and in which individuals are frequently transferred among different holdings).

These three outbreaks were discovered *via* passive surveillance, based on notification by the pig farmer or state veterinarian of suspicious clinical signs (abortions, early return to oestrus). One outbreak was initially thought to have arisen from pigs introduced from a holding found in a *département* where outbreaks had been detected in 2011. However, this assumption was not confirmed.

No outbreaks due to an epidemiological connection were detected in 2012.

Cost

In 2012, for the 80 *départements* for which data is available, the French government invested €4,700 in surveillance and control of porcine brucellosis. Laboratory costs amounted to €2,415 and veterinary costs were €2,364. These figures do not include the compensation that is paid out in cases of confirmed porcine brucellosis outbreaks.

Discussion

The profile of holdings affected by porcine brucellosis outbreaks in France has changed since 2010, with outbreaks involving local breeds and a higher proportion of secondary outbreaks.

In 2012, as observed in 2011 (Marcé *et al.*, 2012), the infection in holdings with local breeds appears to be continuing, although the proportion of secondary outbreaks was limited (one of the three cases was suspected to be a secondary outbreak). Wildlife remains the primary identified or suspected source of infection.

However, 2012 was the first year in which an outbreak was identified in south-eastern France. For the past 20 years, most outbreaks have occurred in western France where outdoor holdings are the most frequent (Figure 1). Generally speaking, there are few isolated cases. Therefore, the outbreaks detected may vary by area (some areas may have higher risks), by the degree of awareness on the part of

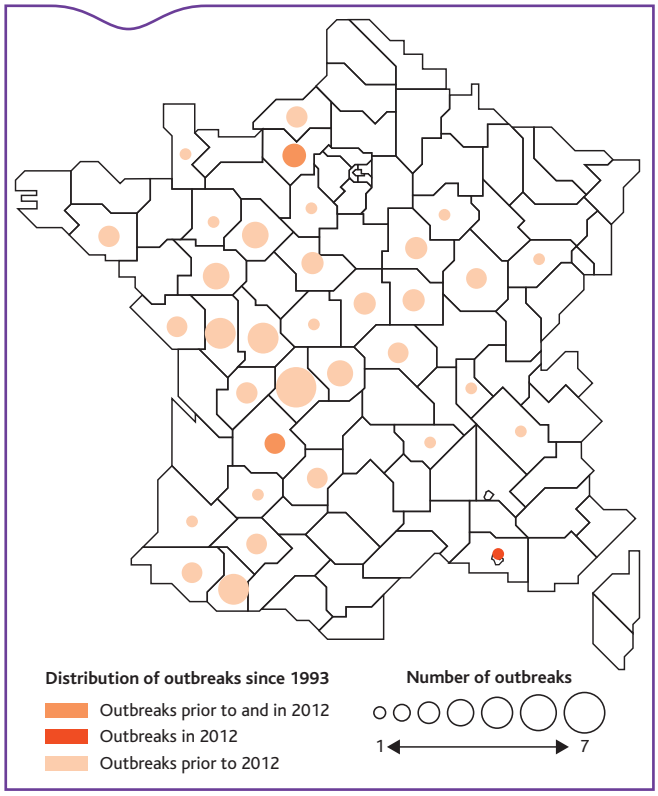

Figure 1. Geographic distribution of confirmed brucellosis outbreaks in pig holdings in France from 1993 to 2012.

farmers and veterinarian service staff who detect the clinical cases, or by the coverage of epidemiological investigations in the case of primary outbreaks. However, the relative importance of these three possibilities is not known.

Although 12 and seven outbreaks were reported in 2010 and 2011 respectively, the detection of only three outbreaks in 2012 does not effectively reflect a decrease in incidence. Outbreaks in outdoor holdings arise sporadically, depending on the frequency of contact with infected wild boars. Thus, from 1993 to 2012, the annual number of outbreaks varied between zero and 12 for a total of 82 outbreaks reported during the 10-year period.

Suspicions in farmed wild boar holdings were raised after non-routine serological testing (e.g. private requests during transfers between farms, when purchasing breeding boars or simply to determine the health status of a holding). Analyses on wild boars are usually done on young individuals, which are easier to catch and handle. The interpretation of results from tests on young wild boars (whose sample size may vary greatly) is very difficult, particularly because clinical signs, even in a declared outbreak, often go undetected in farmed wild boar holdings due to production practices. Suspected cases are resolved by preventive slaughter or further testing to prove the brucellosis-free status of the holding.

The three outbreak holdings reported in 2012 for domestic pig production had proper fencing. Although other contamination routes are possible, the risk of introduction *via* wildlife is very real and current regulations on fencing are not always sufficient to prevent contact between wild boars and sows likely to be in heat. Fencing is currently not mandatory for gilts, gestating sows after the fourth week following mating or artificial insemination, lactating sows and non-pubescent gilts. Thus, despite proper fencing, female pigs in oestrus may be at risk of contamination. Despite there being no regulatory requirement, all pens in outdoor holdings should be fenced according to the standards indicated in the Circular DPEI/SDEPA/2005-4073 of 20 December 2005.

The results of the porcine brucellosis surveillance obtained in 2012 highlight, as in 2010 and 2011, how important it is to encourage farmers to implement biosafety measures, to declare abortions and to properly diagnose abortions due to brucellosis. Active surveillance cannot be generalised or extended due to the limited specificity of the serological tests and the very low incidence of porcine brucellosis in France, making it cost-ineffective. However, active surveillance can occasionally compensate for the limits of passive surveillance, which has very low sensitivity, although it requires close and intensive monitoring of holdings, due to the high risk of false positives.

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Box. Porcine brucellosis surveillance and control measures

Objectives of the surveillance programme
The aim of porcine brucellosis surveillance is, for all swine holdings, to detect outbreak events rapidly, in order to prevent the spread of the disease to other holdings and, according to the strains involved, to prevent the risk of zoonosis. For quarantine centres and artificial insemination (AI) centres, the goal is to ensure that only disease-free boars are used for artificial insemination purposes.

- Passive surveillance in herds: animal health rules and public health protection measures in case of suspicion and confirmation.
- Active surveillance: individual testing of boars before admission to AI centres to prevent the spread of the disease (EC regulations).

Monitored population
Domestic swine and farmed wild boars throughout mainland France

Scope of surveillance programme
Brucella suis biovars 1, 2 and 3, *Brucella melitensis* and *Brucella abortus*

Surveillance procedures
Porcine brucellosis is monitored by passive surveillance (testing after observation of clinical signs) in all holdings, and active surveillance (routine serological testing) in quarantine centres and AI centres. Programmed surveillance was set up (professional initiative) in late 2010 for holdings of the Noir de Bigorre (Gascon) breed and for local breeds shown at the Paris International Agricultural Show.

- Outbreak surveillance

Based on the surveillance of symptoms typical of brucellosis infection: early abortion with early return to oestrus (abortion or embryonic resorption can affect up to 50% of breeding sows in a holding, 95% of breeding sows may be infertile), acute orchitis or any other reproductive disorder of enzootic nature. Arthritis and paresis arising from bone and joint injury can also indicate brucellosis.

- Programmed surveillance

Targets boars used for AI (which are also tested for Aujeszky's disease and classical swine fever), due to the potential role of semen in the spread of brucellosis (the combination of antimicrobials added to collected semen does not eliminate *Brucella*). This serological surveillance is not generalised to other types of holdings that may nonetheless run the risk of spread or introduction of *Brucella* because serological tests are known to have low specificity and frequent false positives.

A herd becomes suspect in one of the following three circumstances:

1. observation of epi- or enzootic clinical signs associated with positive serological tests;

2. herds with an epidemiological connection to an infected holding;

3. in accredited AI or quarantine centres, positive serological reactions as defined in Memorandum 2004/8134 of 12 May 2004.

- Epidemiological investigation during an outbreak (trace-back/trace-forward surveys)

For suspected outbreaks, samples are taken by state veterinarians for serological testing (blood samples in vacutainer collection tubes from all breeding pigs) or bacteriological analyses (peri- or endocervical swabs or samples of vaginal secretions in sows having aborted or those that show reproductive disorders, samples of lymph nodes and/or uterus tissue in sows having aborted, samples of affected testes for boars with orchitis, sample of joint fluid from any arthritic pig, after diagnostic slaughter).

Animal health rules
Given the low specificity of brucellosis symptoms, any suspected holdings will be placed under the surveillance of state veterinary services by prefectural monitoring order when the clinical suspicion is confirmed by positive serological results. However, for quarantine or AI centres, due to the impact that any delay would have for the notification of brucellosis, and given the type of surveillance (clinical and serological), these centres are placed under prefectural monitoring order as soon as positive serological test results are obtained.

- Outbreak definition

An outbreak of porcine brucellosis is confirmed when:

> *the Brucella bacterium has been isolated;*
> *when at least 10% of breeding pigs are seropositive;*
> *for accredited quarantine and AI centres, when the suspected pig(s) originate from a known infected holding.*

Except for quarantine centres, confirmation is thus based on isolation of the pathogen (high specificity, but low sensitivity), or positive serological results (low specificity, but high sensitivity, particularly due to cross-reactions with *Yersinia enterocolitica* O:9). In the absence of any suspicious clinical signs, isolated positive serological reactions do not in any way constitute a suspicion of brucellosis according to the Ministerial Order of 14 November 2005.

- Measures taken in the event of confirmed outbreaks

When the outbreak is confirmed, the prefectural monitoring order is replaced with a prefectural declaration of brucellosis infection. According to whether the bacteria could be typed and according to the *Brucella suis* biovar isolated, the fate of breeding pigs and growing-finishing pigs differs in terms of condemned meat and heat treatment. When an outbreak has been confirmed, the entire herd is culled. Ruminants and dogs on the premises are also tested. Epidemiological trace-back and trace-forward surveys are conducted for the six months preceding the first suspicion of outbreak. Depopulation is followed by cleaning and disinfection.

Regulations

– Directive 90/429/EEC laying down the animal health requirements applicable to intra-Community trade in and imports of semen of domestic animals of the porcine species
– Ministerial Order of 14 November 2005 laying down the animal health measures regarding brucellosis in captive swine
– Ministerial Order of 7 November 2000 laying down the animal health conditions required for disseminating swine semen

Review of surveillance of **Aujeszky's disease** in France in 2012: upholding of Aujeszky's disease-free status in mainland France

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Abstract

This article presents the results of surveillance of Aujeszky's disease in mainland France and Reunion Island in 2012. They show a continuous decrease in screened pigs from outdoor holdings – especially among grow-to-finish farms – despite the real risk of reoccurrence of the disease (circulation of the virus in wildlife) as previously shown with the 2010 episode. The number of screened pigs from breeding holdings is similar in 2012 to that in 2011. The priority is for all stakeholders 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, Aujeszky's disease, epidemiological surveillance, France, official control, swine

Résumé

Bilan de la surveillance de la maladie d'Aujeszky en France en 2012 : maintien du statut indemne de maladie d'Aujeszky en France continentale

Cet article présente les résultats de la surveillance de la maladie d'Aujeszky en France continentale et sur l'Ile de la Réunion en 2012. Ces résultats confirment, comme déjà initiée en 2011, la diminution du nombre de porcs d'élevages plein air dépistés, notamment en élevages engraisseurs, alors que le risque de réapparition de la maladie est réel du fait de la circulation du virus dans la faune sauvage, comme l'a montré l'épisode survenu en 2010. Le nombre de porcs dépistés en élevages sélectionneurs-multiplicateurs reste similaire à celui de 2010. Le maintien de la vigilance de l'ensemble des acteurs reste la priorité. Il est notamment important que les vétérinaires incluent dans leur diagnostic différentiel la maladie d'Aujeszky lors de symptômes (syndrome grippal, avortements) ne pouvant être rattachés avec certitude à une autre maladie.

Mots clés

Maladie réglementée, maladie d'Aujeszky, épidémio-surveillance, France, police sanitaire, suidés

Here, we present the results of the Aujeszky's disease (AD) surveillance programme in mainland France and Reunion Island for 2012 (see Box for details on the surveillance programme). The results presented in this report do not include Corsica (where AD surveillance was not in effect in 2012).

Population counts used in this report come from pig holding registration forms filed by farmers on or before 31 December 2012 (compiled in the BDPORC, the national pig identification database) and transmitted to DGAL's information system SIGAL). All pig keepers must register their pig holdings (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.

Sampling

Surveillance in breeding holdings

Surveillance was conducted in 415 breeding holdings out of the 522 holdings identified in the pig holding registry (80% coverage rate).

On average, 54 samples were taken per holding and per year, i.e. 14 samples per quarter, for a total of 22,625 samples. Compared to the 2011 average of 13 samples, the average number of samples per holding and per quarter increased slightly in 2012, reaching levels similar to those in 2009, 2011 and 2012 (Marcé *et al.*, 2011; Marcé *et al.*, 2012; Bronner *et al.*, 2010).

Overall, assuming that samples were only taken on breeding stock, and according to the pig population count recorded in the BDPORC database, 27% of breeding stock was tested in 2012, or 7% per quarter, i.e. the same levels as in 2011.

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

In all, 1,107 outdoor holdings (domestic swine or farmed wild boars) were listed as having been tested out of 2,620 holdings included in the database (2,124 outdoor pig holdings and 496 holdings with wild boars), i.e. a 42% coverage rate, with 11,524 samples taken.

The rate of routine testing varied with the type of domestic swine

Table 1. Testing for Aujeszky's disease in outdoor domestic swine holdings in 2012

Type of outdoor holding	Number of holdings registered*	Number of holdings tested	Proportion of holdings tested (%)	Number of samples	Mean number of samples per holding
Farrowing	223	148	66	1,595	11
Wean-to-grow	37	15	41	193	13
Grow-to-finish	1,114	408	37	4,365	11
Farrow-to-finish	763	468	61	4,397	9
Total (all types of outdoor holdings)	2,137	1,039	49	10,550	10

* Taken from the BDPORC database in January 2013 for mainland France. All *départements* were included, although 14 *départements* did not provide all information on Aujeszky's disease surveillance (including several *départements* with a high pig density or a high density of pig holdings that must undergo routine testing) and 37 *départements* did not validate their pig population counts. Holding categories differed slightly to those used in the 2011 report: the farrowing category includes farrowing and wean-to-grow farms, and wean-to-finish holdings were included in the grow-to-finish category.

holding, from 37% in grow-to-finish holdings to 66% in farrowing holdings (Table 1).

Of a total of 2,137 outdoor domestic pig holdings, the surveillance programme effectively covered 1,039 of them (48.6% surveillance coverage rate), for a total of 10,550 samples.

Surveillance in indoor production holdings (production of slaughter pigs)

Although routine testing is not mandatory, 116 holdings were screened for AD (4,460 samples).

In all, including all the holdings mentioned previously, 38,609 samples were taken for serological screening of AD.

Non-negative results

According to the data available for outdoor and indoor holdings, 13 sites obtained positive serological results (1% of tested holdings).

In outdoor holdings, 10 pig holdings showed at least one non-negative result for the first-line ELISA gB test (22 samples) and 4 holding sites showed at least one non-negative result for the first-line ELISA gE test. Due to these results, seven sites were placed under APMS (6 due to at least 1 non-negative result for the first-line ELISA gB test and 1 due to at least 1 non-negative result for the first-line ELISA gE test). Ten sites were visited again to collect enough serum for confirmatory tests (gE in particular). One site showed a non-negative second-line ELISA gB test (1 positive sample out of 20 analysed samples). However, the introduction of the AD virus in a pig holding rapidly causes high seroprevalence. Therefore, the absence of any active seroconversion between two series of analyses on samples from the holding demonstrates that the non-negative results are in fact non-specific serological reactions; these positive samples are described as coming from «single reactor» individuals (Annelli *et al.*, 1991). Therefore, none of the non-negative results were confirmed.

Additionally, high suspicions in a wild boar holding after annual routine testing (10 positive results out of the 12 sera tested) led to culling of the boars in the holding. Epidemiological investigations did not lead to detection of any outbreak.

In indoor production systems, three pig holding sites showed non-negative results for the first-line ELISA gB test. Two of the three holdings were placed under APMS. None of these results were validated by the NRL.

Data are not available for breeding holdings.

Clinical suspicions

One indoor holding showed suspicious clinical signs (Hautes-Alpes *département*). Forty pigs underwent serological tests, none were screened using PCR. The clinical suspicion was not confirmed. For three other holdings in mainland France, the NRL was asked to carry out diagnostic PCR tests on organs or nasal swabs. Following this AD-specific test, none of these holdings were placed under APMS as part of differential diagnosis recommended by a Memorandum but the PCR test effectively excluded the possibility of infection with the AD virus.

Costs

In 2012, in the 85 *départements* for which there was usable data, the French government invested more than €26,740 for surveillance and control of AD. Laboratory costs totalled €11,773 for routine testing (preventive measures) and €1,660 for control measures. Veterinary costs were €12,309 for routine testing and €1,268 for enforcing animal health rules. These figures do not include testing in breeding holdings that belong to the French pig breeding agency (ASP): €34,510 for sampling and serological analyses.

Discussion

No outbreaks of AD were identified in any holding in 2012. Only one holding showed suspicious clinical signs and at least four underwent confirmatory testing. PCR analyses carried out by accredited departmental testing laboratories are not yet systematically reported to the central database. Therefore, some confirmatory tests may not have been recorded and the number of tests may therefore be underestimated. As reporting of confirmatory PCR tests is useful for estimating the degree of surveillance required in French pig holdings, it should therefore be improved so that the NRL can document the analyses actually conducted by its network of accredited laboratories.

Furthermore, the episode that occurred in 2010 (Rose *et al.*, 2010) underscores the undeniable risk of the disease reappearing in mainland France *via* outdoor holdings and wildlife. 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 due to clinical signs of infection that can be attenuated at low pig densities. Combining outbreak and serological surveillance, in holdings with domestic pigs or those with 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 2012 serological surveillance with those of 2011 (Marcé *et al.*, 2012), although the same method (based on the pig holding registry) was used to identify the number of pig holdings in France. This is because the holdings were not categorised similarly. In 2011, wean-to-finish farms were wrongly counted in the wean-to-grow category, but included in the grow-to-finish category in 2012. Moreover, the pig holding registry is based on a single filing that is only amended when the initial information has changed. Given that the population counts in holding registries are based on the authorised holding population size and not on the effective number of pigs present, some farmers, when ceasing part of their activity, thinking that they are still authorised, may wrongly or inadvertently fail to change the declared head counts when their actual pig populations are lower than the authorised counts. As part of the controls set up for meeting the new European "animal welfare" standards, such as group pens for gilts and sows, at least holdings that have or had farrowing activity, will be updated in the database. Moreover, the holding registry cannot be used for farmed wild boar holdings or outdoor holdings, even though these holdings are targeted for routine serological testing for AD. Population counts of farmed wild boars are therefore probably currently underestimated in the BDPORC database on which this report is based. Finally, it is important to note that 37 of 95 *départements* (mainland France and Reunion Island) have not validated their data in the database for the 2012 report. There was thus missing data and obsolete data. Daily data transmission between BDPORC and SIGAL in late 2012 should help the departmental services in charge of population protection (DDecPP) ensure that data are synchronised and updated throughout the year.

In 2012, there was a slight increase in the number of farrowing and wean-to-grow holdings tested, a strong decrease in the number of grow-to-finish holdings tested and a decrease in the number of farrow-to-finish holdings tested. These changes, although difficult to explain, may have arisen due to the special attention governmental services are giving to holdings that must comply with the new «animal welfare» regulations, i.e. holdings with breeding pigs. The coverage rate of outdoor holdings (domestic swine) was lower than 50%, representing a 10% decrease compared to 2011, although these holdings are the most at risk. This decrease can be attributed at least in part to *départements* for which data could not be used in 2012. A decrease in the number of breeding holdings analysed was also observed, probably due to the decrease in population counts in this holding category. For this type of holding, the number of samples per holding and per year remained stable between 2011 and 2012, and the average number of samples per holding and per quarter increased compared with 2011. It is also surprising that indoor holdings in some *départements* continue to be subjected to routine testing even though this type of holding is not

targeted by the mandatory active surveillance programme (because these holdings are considered to be at a lower risk of introduction or spread of the virus).

Annual serological surveillance in outdoor holdings, particularly in farrowing holdings, should help compensate for the limits of passive surveillance. It is now necessary to ensure that serological testing is effectively carried out, because the 11 serological tests performed on average only ensure the detection of a seroprevalence level of at least 30%, whereas seroprevalence levels that can occur in outdoor holdings are lower (routine tests include 15 samples to target a seroprevalence of 20% with a 5% error rate).

Of the 14 outdoor holdings that showed positive serological results, all required a second round of blood 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, particularly when an outbreak is suspected, to rapidly confirm or refute the presence of an outbreak of AD. Although the new continuing vocational training course for state veterinarians on taking blood samples from pigs could not be set up in 2011 due to technical constraints (unsuitable material, difficulty in finding animals of different ages for the different samples to be taken, etc.), initiatives taken by local DDecPPs have combined training on regulated pig diseases with a practical course on taking blood samples. This initiative should be extended in the future to all training sessions of this type.

All players in the pig industry must remain vigilant, because this is the only way to ensure early detection of an outbreak. Only one clinical suspicion was notified in 2012 (a decrease compared to 2011 (1 suspected case) and 2010 (4 suspected cases)), but some veterinarians requested exclusion tests for AD. This new approach for suspected cases should be pursued, and all veterinarians should be encouraged to include AD in their confirmatory testing when flu-like symptoms and abortions cannot be attributed to another disease with certainty. Exclusion tests can effectively facilitate the notification of suspected cases by decreasing the consequences for the holding affected. However, currently, this test and its results are not always reported, and therefore actual clinical surveillance activities are not clearly or fully described. It is therefore necessary to improve the tools used for monitoring and communicating epidemiological information. It is also important to stress that outdoor holdings run the highest risk of AD. It is fundamental therefore that routine testing be carried out on all outdoor holdings, and on at least 15 pigs per holding to detect any infection as quickly and as close to the source as possible.

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Box. Aujeszky's disease surveillance and control measures

Objectives of the surveillance programme

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

Monitored population

Domestic swine and farmed wild boars throughout mainland France.

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). In the case of suspicion, the DDecPP must be notified and sampling must be carried out for serological and virological testing.

- 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 breeding holdings).

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

In breeding holdings: quarterly surveillance (15 samples per quarter for serological analyses).

Holdings for which AD-free status has been revoked or suspended for administrative reasons (due to absence of routine testing 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.

Animal health rules (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. 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 survey results.

Serological suspicion is based on non-negative serological results. Any animal 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 serological tests. The holding is placed under APMS when an accredited laboratory produces a positive or ambiguous result in any individual test. When only one or two samples are positive or inconclusive, APMS can be "relaxed", allowing movements of pigs to slaughter or to authorised terminal holding pens, providing that the holding (of origin) with serological suspicion has been clinically and epidemiologically sanctioned to do so and that the destination holding or slaughterhouse has agreed in writing that pigs can be introduced from a holding with serological suspicion for AD and that the destination holding is also placed under APMS.

Box. Aujeszky's disease surveillance and control measures

An animal is considered infected by AD when, even in the absence of any suggestive symptoms 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.

Regulations

- Directive 90/429/EEC laying down the animal health requirements applicable to intra-Community trade in and imports of semen of domestic animals of the porcine species
- Decision 2008/185/EEC amended of 21 February 2008 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

Review of classical and African swine fever surveillance in France in 2012

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Abstract

As in previous years, vigilance with respect to Classical Swine Fever in 2012 was based on serological surveillance at the slaughterhouse and in breeding holdings, as well as on outbreak surveillance. Surveillance of wild boars in the north-eastern part of France was maintained in 2012. Vigilance with respect to African Swine Fever was based on outbreak surveillance. This outbreak surveillance led to only one clinical suspicion being reported in 2012. The risks posed by Classical Swine Fever and African Swine Fever are indeed still present, highlighting the need to raise the awareness of professionals in the field. This latter disease emerged in Belarus in June 2013 after being reported in Ukraine in August 2012. In 2012, outbreaks of Classical Swine Fever were reported in Hungary, Latvia and Russia and positive serological results were detected in wild boars in Croatia in 2012 and 2013.

Keywords

Regulated disease, CSF, ASF, epidemiological surveillance, swine, France

Résumé

Bilan de la vigilance à l'égard des pestes porcines classique et africaine en France métropolitaine et d'Outre-mer en 2012

Comme les années précédentes, la vigilance à l'égard de la peste porcine classique a reposé en 2012 sur une surveillance sérologique à l'abattoir et dans les élevages de sélection-multiplication, et sur une surveillance événementielle. La surveillance des sangliers dans l'Est de la France a été poursuivie en 2012. La vigilance à l'égard de la peste porcine africaine a reposé sur une surveillance événementielle. En 2012, la surveillance événementielle a conduit à la notification d'une seule suspicion clinique. Les risques présentés par la peste porcine classique et par la peste porcine africaine sont bien présents et la vigilance des acteurs de terrain doit être renforcée. Cette dernière maladie continue de sévir en Russie et en Sardaigne. Elle a fait son apparition en Biélorussie en juin 2013 après avoir également touché l'Ukraine en août 2012. En 2012, des foyers de peste porcine classique ont été notifiés en Hongrie, Lettonie et Russie, et des cas de séropositivité chez des sangliers ont également été détectés en 2012 et 2013 en Croatie.

Mots clés

maladie réglementée, PPC, PPA, épidémiosurveillance, suidés, France

Here, we present the results of the classical and African swine fever surveillance programme for 2012 (see Box for details on the surveillance programme). Of the 101 *départements* in mainland France and overseas, 84 *départements* answered the questionnaires, at least in part, that were sent to them.

Slaughterhouse surveillance

The main results from surveillance at slaughterhouses (Table 1) show that:

- 10,210 pigs were targeted nationally for serological testing (ELISA assays); 7,992 breeding pigs and 1,431 slaughter pigs were actually sampled (i.e. a 92% coverage rate – a conservative estimate based on the initial target, without removing *départements* that did not provide data). A total of 1,212 pig holdings were screened, with an average of eight samples per site.
- 3,000 samples were targeted nationally for virological (PCR) tests; 2,938 blood samples (from 1699 breeding pigs and 1,239 slaughter pigs) were actually sampled (98% coverage rate – conservative estimate based on the initial target, without removing *départements* that did not provide data). A total of 565 holdings were screened, with an average of five samples per site.

Table 1. Results of the classical swine fever slaughterhouse surveillance programme for 2012

	Serological tests (ELISA)	Virological (PCR) tests
Target number of samples	10,210	3,000
Actual number of samples	9,423	2,938
Coverage rate	92%	98%
Number of pig holdings sampled	1,212	565
Average number of pigs tested per holding	8	5
Proportion of culled breeding pigs tested	2.4%	0.8%

In total, 2.5% (7,992 serological tests and 1,699 virological tests) of culled breeding pigs were serologically or virologically tested as part of slaughterhouse surveillance, a rate comparable to that observed in 2011 (i.e. 2.4% culled breeding pigs tested).

Due to the proximity of infected countries such as Madagascar, the Reunion Island DDecPP set up a serological testing programme for African swine fever (ASF) at the slaughterhouse on the 250 samples taken as part of routine testing under the active surveillance programme for classical swine fever (CSF).

Surveillance in breeding holdings

Regarding surveillance in breeding holdings, 5,804 samples were taken in 2012 in 459 holdings, representing an average annual testing rate of 7.2% of the breeding pigs in these holdings.

On average, 13 samples were taken per holding and per year, slightly lower than the number taken in 2011 (n = 16).

Overall, serological CSF surveillance in France (in slaughterhouses and in holdings) covered 16.4% of all breeding pigs (i.e. 7,992 sampled at slaughterhouses and 5804 in holdings), compared to 15.5% and 18.8% in 2011 and 2010, respectively.

Results

For a total of 15,227 serological samples taken to test for CSF, 152 showed non-negative results, 136 of which were from breeding holdings (15 pig holdings) and 16 from slaughterhouse screening (animals from 2 pig holdings, based on available information).

The samples that gave positive results were sent to the NRL and none of these positive results were confirmed.

These 152 non-negative first-line serological test results represent a false positive rate of 1%, compared with 0.34% in 2011.

Regarding ASF testing, 83 serological samples taken in Reunion Island were sent to the NRL for analysis due to technical difficulties with a batch of ELISA kits; all sera were confirmed to be negative.

Regarding CSF virological testing (at slaughterhouses), no positive reactions were detected, demonstrating the high specificity of this PCR test.

Wildlife surveillance

Wildlife surveillance in north-eastern France (Bas-Rhin and Moselle *départements*) (Rossi *et al.*, 2011) was conducted on 10,301 wild boars in 2012 (with 90% sampled in the high surveillance zone and 10% in the original surveillance zone near Thionville), including more than half of the boars hunted during the fourth quarter. In total, 10,491 virological tests and 10,440 serological tests were performed. Of these, 671 were seropositive, with 668 observed in the former vaccination zone. The three seropositive samples found in the original surveillance zone were tested using the specific virus neutralisation test. The results of these additional analyses confirmed the absence of antibodies against the CSF virus in the tested sera. Eight first-line virological analyses tested positive, but none were confirmed by the NRL.

Clinical suspicions

In 2012, there was one pig holding with a clinical suspicion. This suspicion was not confirmed either for CSF or for ASF.

Cost

In 2012, for the 79 *départements* for which data are available, the French government invested more than €131,500 in the surveillance and control of CSF and ASF. Laboratory costs amounted to €126,348 for test-ing (preventive measures) and €5,148 for control measures. The veterinary costs incurred for control meas-ures were €38. These figures do not include government funds spent for testing in breeding holdings that belong to the French pig breeding agency (ASP), i.e. €31,006 for the serological analyses. French gov-ernment funds of €1.4m were invested in wildlife surveillance.

Discussion

As in previous years, the results of the CSF and ASF surveillance programme for 2012 demonstrate that France has maintained its disease-free status.

Serological and virological surveillance in slaughterhouses involved more holdings in 2012 than in 2011 (Marcé *et al.*, 2012), while the average number of samples taken per site was stable. Regarding serological CSF surveillance, the number of samples taken increased in breeding pigs and decreased slightly in slaughter pigs, in accordance with the two main objectives of this surveillance programme. Blood samples taken at the slaughterhouse aim (1) to provide information fundamental to confirming France's disease-free status and to provide proof to the European Community and international authorities that France is free of CSF and ASF; (2) to maintain the operational capacity of the network of serological and virological laboratories accredited for CSF diagnosis (16 laboratories accredited for serological CSF tests and 8 laboratories accredited for virological CSF tests), so as to be able to respond efficiently in case of an epizootic. In 2012, two inter-laboratory proficiency tests (ILPTs) were organised, one for serological assays (ELISA and virus neutralisation tests) and one for virological (PCR) tests. Both showed satisfactory results for all the accredited laboratories.

Ideally, breeding pigs reflect the health status of the entire herd, due to their long presence in the holding -- much longer than that of slaughter pigs – making them a target of choice for meeting the first

surveillance goal. The age of the pig is not a limiting factor for the second goal. Nonetheless, due to the difficulties in procuring breeding pigs to sample at the slaughterhouse, because some slaughterhouses that processed this type of pig have closed down or because, in certain *départements*, slaughterhouses have decided to refocus their activity on slaughter pigs, exceptions were allowed such as those defined in the DGAL Memorandum DGAL/SDSPA/N2006-8033 of 7 February 2006 as amended, when samples could not be taken on breeding pigs. In these cases, samples were taken on slaughter pigs. Regarding sampling for virological tests, the total number of samples taken increased in 2012 and was very close to the target number (2,938 samples of the 3,000 targeted). There was a decrease in samples taken on breeding pigs, and a large increase in the number of samples taken on slaughter pigs. For this part of the surveillance programme, better coverage favours the second objective and helps maintain testing laboratories' capacity for diagnosis.

In breeding holdings, serological surveillance involved a fewer number of breeding pigs in 2012 compared to 2011, but the number of sites sampled was higher. The number of samples per holding decreased compared to 2011 and 2010. This surveillance using serology assays is a safeguard for the disease-free status for France for CSF and ASF, which can be detected at a prevalence threshold of 0.05% with a 95% confidence level.

In parallel, passive surveillance identified one case of clinical suspicion (2 were identified in 2011, 4 in 2010 and none in 2009). This demonstrates that the pig and pork industry is maintaining its vigilance, a commendable practice that should be encouraged. This vigilance is even more important in light of the existence of a low-virulence CSF virus strain that can lead to attenuated clinical signs; furthermore, the CSF virus is still present in some wild boar populations in Europe, and the ASF virus continues to affect areas just outside of Europe (Sánchez-Vizcaíno *et al.*, 2012 Oganesyan *et al,* 2013) and in Sardinia. ASF has appeared in the Caucasus where it is now enzootic. After ASF outbreaks reported in Russia, Ukraine was affected between August and December 2012. Belarus declared for the first time that it detected an ASF outbreak in June 2013. In addition, a resurgence of ASF cases was observed in Sardinia in late 2011 and early 2012 (Le Potier and Marcé, 2013).

Of the 17 holdings that fell under serological suspicion for CSF, only one was placed under a prefectural monitoring order (APMS). Management measures implemented in "suspicious" holdings should be adjusted considering the favourable disease status in France and the actual risk of introduction. In 2012, regulations introduced the concepts of "high" and "low" serological suspicion and any case of serological suspicion must be placed under APMS (see Box), with different restrictions on animal movements, depending on the situation. The one holding for which a case of clinical suspicion was reported in 2012 was placed under APMS.

In January 2012, due to the favourable trend in the situation in wildlife in north-eastern France (no new observed cases and a decrease in seroprevalence), the high level of surveillance was lifted in the former observation zone located around the infected zone. Surveillance measures continue in the former infected zone in the northern Vosges, which has now become a high observation zone (ZOR), and in the surveillance zone due to the observation of young seropositive wild boars. The presence of antibodies in wild boars born after 2010, which therefore could not have been vaccinated (because the emergency vaccination plan ended in June 2010), raises the issue of potential persistence of the CSF virus in these two zones. The analysis conducted on these 2012 data suggests sporadic dissemination of seropositive animals from the vaccinated zone to the surveillance zone and an absence of virus circulation in the high observation zone. (ZOR) The possibility of low-frequency virus circulation in the ZOR after the end of the vaccination programme could not be excluded based on the data analysis. Despite the overall decrease in seroprevalence, certain areas still showed seroprevalences greater than 10% in young wild boars. The detection of antibodies in young wild boars suggests that:

- either some young boars still show maternal antibodies at up to 5 to 10 months, although when raised in captivity, young boars generally lose their maternal antibodies by 3 months of age (this may be the result of intense vaccination in females which were exposed to several vaccination campaigns between 2004 and 2010),
- or the CSF virus continues to circulate at low frequencies in the northern Vosges mountain range with higher dynamics in the centre of this geographic area.

Unfortunately, it is impossible to determine whether the antibodies are of maternal origin from samples acquired from hunting campaigns. A capture-recapture study was initiated in 2013 to better understand the origin of the antibodies in this age class (i.e. young boars).

One of the medium-term goals of the CSF/ASF surveillance programme is to redefine the slaughterhouse surveillance plan, given the expected levels of prevalence in pig holdings for low-virulence CSF strains (which are not easily detected clinically), estimated using a model developed by the ANSES Ploufragan Laboratory (personal communication). Meanwhile, the whole pig and pork industry is encouraged to maintain their vigilance with respect to swine fevers and promote passive surveillance, thereby guarding against the spread of CSF or ASF in the event of their introduction.

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Objectives of the surveillance programme
- To detect as early as possible any appearance of an outbreak in domestic swine.
- To provide evidence that France is free of CSF and ASF.
- To maintain the operational capacity of the network of serological and virological laboratories accredited for CSF diagnosis so as to provide an efficient response in case of an epizootic.

Monitored population
Domestic pigs and farmed wild boars throughout mainland France and its overseas *départements*, and wild boars in north-eastern France.

Surveillance procedures
- Outbreak surveillance

Passive surveillance targets both CSF and ASF and is based on the principle that any person (veterinarian, farmer, animal trader, hunter, etc.) suspecting a case of CSF or ASF must notify the DDecPP.
- Programmed surveillance

Programmed surveillance is carried out in slaughterhouses and in holdings (only in breeding holdings).
In slaughterhouses, random serological and virological tests are carried out on slaughtered breeding pigs throughout France:
> *in serological tests, 10,000 samples should be tested annually to detect a prevalence rate of 0.05% (at a 99% confidence level, providing that sampling is random), and thus attest to the disease-free status of mainland France;*
> *in virological tests, 3,000 samples should be tested to detect a prevalence of at least 0.1% (with a 95% confidence level); given that viraemia is short-lived (2-3 weeks at most), these samples are used first and foremost to maintain the technical skills in the network of accredited PCR laboratories.*

In breeding holdings (in which the spread of CSF/ASF is potentially high), annual testing is carried out in each holding: 15 samples for serological tests (for *a minima*l within-holding prevalence rate of 20% with a 95% confidence level).
- CSF surveillance in wild boars in north-eastern France

As France recovered its disease-free status for wild boars on 14 November 2011, surveillance has been restricted to a smaller area since 1 January 2012 (DGAL/SDPSA/N2011-8283). Any wild boar hunted or found dead must be sampled for virological analysis (PCR) and a blood sample for ELISA analysis must be taken in a vacutainer blood collection tube. This surveillance will be relaxed even further and restricted to a smaller, specific area during the 2013-2014 hunting season.

Definition of suspected cases and confirmed cases
Swine "suspected to be infected with swine fever": any swine showing symptoms and/or *post mortem* lesions suggestive of swine fever that cannot be attributed with certainty to any other disease or showing non-negative first-line test results.
Swine "suspected to be contaminated": any swine likely, according to epidemiological information, to have been exposed directly or indirectly to a swine fever virus.
A holding is suspect when it holds at least one suspect animal or when it has an epidemiological connection with a confirmed outbreak.

Distinction between low serological suspicion and high serological suspicion
An outbreak of swine fever must be notified when a holding meets one or more of the following criteria:
1. CSF or ASF virus isolated in an animal or any derived product thereof;
2. clinical signs suggestive of swine fever observed in an animal and viral antigen or genome (RNA, for CSF; DNA for ASF) detected and identified in samples taken from an individual pig or a cohort;
3. clinical signs suggestive of swine fever observed in an individual of a susceptible animal species and the animal or members of its cohort show specific antibodies against CSF or ASF viral proteins;
4. CSF or ASF viral antigen or genome observed and identified in samples taken from swine and the swine show specific antibodies against CSF or ASF viral proteins;
5. clear epidemiological connection with the appearance of a confirmed swine fever outbreak and when at least one of the following conditions is met:
a) at least one individual shows specific antibodies against CSF or ASF viral proteins,
b) the antigen or the CSF or ASF viral genome is detected and identified in samples taken from at least one individual of a susceptible species.

Animal health rules
CSF and ASF are Category 1 health hazards, notifiable diseases, regulated and subject to emergency response plans.
When an accredited laboratory announces that one or more individual serological tests resulted in positive or ambiguous results, the holding is placed under APMS surveillance. There are two levels of suspicion, defined since February 2012.
If only one or two samples are positive or ambiguous and there are no suspicious clinical signs or unfavourable epidemiological conditions, suspicion is low and the APMS is adapted to this less threatening situation: movements are allowed providing that the holding (of origin) with serological suspicion has been visited and clinically and epidemiologically sanctioned and that the destination holding or slaughterhouse has agreed in writing that animals can be introduced from a holding with serological suspicion, and that the destination holding is also placed under APMS. Culled animals are consigned until there are results disproving the suspicion.
In the case of high CSF or ASF suspicion based on clinical signs or epidemiological connections, an APMS is ordered and no exceptions are possible for the movement of animals. When infection is confirmed, the holding is placed under APDI. All swine are culled immediately, the carcasses are disposed of, the holding is disinfected, and all animal and animal by-products are disposed of. Repopulation cannot take place until after 30 days. This period is longer in the case of ASF infection if the intermediate host (i.e. Ornithodoros ticks) is likely to have been involved.
In holdings with an epidemiological connection (contact holdings), conservative measures are taken under APMS and call for enhanced surveillance.
In the vicinity of the outbreak, a protection zone with a radius of 3 km is established as well as a surveillance zone with a radius of 10 km within which surveillance, movements and possible exceptions are not as strict as within the protection zone. The measures specific to these zones are available in the Memorandum DGAL/SDSPA/N2006-8194 as amended on the swine fever contingency plan.

Regulations
- Directive 2001/89/EC on Community measures for the control of classical swine fever
- Directive 2002/60/EC laying down specific provisions for the control of African swine fever
- Decision 2008/855/EC concerning animal health control measures relating to classical swine fever in certain Member States
- Decision 2004/832/EC approving the plans for the eradication of classical swine fever in feral pigs and the emergency vaccination of such pigs in the Northern Vosges, France
- Decision 2002/106/EC approving a Diagnostic Manual establishing diagnostic procedures, sampling methods and criteria for evaluation of the laboratory tests for the confirmation of classical swine fever
- Ministerial Order of 23 June 2003 laying down the measures for the control of classical swine fever
- Ministerial Order of 11 September laying down the measures for the control of African swine fever

Update on the surveillance of avian *influenza* and Newcastle disease in France in 2012

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Abstract
France maintained its status in 2012 as "free from high and low pathogenic avian *influenza*" and "free from Newcastle disease" according to the OIE Animal Health Code. The active surveillance in poultry holdings revealed, as in other years, H5 seropositive waterfowl holdings without detecting any virus. The year 2012 was marked by a decrease in the number of suspicions. The evaluation in 2012 of the avian *influenza* and Newcastle disease surveillance system by the OASIS method within the framework of the activities of the French surveillance platform for animal health (ESA Platform) has led to a number of recommendations designed to increase the efficiency of this surveillance programme.

Keywords
Avian *influenza*, Newcastle disease, Paramyxovirosis, pigeon, poultry, birds, France

Résumé
Bilan de la surveillance de l'influenza aviaire et de la maladie de Newcastle en france en 2012
La France a conservé en 2012 son statut indemne vis-à-vis de l'influenza aviaire hautement et faiblement pathogène et de la maladie de Newcastle au sens du code sanitaire de l'OIE. La surveillance programmée en élevage a mis en évidence, comme les autres années, des lots séropositifs H5 au sein d'élevages de palmipèdes, sans pour autant mettre en évidence de virus. L'année 2012 a été marquée un nombre de suspicions en chute libre pour la surveillance événementielle. L'évaluation menée en 2012 du dispositif de surveillance des pestes aviaires par la méthode OASIS dans le cadre des activités de la Plateforme nationale d'épidémiosurveillance en santé animale (Plateforme ESA) a débouché sur un certain nombre de recommandations visant augmenter l'efficience de ce dispositif.

Mots clés
influenza *aviaire, maladie de Newcastle, paramyxovirose pigeon, volailles, oiseaux, France*

Here, we present the results from the avian *influenza* (AI) and Newcastle disease (ND) surveillance programme in 2012 as well as the main 2012-2013 changes and activities. An overview of the surveillance programme is given in Boxes 1 and 2.

Active surveillance for avian *influenza* in poultry holdings

Objectives and design of surveillance programme: targeting at-risk populations (Box 1)

As in previous years, surveillance consisted in taking serological samples to test for infection by AI virus subtypes H5 and H7 in selected poultry farms throughout French *départements*, in accordance with the recommendations made in European Commission Decision 2010/367/EU of 25 June 2010.

This surveillance programme targets holdings with certain risk factors: possible contamination by wildlife in outdoor holdings, or holdings located near wetlands or areas where wild birds may gather, and those with more susceptible species (e.g. turkeys in particular). Sampling preferentially targets *départements* with a high density of poultry holdings (Figures 1 and 2).

The targeted species included gallinaceans (chicken, turkey, guinea fowl, pheasant, partridge and quail), ratites and palmipeds.

Farmed game birds (pheasants, partridges, mallard ducks) and palmipeds are the poultry production categories that have shown the highest serological prevalence in previous surveys and therefore must be specifically targeted, as well as holdings in contact with them.

Results

Number of holdings: Out of the 922 targeted holdings (other than ratite holdings), 898 were sampled. Seventeen were found to be seropositive for H5 (12 breeder duck holdings, 3 ready-for-gavage duck holdings and 2 breeder geese holdings), three batches were deemed to be ambiguous for H5 (2 ready-for-gavage duck holdings and 1 breeder duck holding) and one batch was seropositive for H5 and ambiguous for H7.

For these 21 batches with positive and/or ambiguous results, 14 were no longer present in the holdings of origin and 7 were sampled (swabs) for PCR testing, all of which gave negative results.

These 21 seropositive and/or ambiguous batches involved seven *départements* (Vendée, Maine-et-Loire, Loire-Atlantique, Deux-Sèvres, Landes, Gers and Indre-et-Loire).

Categories of holdings: The categories for which the coverage rate was less than 80% of the target announced in Memorandum DGAL/SDSPA/N2012-8134 of 27 June 2012 were breeder quails (68%) and (non-accredited) slaughterhouses (77%) — the same as in 2011 — and, in 2012, caged laying hens (78%). Only four ratite holdings were sampled nationally.

Based on the population counts available in SIGAL, it was not always possible to adapt the sampling plan to the actual population counts and holdings found in each *département*.

Time to results: An improvement is necessary in regard to transmission of the samples to the accredited laboratories: the average time is 28 calendar days (calculated from available information). However, the interval between when laboratory reports are sent for seropositive cases and the return visit to the holding of origin is relatively short,

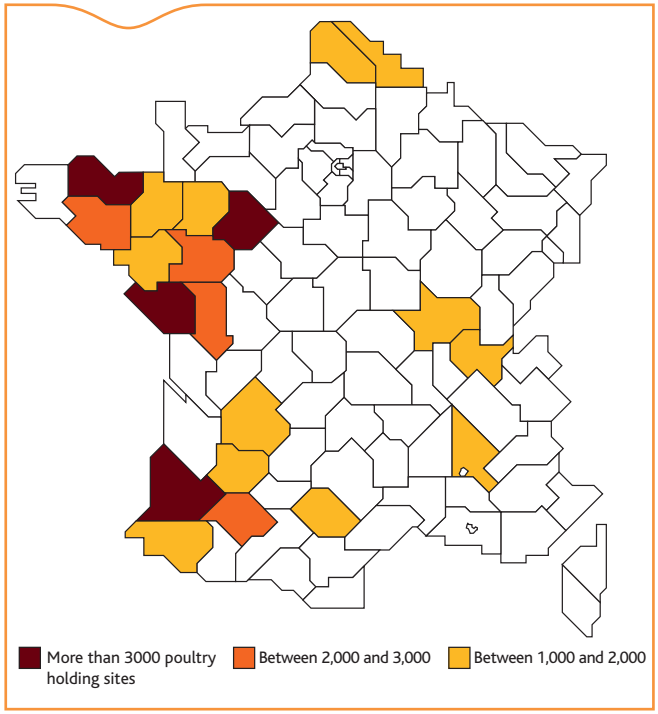

Figure 1. Distribution of poultry holdings (all production categories) in France registered in the national database

Source: SIGAL database

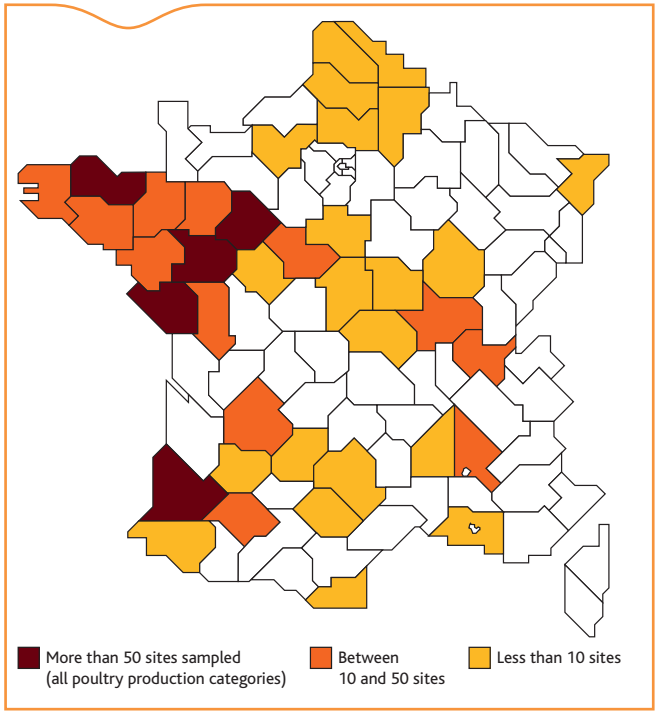

Figure 2. Distribution of poultry holdings in France sampled for the annual serological testing

Source: SIGAL database

Table 1. Review of avian *influenza* surveillance in holdings – 2012

	Data extracted from SIGAL on 13-12-2012				Data from the NRL				Additional analyses: molecular analyses according to results reported to the NRL on 21-12-2011	
	No. batches	No. holdings	Target no. holdings (see Memorandum DGAL/SDSPA/ N2012-8134)	Coverage rate (farms sampled compared to target)	No. batches sent to the NRL	No. AI positive holdings (IDG)	No. H7 seropositive holdings	Nombre d'élevages H7 séropositifs	No. retested batches	No. positive batches[c]
Breeder quail	16	15	22	68%	0	/	0	0	/	/
Broiler duck	77	76	90	84%	1	/	0	0	/	/
Mallard duck	18	18	20	90%	0	/	0	0	/	/
Multiplier and breeder duck	76	72	80	90%	17	/	13[a] + 1 ambiguous	1 ambiguous[a]	3/14	0/3
Muscovy multiplier (≤ 24 weeks)	*11*	*11*	/	/	*0*	/	*0*	*0*	/	/
Muscovy breeder	*31*	*28*	/	/	*5*	/	*3*	*0*	*1/3*	*0/1*
Peking multiplier (≤ 18 weeks)	*6*	*6*	/	/	*0*	/	*0*	*0*	/	/
Peking breeder	*28*	*27*	/	/	*12*	/	*10[a] + 1 ambiguous*	*1 ambiguous[a]*	*2/11*	*0/2*
Ready-for-gavage duck	93	93	90	103%	7	/	3 + 2 ambiguous	0	2/5	0/2
Fattening turkey	69	69	60	115%	0	0	/	/	/	/
Free-range turkey	58	58	60	97%	0	0	/	/	/	/
Breeder turkey	65	49	60	82%	0	0	/	/	/	/
Pheasant	37	37	30	123%	3	/	0	0	/	/
Multiplier and breeder goose	16	16	20	80%	4	/	2	0	2/2	0/2
multiplier goose (≤ 24 weeks)	*2*	*2*	/	/	*0*	/	*0*	*0*	/	/
breeder goose	*14*	*14*	/	/	*4*	/	*2*	*0*	*2/2*	*0/2*
Partridge	28	28	30	93%	0	/	0	0	/	/
Guinea fowl	56	56	60	93%	6	/	0	0	/	/
Laying hen	48	47	60	78%	0	0	/	/	/	/
Free-range laying hen	67	67	60	112%	0	0	/	/	/	/
Breeder hen	61	60	60	100%	1	1	0b	0	/	/
Free-range broiler	91	91	60	152%	1	1	0	0	/	/
Slaughterhouse	46	46	60	77%	0	0	/	/	/	/
Ratite	4	4	91	4%	0	/	0	0	/	/
Other[d]	9	9		/	0	0	/	/	/	/
Total	**926**	**902**	**1,013**	**89%**	**40**	**2**	**18[a] + 3 ambiguous**	**1 ambiguous[a]**	**7/21**	**0/7**

/ : not applicable

a 1 batch seropositive for H5 and ambiguous for H7

b 1 batch inconclusive for H5, due to insufficient serum; holding was visited a second time and an epidemiological survey was carried out (culled batch).

c tested directly with rRT-PCR for the H5 gene

d 9 batches, for which the production category was not specified, were removed from the final count

with an average of 10.9 calendar days. Therefore, the local veterinary services (DDecPPs) show good response times.

Table 1 gives a summary of the main results.

Comparison with previous years

Table 2 gives an overview of the seropositive results for H5 for the last three annual surveys in holdings, indicating the proportion of H5 positives per species and per year, as well as the 95% confidence interval according to a normal distribution or a binomial distribution, depending on statistical test (according to sample size). For each type of poultry production category, the confidence intervals show overlapping values for the three surveyed years. There are therefore no significant differences in seropositivity among the last three serological surveys.

Outbreak surveillance in holdings

Procedures (Boxes 1 and 2)

Passive surveillance entails the notification of clinical suspicions of AI or Newcastle disease (ND) in a holding.

As stipulated in Article L201-7 of the French Rural Code, all poultry farmers/breeders and veterinarians are required to notify the DDecPP of any sign suggestive of AI or ND.

For AI, the alert criteria in flocks of more than 1000 individuals are listed in Annex 3 of the Ministerial Order of 24 January 2008. These criteria give the thresholds of mortality or decrease in water and food consumption and drop in egg-laying below which the poultry farmer must notify his/her state veterinarian. The veterinarian must determine the causes of these symptoms and, in case of a suspected AI case, must notify the DDecPP immediately.

Suspicions of AI lead to sampling (tracheal and cloacal swabs) to screen for the AI virus genome using RT-PCR targeting the M gene. If the test results are positive, H5- and H7-specific PCR tests are carried out respectively at an accredited laboratory and at the NRL. Suspicions of Newcastle disease and/or pigeon paramyxovirosis are tested by screening for and isolating the incriminating virus (LVD01 and LDA22) from organs.

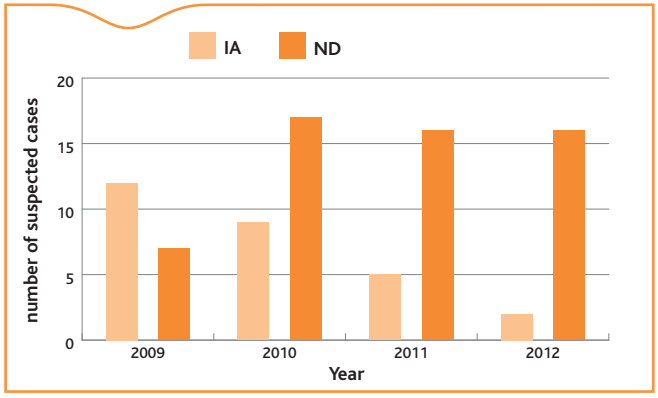

Figure 3. Annual number of cases of clinical suspicions of avian *influenza* (AI) and Newcastle disease (ND) between 2009 and 2012.

Results

Only two cases of clinical suspicion of AI were reported in holdings in 2012: one suspicion of a low pathogenic AI (LPAI) in breeder hens and one case in ducks. There were 16 reported cases of clinical suspicions of ND, and 6 of these revealed a pigeon paramyxovirus serotype 1 (PPMV-1) in sites with captive show pigeons held by private individuals.

These passive surveillance results underscore two important points: (1) passive surveillance needs to be reactivated and (2) poultry owners need to be reminded that vaccination against Newcastle disease is mandatory in pigeons.

Reactivation of outbreak surveillance

The ever-decreasing number of clinical suspicions of AI (Figure 3) is a sign of a flagging passive surveillance programme. Memorandum DGAL/SDSPA/N2013-8047 of 27 February 2013, and the recent appearance of a highly pathogenic H7N7 AI strain in Italy reiterate that symptoms suggestive of AI must be reported and vigilance should be redoubled in holdings.

Nevertheless, this ever-decreasing number of clinical suspicions of AI also demonstrates that reporting an official suspicion of AI, particularly LPAI, may be difficult because it leads to an APMS monitoring order, and

Table 2. Comparison of results from the surveillance campaigns in 2010, 2011 and 2012

	2012			2011			2010		
	No. holdings	No. H5 seropositive holdings	percentage of H5 positive holdings [95% CI]	No. holdings	No. H5 seropositive holdings	percentage of H5 positive holdings [95% CI]	No. holdings	No. H5 seropositive holdings	percentage of H5 positive holdings [95% CI]
Breeder quail	15	0	0% [0.0-21.8]	12	0	0% [0.0-26.5]	12	0	0% [0.0-26.5]
Broiler duck	76	0	0% [0.0-4.7]	74	0	0% [0.0-4.9]	74	2	2.7% [0.3-9.4]
Mallard duck	18	0	0% [0.0-18.5]	18	1	5.6% [0.1-27.3]	20	3 + 1 ambiguous	20.0% [5.7-43.7]
Multiplier and breeder duck	72	13[a] + 1 ambiguous	19.4% [10.3-28.6]	72	17 + 2 ambiguous	26.4% [16.2-36.6]	85	18[c]	21.2% [12.5-29.9]
Ready-for-gavage duck	93	3 + 2 ambiguous	5.4% [1.8-12.1]	91	6	6.6% [2.5-13.8]	85	1 ambiguous	1.2% [0.0-6.4]
Fattening turkey	69	/	0% [0.0-5.2]	65	0	0% [0.0-5.5]	77	0	0% [0.0-4.7]
Free-range turkey	58	/	0% [0.0-6.2]	59	0	0% [0.0-6.1]	78	0	0% [0.0-4.6]
Breeder turkey	49	/	0% [0.0-7.3]	41	0	0% [0.0-8.6]	83	0	0% [0.0-4.4]
Pheasant	37	0	0% [0.0-9.5]	39	0	0% [0.0-9.0]	48	1	2.1% [0.1-11.1]
Multiplier and breeder goose	16	2	12.5% [1.6-38.6]	19	2	10.5% [1.3-33.1]	13	6	46.2% [19.2-74.9]
Partridge	28	0	0% [0.0-12.3]	29	0	0% [0.0-11.9]	41	0	0% [0.0-8.6]
Guinea fowl	56	0	0% [0.0-6.4]	54	0	0% [0.0-6.6]	60	1	1.7% [0.0-8.9]
Laying hen	47	/	0% [0.0-7.6]	51	0	0% [0.0-7.0]	54	0	0% [0.0-6.6]
Free-range laying hen	67	/	0% [0.0-5.4]	66	0	0% [0.0-5.4]	/	/	/
Breeder hen	60	0[b]	0% [0.0-6.0]	56	0	0% [0.0-6.4]	/	/	/
Free-range broiler	91	0	0% [0.0-4.0]	63	0	0% [0.0-5.7]	133	0	0% [0.0-3.59]
Slaughterhouse	46	/	0% [0.0-7.7]	47	0	0% [0.0-7.6]	96	0	0% [0.0-3.8]
Ratite	4	0	0% [0.0-60.2]	4	0	0% [0.0-60.2]	0	0	/
Total	**902**	**18[a et b] + 3 ambiguous**		**860**	**26 + 2 ambiguous**		**959**	**31[c] + 2 ambiguous**	

a 1 batch both seropositive for H5 and ambiguous for H7

b 1 batch with inconclusive results

CI = confidence interval

The 95% confidence intervals were calculated for a normal distribution or a binomial distribution, according to the statistical test applied (i.e. depending on sample size)

The ambiguous batches are considered to be positive.

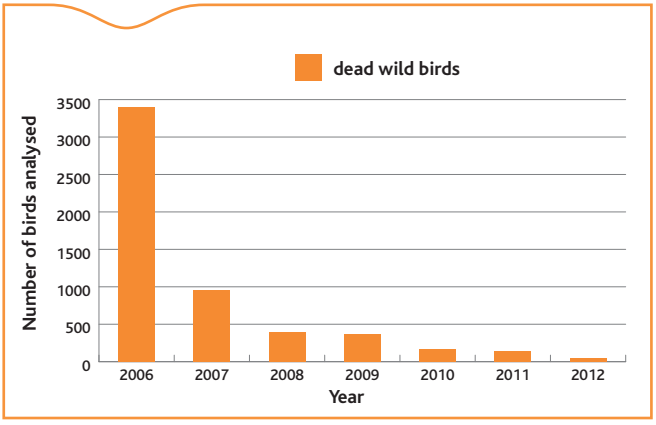

Figure 4. Annual number of wild birds found dead and screened for the AI virus using PCR.

to movement restrictions on the holding while waiting for test results. In the case of Newcastle disease, these movement restrictions are even longer because the diagnosis technique requires virus isolation, which is a long process.

This opens the debate on the possibility of conducting a diagnosis of exclusion and to be able to classify a suspicion (low, medium, high). A low suspicion would require AI and/or ND tests by state veterinarians to exclude these diseases, without leading to an APMS order. High suspicion would entail emergency testing procedures.

Pigeon ND vaccination programme: authorisation of the use of IMOPEST in pigeons

Vaccination against ND is mandatory in domestic pigeons as stipulated in Article 24.3 of the Ministerial Order of 8 June 1994 and for all categories of pigeon (sport, breeding, show, food, etc.).

Previously there were three ND vaccines authorised for pigeons: NOBILIS PARAMYXO P201® (Intervet), COLOMBOVAC PMV/Pox® (Pfizer), COLOMBOVAC PMV® (Pfizer).

Using other vaccines under the cascade approach is not authorised because there are products available with an MA indicated for use in pigeons.

Despite the existence of these pigeon-specific vaccines, the major obstacles in the mandatory ND vaccination of pigeons were the cost and the packaging of vaccines with an MA. The IMOPEST® vaccine marketed by Merial, which could overcome these obstacles, had an MA for ND vaccination in chickens only. This vaccine did not have an MA for use in pigeons; however, following a request to modify the target species, the modified summary of product characteristics was approved on 26 June 2013 by ANSES-ANMV. This vaccine is therefore now authorised for use in pigeons. As a result, Memorandum DGAL/SDSPA/N2012-8145 of 9 July 2012 regarding the mandatory vaccination of pigeons against ND and ensuring effective vaccination was modified.

Surveillance of wild birds and decoy ducks with respect to avian *influenza*

Procedures (Box 2)

Active surveillance on wild birds captured or hunted ended in 2011, after publication of the recommendations made in European Commission Decision 2010/367/EU, which indicated that, since its implementation in Europe, this type of surveillance had only helped to detect LPAI, but never the highly pathogenic AI virus (HPAI), with few exceptions. The National Office for Hunting and Wildlife (ONCFS) oversaw this surveillance from 2003 to 2011 through an agreement between the DGAL and ONCFS.

Passive surveillance remains the only type of surveillance that is currently used for wild birds and decoy ducks.

Results

In 2012, only 49 dead birds were screened for AI by PCR and no decoy ducks were tested. All tests showed negative results. This number continues to decrease (Figure 4), thus calling for reactivation of passive surveillance which requires training and information campaigns and new and revised surveillance procedures. This item has been included in the agenda for the working group on monitoring wildlife at the ESA Platform and was noted during the assessment of the surveillance programme using the OASIS method (see below).

Box 1. Newcastle disease (ND) surveillance and control measures

Objectives of the surveillance programme
- To confirm and maintain the ND-free status (as defined by the OIE health code) of France.
- To detect as early as possible any evidence of virus circulation in poultry and captive birds.

Monitored population
Poultry species and captive birds throughout France.

Surveillance procedures
- Outbreak surveillance: notification of clinical suspicions in poultry and captive birds to the DDecPP.
- Programmed surveillance: none.

Vaccination
Mandatory vaccination in pigeons.

Definition of a case
Newcastle disease: infection caused by any avian strain of paramyxovirus type 1 in day-old chicks with an intracerebral pathogenicity index (ICPI) greater than 0.7.

Poultry suspected to be infected: any poultry showing symptoms or *post mortem* lesions that lead to suspicion of ND.

Confirmed case of Newcastle disease: confirmation by the NRL of the presence of a paramyxovirus type 1 showing the characteristics of a virulent strain.

Animal health rules
- In the case of suspicion:
  - > *The holding is placed under an APMS surveillance order, samples (organs) are taken for virological analyses that entail inoculation on embryonated eggs and sent to one of the two laboratories accredited for virus isolation.*
  - > *Trace-back/trace-forward epidemiological survey: traceability of animals introduced to or leaving the holding during the risk period (21 days before the onset of symptoms).*

The objective is:
- > *to date the infection event and identify the origin of infection,*
- > *to estimate the risk of the virus spreading and thus take control measures according to this risk,*
- > *to determine which holdings are at risk, i.e. holdings with epidemiological connections with a suspect holding, as well as poultry farms located near the suspect holding.*

- When an outbreak has been confirmed, the holding is placed under an APDI order, the poultry stock is slaughtered (exceptions possible for captive birds with a 60-day confinement period), cleansing and disinfection operations, protection and surveillance zones of 3 and 10 km, respectively, except for outbreaks in captive birds.

Regulations

– Ministerial Order of 8 June 1994 laying down the control measures for Newcastle disease

Box 2. Avian *influenza* surveillance and control measures

Objectives of the surveillance programme
- To confirm and maintain the avian *influenza*-free status (as defined by the OIE health code) of France.
- To detect as early as possible any appearance of circulation of low pathogenic (LPAI) and high pathogenic avian *influenza* (HPAI) strains in poultry and captive birds and HPAI strains in wild birds.

Monitored population
Poultry, captive birds and wild birds found in France.

Surveillance procedures
- Outbreak surveillance:
  - > *Poultry holdings: notification of clinical suspicion based on alert criteria (Ministerial Order of 24 January 2008).*
  - > *Wild birds: notification of mortality and collection of dead wild birds according to the definition of abnormal death (Memorandum DGAL/SDSPA/N2007-8056 of 28 February 2007): one swan carcass or five birds on one given site within a period of seven days or less.*
  - > *Decoy ducks: notification to the state veterinarian and FDC of the death of more than five birds or symptoms affecting the nervous system observed in more than five individuals in a seven-day period (Memorandum DGAL/SDSPA/N2011-8007 of 4 January 2011).*
- Programmed surveillance:
  - > *Poultry holdings: Annual serological testing (H5, H7) targeting the most at-risk holdings, i.e. outdoor holdings, near wetlands, holdings with insufficient biosafety measures, multi-species holdings, species with higher susceptibility such as turkeys, holdings with long-lived poultry production categories or holdings with frequent commercial movements (breeding holdings), species having shown higher serological prevalence (ducks, game birds) and also départements with a high density of poultry holdings,*
  - > *wild birds and decoy ducks: active surveillance ended for these categories in 2012 and 2011, respectively.*

Vaccination
Vaccination prohibited in France except for any vaccination programme approved by the European Commission.

Definition of a case
HPAI: Infection caused by an avian *influenza* virus:
- > *belonging to subtypes H5 or H7 with genomic sequences coding for multiple basic amino acids at the haemagglutination cleavage site, similar to those observed for other HPAI viruses, indicating that haemagglutinin can undergo cleavage by a ubiquitous host protease;*
- > *or showing, in six-week old chickens, an intravenous pathogenicity index greater than 1.2.*

LPAI: infection caused by avian *influenza* virus subtype H5 or H7 that does not fit the previous definition.

Suspicion of avian influenza: suspicion based on:
- > *Epidemiological or clinical evidence or lesions. According to the evidence, suspicion can be oriented towards LPAI or HPAI and/or*
- > *non-negative results in laboratory tests lead to suspicion of infection by an AI virus (positive H5 or H7 serology confirmed by the NRL or positive PCR for the M or H5 gene in an accredited laboratory).*

Confirmation of avian influenza: confirmation of infection by a LPAI or HPAI virus by the NRL.

Animal health rules
- In the case of (clinical or analytical) suspicion:
  - > *holding is placed under an APMS order,*
  - > *samples are taken for virological PCR analyses in an accredited laboratory (Memorandum DGAL/SDSPA/N2007-8162 of 5 July 2007) or sent to the NRL for confirmation of a positive PCR in an accredited laboratory and LPAI/HPAI strain determination.*
- In the case of analytical suspicion from a waterfowl holding without clinical symptoms (positive serological tests for H5 or H7 confirmed by the NRL), additional samples are taken depending on whether the original incriminated batch is still present in the holding, whether biosafety measures have been implemented, and the rate of seropositive individuals in the batch tested originally (Memorandum DGAL/SDSPA/N2008-8287 of 18 November 2008).
- Trace-back/trace-forward epidemiological survey:

The objective is:
- > *to date the event and identify the origin of the contamination;*
- > *to estimate the risk of the virus spreading and thus take appropriate control measures according to this risk;*
- > *to determine which holdings are at risk, i.e. holdings with epidemiological connections with a suspect holding, as well as poultry farms located near the suspect holding.*

- In the case of a confirmed outbreak, the holding is placed under an APDI order, animals are slaughtered or sent to a slaughterhouse if infection with LPAI, cleansing and disinfection operations are undertaken, zones (3 and 10 km) are set up for HPAI and 1 km for LPAI.

Reglementation

– Council Directive 2005/94/EC of 20 December 2005 on Community measures for the control of avian *influenza* and repealing Directive 92/40/EEC

– Commission Decision 2010/367/EU of 25 June 2010 on the implementation by Member States of surveillance programmes for avian *influenza* in poultry and wild birds

– Ministerial Order of 18 January 2008 laying down the technical and administrative measures for the control of avian *influenza*

– Ministerial Order of 24 January 2008 regarding the level of epizootic risk due to infection of birds by a highly pathogenic avian *influenza* virus and the surveillance system and control measures for captive birds

Assessment of the surveillance programme

In March 2012, as part of ESA Platform activities, the DGAL requested an assessment of the surveillance programme for avian diseases in France using the OASIS method. This semi-quantitative method was developed by ANSES and helps to identify and illustrate the strengths and weakness of a surveillance programme.

This assessment was conducted from March 2012 to October 2012. It included interviews with those involved in the surveillance programme at the national administrative level and at the local and regional levels in four *départements* (Vendée, Pas de Calais, Ain and Moselle) chosen according to their poultry production activities and their exposure to risk factors favouring the appearance of an infection. Validated by the assessment team and by the monitoring group at the ESA Platform, the report was delivered to the DGAL in August 2013.

The main recommendations mentioned in this report involve the organisation of government bodies to create a central coordination unit that can be extended to the entire poultry production sector. The improvement in acceptability of suspicions is a priority issue, with the implementation of differential diagnosis (to classify suspicions) and with the transfer of some techniques to accredited laboratories (H7 PCR test, ND PCR test) to shorten the interval for confirming or dispelling suspicions. The enhancement of passive surveillance in wild birds and the improvement in data management to produce indicators to determine how well the surveillance plan is functioning are the other points highlighted in this assessment.

These recommendations will be studied by DGAL to determine which ones will be followed up and implemented.

Conclusion and outlook

Since the last HPAI outbreak in French holdings in 2006, and the summer outbreaks involving wild birds in Moselle in 2007, no HPAI viruses have been detected in France.

However, the recent HP H7N7 outbreak in Italy was a potent reminder that surveillance and biosafety measures must be taken in holdings and any suspicion of AI must be reported to maintain effective and efficient passive surveillance.

Based on the OASIS assessment and recent AI events, the surveillance programme can be adjusted to respond to the current disease situation and to meet the goal of an efficient surveillance programme.

Acknowledgments

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References

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[2] Memorandum DGAL/SDSPA/N2012-8134 of 27 June 2010 "Survey on avian *influenza* in 2012".

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[4] Memorandum DGAL/SDSPA/N2013-8047 of 27 February 2013: "Reiteration of the surveillance of avian *influenza* and the relevant biosafety measures"

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Overview of implementation of the control programme for *Salmonella* in *Gallus gallus* and *Meleagris gallopavo* flocks in 2012

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Abstract

The mandatory programme to control salmonella involved all *Gallus gallus* and *Meleagris gallopavo* flocks. The infection rate in *Gallus gallus* breeding flocks remained relatively stable. The infection rate for *Salmonella* Enteritidis and Typhimurium in farms of table egg laying hens has remained stable since 2011. And lastly, due to the low number of broiler chicken and fattening turkey farms in which *Salmonella* Enteritidis and *Salmonella* Typhimurium were detected, France was able to comply with the European goal of prevalence reduction. The overall cost of the control programme continued to fall. In addition, a non-regulated, multi-antibiotic-resistant strain of *Salmonella* Kentucky CIP R - also resistant to ciprofloxacin - was identified in late 2012 in two fattening turkey farms in the Morbihan *département*, probably imported from a country in which this serotype is present in poultry farms.

Keywords

Salmonella, epidemiological surveillance, health control, *Gallus gallus*, *Meleagris gallopavo*, France

Résumé

Bilan d'exécution du programme de lutte contre* Salmonella *dans les troupeaux des espèces* Gallus gallus *et* Meleagris gallopavo *en 2012

Le programme de lutte obligatoire contre les salmonelles concerne tous les troupeaux de Gallus gallus *et de* Meleagris gallopavo. *Le taux d'infection dans les troupeaux de reproducteurs de l'espèce* Gallus gallus *reste relativement stable. Le taux d'infection vis-à-vis de* Salmonella *Enteritidis et Typhimurium à l'étage poules pondeuses d'œufs de consommation est stable depuis 2011. Enfin, le nombre de troupeaux de poulets de chair et de dindes d'engraissement, dans lesquels* Salmonella *Enteritidis et* Salmonella *Typhimurium ont été détectées, permet à la France de respecter l'objectif européen de réduction de la prévalence. Le coût global du programme de lutte continue de diminuer. Par ailleurs une souche non réglementée de* Salmonella *Kentucky CIP R multirésistante aux antibiotiques, dont la ciprofloxacine, a été identifiée en fin d'année 2012 dans deux élevages de dindes du Morbihan, probablement importée à partir d'un pays tiers dans lequel la présence de ce sérotype est implanté dans les élevages de volailles.*

Mots clés

Salmonella, *épidémiosurveillance, police sanitaire,* Gallus gallus, Meleagris gallopavo, *France*

The objective of Salmonella surveillance in poultry flocks is to prevent the occurrence of food-borne diseases. Salmonella bacteria are transmitted through the different levels of the breeding scheme; control measures therefore involve not only poultry raised for food production (eggs, meat), but also poultry reared for breeding purposes.

European Regulation (EC) No. 2160/2003 lays down the general framework for controlling Salmonella infections in the poultry sector in Member States. Specific regulations for implementing EU legislation define the prevalence targets and the details of the screening programme (sampling protocol, duties on the part of the farmers and competent authorities, laboratory analyses):

- European Regulation (EC) No. 200/2010 for adult breeding flocks of *Gallus gallus*;
- Regulation (EC) No 517/2011 for laying hens of *Gallus gallus*;
- Regulation (EC) No 200/2012 for flocks of broilers;
- Regulation (EC) No 584/2008 (repealed and replaced by Regulation (EC) No 1190/2012 for 2013) for adult breeding and fattening turkeys.

The French national control programme was progressively aligned with the European regulations as it was being developed:

- Ministerial Order of 26 February 2008 as amended regarding flocks of *Gallus gallus* breeding hens and table egg-laying hens;
- Ministerial Order of 4 December 2009 as amended regarding breeding turkeys;
- Ministerial Order of 22 December 2009 as amended (repealed and replaced by Ministerial Order 24 April 2013 for 2013) regarding broilers and fattening turkeys.

Screening programme in 2012

The screening programme has been the same since 2010 for flocks of all poultry species (Box).

French regulations include the following Salmonella Typhimurium variants: 1,4,[5],12,i :- , 1,4,[5],12,- :1,2 and 1,4,[5],12,- :- (Table 1).

The strains isolated from screening are held at the ANSES Ploufragan-Plouzané Laboratory, the designated NRL for Salmonella. This strain collection can be used for retrospective typing studies, if required.

National coverage of the screening programme

Since 2010, screening for avian Salmonella infections has been mandatory for flocks of Gallus gallus and Meleagris gallopavo, irrespective of their level in the breeding scheme, their geographic location or the epidemiological situation of the flock, with the exception of small flocks of less than 250 individuals, whose products are distributed in small quantities directly to the end consumer.

Screening results in 2012 (Tables 2 and 3)

The positive cases in regard to Category 1 health hazards are given for all Gallus gallus and Meleagris gallopavo production sectors in Table 2 and a comparison with previous years' results is given in Table 3.

Breeding flocks of Gallus gallus *(Table 4)*

In 2011 and 2012, serovar Enteritidis was absent from the table-egg laying and broiler sectors, representing an improvement over 2010.

At the breeding level of the table-egg sector, no flocks were identified as positive for any of the five serovars. The 11 positive flocks were all breeder flocks from the broiler sector. In this sector, serovar Typhimurium was the predominant Salmonella serovar isolated.

Serovar Typhimurium (sensu stricto) was found in four pre-adult multiplier flocks, one flock of pre-adult primary breeders and two flocks of adult primary breeders.

The Typhimurium variant 1,4,[5],12,i :- was found in two pre-adult multiplier flocks, one adult multiplier flock and one pre-adult primary breeder flock.

The overall infection rate, for both sectors and all levels of the breeding scheme, was 0.47% for pre-adult breeders (0% in 2011). It was 0.13% for adult breeders (0.47% in 2011), which is lower than the European target, set to 1% for adult breeders by Regulation (EC) No. 200/2010. The infection rate at the adult breeding level thus decreased in 2012, although the total number of positive flocks remained relatively stable across the whole breeding scheme.

Laying hen flocks (Table 5 and Figure 1)

At the production level in the French layer sector, European Regulation (EC) No. 517/2011 targets a 10% reduction in the prevalence of Salmonella Enteritidis and Salmonella Typhimurium every year, or a stable rate of less than 2%. The targeted reduction in prevalence was set according to the 2005 EU-wide baseline survey; a prevalence of 8% was observed in France in 2005.

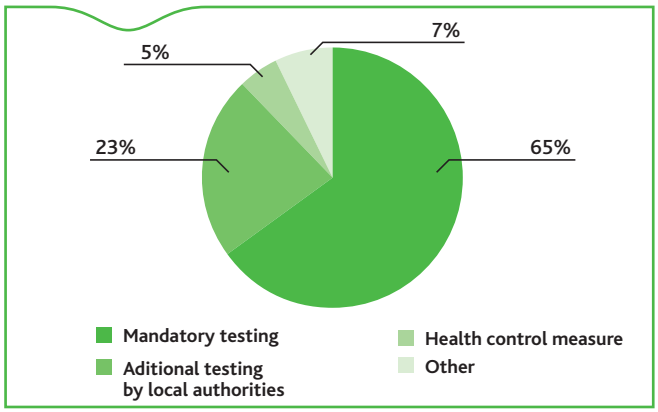

Figure 1. Detection of suspected cases of infection with *Salmonella Enteritidis* and *Salmonella Typhimurium* for table egg layers in 2012

France achieved this reduction especially in 2010 (a reduction of more than 35% was observed between 2009 and 2010). However, this change in the infection rate should be interpreted with caution because the way the denominator is calculated changed between 2009 and 2010. In 2009, the denominator was the number of flocks declared for testing whereas since 2010, the denominator has been the number of flocks that were tested in accordance with European regulations. The infection rate was thus overestimated prior to 2010.

In 2011 and 2012, the targeted prevalence was less than 2% in laying hen flocks, with observed values of 1.62% in 2010, 1.45% in 2011 and 1.42% in 2012 (Table 5).

Among the variants of serovar Typhimurium, only the 1,4,[5],12,i :- variant was found in 2011 and 2012.

The proportion of suspected cases of infection with Salmonella Enteritidis and Salmonella Typhimurium is given in Figure 1 according to how it was detected in the surveillance programme. Two-thirds of the suspected cases were detected via routine testing carried out by poultry farmers.

Breeder turkey flocks (Table 6)

The prevalence, as defined by European Regulation (EC) No. 584/2008, was 0.52% for adult breeder turkey flocks and 0% for pre-adult breeder turkey flocks. This result can be attributed to the Salmonella control programme that poultry farmers had already applied in their breeder flocks, under a scheme called the "Contract for Progress". This "contract" requires sampling, preventive culling of any positive flocks and good hygiene practices in turkey holdings.

Table 1. Classification of serovars as Category 1 health hazards (formerly "notifiable (animal) disease with compulsory control measures (NDCCM)") or Category 2 health hazards (formerly notifiable diseases, ND), according to the type of flock

	Enteritidis	Hadar	Infantis	Typhimurium (and its variants)	Virchow	Other serovars
***Gallus gallus* – broiler sector**						
Primary breeder flock - pre-adults	1	1	1	1	1	2
Primary breeder flock - adults	1	1	1	1	1	2
Multiplier flock - pre-adults	1	1	1	1	1	2
Multiplier flock - adults	1	1	1	1	1	2
Broiler production flocks	1	2	2	1	2	2
***Gallus gallus* – layer sector**						
Primary breeder flock - pre-adults	1	1	1	1	1	2
Primary breeder flock - adults	1	1	1	1	1	2
Multiplier flock - pre-adults	1	1	1	1	1	2
Multiplier flock - adults	1	1	1	1	1	2
Layer flocks - pre-adults (pullets)	1	2	2	1	2	2
Layer production flocks (laying hens)	1	2	2	1	2	2
Meleagris gallopavo						
Primary breeder flock - pre-adults	1	2	2	1	2	2
Primary breeder flock - adults	1	2	2	1	2	2
Multiplier flock - pre-adults	1	2	2	1	2	2
Multiplier flock - adults	1	2	2	1	2	2
(fattening turkeys)	1	2	2	1	2	2

Table 2. Summary of the results of Category 1 *Salmonella* for 2012

	Total number of individuals covered by the programme	Number of flocks tested	Number of flocks positive for category 1 health hazard Salmonella in 2012	Number of flocks culled preventively	Number of individuals culled preventively	Number of eggs destroyed or heat-treated
***Gallus gallus* – broiler sector – breeding level**						
Primary breeder flock - pre-adults	2,771,086	304	2	2	12,632	0
Primary breeder flock - adults	2,311,562	280	2	2	8,613	64,440
Multiplier flock - pre-adults	13,602,449	1,290	6	6	82,290	0
Multiplier flock - adults	14,950,449	1,856	1	1	5,475	41,474
***Gallus gallus* – layer sector – breeding level**						
Primary breeder flock - pre-adults	80,806	7	0	0	0	0
Primary breeder flock - adults	542,594	41	0	0	0	0
Multiplier flock - pre-adults	1,413,993	96	0	0	0	0
Multiplier flock - adults	1,922,984	160	0	0	0	0
***Gallus gallus* – layer sector – production level**						
Layer flock - pre-adults (pullets)	68,218,530	3,156	3	2	21,534	0
Layer flock (laying hens)	48,587,262	4,026	58	46	466,430	34,251,381
***Meleagris gallopavo* – breeding level**						
Primary breeder flock - pre-adults	133,263	34	0	0	0	0
Primary breeder flock - adults	105,774	51	0	0	0	0
Multiplier flock - pre-adults	2,537,080	520	0	2	6,584	0
Multiplier flock - adults	3,150,960	912	5	6	15,302	232,500
***Gallus gallus* – broiler and *Meleagris gallopavo* - production**						
Meat production (broilers and fattening turkeys)	810,915,255	64,563	328	328	666,304	0

Serovar Typhimurium (and its variants) is frequent in the turkey sector and is the main serovar isolated at the breeding level (Table 6).

Broiler chicken flocks (Table 7)

The results of the analyses have been compiled more exhaustively since 2011. For 2012, results on broiler chickens were pooled with fattening turkeys (Table 9).

The results obtained in 2011 met the European target set for late 2011, i.e. less than 1% (Table 7). The observed prevalence was higher than that observed during the EU-wide baseline survey in 2006-2007. This result can be attributed to a probable underestimation of the number of flocks tested and also to the inclusion of overseas *départements* since 2009, particularly Reunion Island, which alone accounts for more than 15% of broiler flocks found to be positive for Salmonella Typhimurium nationally. Therefore, in 2014, a specific vaccination programme, using an attenuated live vaccine, will be launched on Reunion Island to reduce the number of holdings contaminated due to recurrent infection with Salmonella Typhimurium. The ultimate objective is to reduce the prevalence of Salmonella Typhimurium in this *département*.

Variants of Salmonella serovar Typhimurium were present in broiler flocks, particularly the monophasic variant 1,1,[5],121i :-.

Table 3. Annual number of positive cases of infections with Category 1 *Salmonella* from 2006 to 2012. Since 2010, serovar Typhimurium also includes the following variants: 1,[5],121i :- , 1,1,[5],121- :1,1 and 1,1,[5],121- :-<

Year		2006	2007	2008	2009	2010	2011	2012
***Gallus gallus* broiler sector**								
Pre-adult breeders								
Number of flocks tested		ND	1,045	1,049	1,070	1,186	1,336	1,594
Number of positives	Enteritidis	1	3	5	2	0	0	0
	Typhimurium	3	2	0	0	0	0	5 + 3 variants « i :- »
	Hadar	Not tested	1	0	0	0	0	0
	Infantis	Not tested	0	0	0	0	1	0
	Virchow	Not tested	1	0	0	0	0	0
Adult breeders								
Number of flocks tested		1,023	1,030	998	1,041	1,487	1,529	2,136
Number of positives	Enteritidis	4	3	1	3	4	0	0
	Typhimurium	1	0	4	0	3	5	2 + 1 variants « i :- »
	Hadar	Not tested	2	0	0	0	0	0
	Infantis	Not tested	1	0	0	0	0	0
	Virchow	Not tested	0	0	0	1	0	0
***Gallus gallus* table egg sector**								
Pre-adult breeders								
Number of flocks tested		45	111	70	80	115	87	103
Number of postive flocks	Enteritidis	0	0	0	1	0	0	0
	Typhimurium	0	0	0	0	0	0	0
	Hadar	Not tested	0	0	0	0	0	0
	Infantis	Not tested	0	0	0	0	0	0
	Virchow	Not tested	0	0	0	0	0	0
Adult breeders								
Number of flocks tested		88	147	105	108	182	132	201
Number of postive flocks	Enteritidis	0	1	0	0	1	0	0
	Typhimurium	0	0	0	0	0	0	0
	Hadar	Not tested	0	0	0	0	0	0
	Infantis	Not tested	0	0	0	0	0	0
	Virchow	Not tested	0	0	0	0	0	0
Pre-adult layers (pullets)								
Number of flocks tested		1,607	2,115	2,093	2,050	2,330	2,060	3,156
Number of postive flocks	Enteritidis	7	7	6	2	1	0	3
	Typhimurium	3	7	3	9	2 (+ 1 variant « i:- »)	1 (+ 2 variants « i:- »)	0
Table-egg laying hens								
Number of flocks tested		3,099	2,980	3,067	2,855	4,013	4,000	4,026
Number of postive flocks	Enteritidis	104	81	62	51**	48****	38	26
	Typhimurium	(18*)	33	36	22	20 (+ 6 variants « i:- » and 1 variant « -:1,2 »****)	19 (+ 1 variant « i:- »)	28 + 3 variants « i :- » + 1 variant « -:- »
Meleagris gallopavo								
Pre-adult breeders								
Number of flocks tested		ND	ND	ND	ND	455	459	554
Number of positives	Enteritidis	Not tested	Not tested	Not tested	Not tested	0	1	0
	Typhimurium	Not tested	Not tested	Not tested	Not tested	1	2	0
Adult breeders								
Number of flocks tested		ND	ND	ND	ND	785	689	963
Number of positives	Enteritidis	Not tested	Not tested	Not tested	Not tested	0	0	1
	Typhimurium	Not tested	Not tested	Not tested	Not tested	0 (+ 4 variants « i:- »)	2	2 + 2 variants « i :- »
Gallus gallus* broiler sector and *Meleagris gallopavo								
Meat chickens and turkeys								
Number of flocks tested		ND	ND	ND	35,911***	58,418	65,228	64,563
Number of positive flocks	Enteritidis	Not tested	Not tested	Not tested	78	75	96	93
	Typhimurium	Not tested	Not tested	Not tested	109	191(+ 24 variants « i:- » , 12 variants « -:1,2 » and 2 variants « -:- »)	226 (+ 27 variants « i:- » , 2 variants « -:1,2 » and 7 variants « -:- »)	191 (+37 variants « i:- », 5 variants « -:1,2 » and 2 variants « -:- »)

* Screening for S. Typhimurium was not mandatory - ** 51 infections in flocks targeted in the national control programme for Salmonella, other than backyard flocks identified in domestic foodborne disease outbreaks - *** number of flocks tested is probably underestimated due to difficulties in counting the number of negative flocks - **** including 3 flocks positive for both Enteritidis and Typhimurium

1 One flock positive for ST.m, ST.m - :- variant and - :1,2 variant

Table 4. Annual infection rate in *Gallus gallus* breeder flocks in France from 2004 to 2012. Calculation method: the denominator is the sum of all tested flocks in a given year, the numerator is the number of flocks showing a positive sample that year

Level	Stage	Enteritidis									Typhimurium									Hadar					
		04	05	06	07	08	09	10	11	12	04	05	06	07	08	09	10	11	12	07	08	09	10	11	12
Gallus gallus breeders - layer sector																									
Breeder flocks	pre-adult	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	adult	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Multiplier flocks	pre-adult	0	0	0	0	0	1.6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	adult	0	0	0	0.88	0	0	0.77	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gallus gallus breeders - broiler sector																									
Breeder flocks	pre-adult	0	0	0	1.05	0	0	0	0	0	0	0	0	0	0	0	0	0	0.66	0	0	0	0	0	0
	adult	0	0	1.4	0	0	1.7	0	0	0	0	0	0	0	0	0	0	0.52	0.71	0	0	0	0	0	0
Multiplier flocks	pre-adult	0	0	0.1	0.12	0.6	0.2	0	0	0	0	0	0.1	0.2	0	0	0	0	0.47	0.1	0	0	0	0	0
	adult	0.2	0.6	0.2	0.33	0.1	0.1	0.3	0	0	0.1	0.1	0.1	0	0.6	0	0.23	0.3	0.05	0.2	0	0	0	0	0

Level	Stage	Infantis						Virchow						SE SI SH ST SV					
		07	08	09	10	11	12	07	08	09	10	11	12	07	08	09	10	11	12
Gallus gallus breeders - layer sector																			
Breeder flocks	pre-adult	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	adult	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Multiplier flocks	pre-adult	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1.6	0	0	0
	adult	0	0	0	0	0	0	0	0	0	0	0	0	0.9	0	0	0.77	0	0
Gallus gallus breeders - broiler sector																			
Breeder flocks	pre-adult	0	0	0	0	0	0	0.5	0	0	0	0	0	1.6	0	0	0	0	0.66
	adult	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1.7	0	0.52	0.71
Multiplier flocks	pre-adult	0	0	0	0	0.09	0	0	0	0	0	0	0	0.4	0.6	0.2	0	0.09	0.47
	adult	0.1	0	0	0	0	0	0	0	0	0	0	0	0.7	0.7	0.1	0.53	0.3	0.05

Table 5. Annual infection rate in laying hen flocks in France from 2007 to 2012. Calculation method: the denominator is the sum of all tested flocks in a given year, the numerator is the number of flocks showing a positive sample that year

Stage	Salmonella Enteritidis						Salmonella Typhimurium					
	2007	2008	2009	2010	2011	**2012**	2007	2008	2009	2010	2011	**2012**
Pre-adult table-egg laying hens	0.33	0.19	0.10	0.04	0.00	**0.10**	0.33	0.29	0.44	0.10	0.05	**0.00**
Table egg-laying hens	2.74	2.02	1.79	1.20	0.95	**0.65**		1.17	0.77	0.50	0.48	**0.79**

Stage	Salmonella Typhimurium Variant i;-			Salmonella Typhimurium Variant -;1,2			Salmonella Typhimurium Variant -;-			Salmonella Enteritidis/Salmonella Typhimurium				
	2010	2011	**2012**	2010	2011	**2012**	2010	2011	**2012**	2008	2009	2010	2011	**2012**
Pre-adult table-egg laying hens	0	0	**0**	0.33	0.19	**0.10**	0.00	0.00	**0.00**	0.48	0.54	0.13	0.15	**0.10**
Table egg-laying hens	0	0	**0**	2.74	2.02	**1.79**	0.00	0.00	**0.02**	3.16	2.56	1.62	1.45	**1.42**

Table 6. Annual infection rate in breeder turkey flocks in France from 2010 to 2012. Calculation method: the denominator is the sum of all tested flocks in a given year, the numerator is the number of flocks showing a positive sample that year

	Number of positive flocks in 2010	Infection rate in 2010 (49,024 flocks declared for testing)	Number of positive flocks in 2011	Infection rate in 2011 (57,182 flocks declared for testing)	Number of positive flocks in 2012	Infection rate in 2012 (1 517 flocks declared for testing)
Enteritidis	0	0.00%	1	0.09%	1	0.07%
Typhimurium strict	1	0.08%	4	0.35%	2	0.13%
SE ST strict	**1**	**0.08%**	**5**	**0.44%**	**3**	**0.20%**
1,4,[5],12,i :-	4	0.32%	0	0.00%	2	0.13%
1,4,[5],12,- :1,2	0	0.00%	0	0.00%	0	0.00%
1,4,[5],12,- :-	0	0.00%	0	0.00%	0	0.00%

Table 7. Annual infection rate in broiler chicken flocks in France from 2009 to 2011. Calculation method: the denominator is the sum of all tested flocks in a given year, the numerator is the number of flocks showing a positive sample that year, from 2009 to 2011

	Prevalence in 2006-2007 survey	Number of positive flocks in 2009	Infection rate in 2009 (35,911 flocks declared for testing)	Number of positive flocks in 2010	Infection rate in 2010 (49,024 flocks declared for testing)	Number of positive flocks in 2011	Infection rate in 2011 (57,182 flocks declared for testing)
Enteritidis	0.20 %	81*	0.23	61	0.12	82	0.14
Typhimurium *sensu stricto*	0.10 %	109*	0.30	149	0.30	179**	0.31
SE ST strict	**0.30 %**	**188***	**0.52**	**210**	**0.43**	**261**	**0.45**
1,4,[5],12,i :-	ND	ND	ND	21	0.04	19	0.03
1,4,[5],12,- :1,2	ND	ND	ND	10	0.02	2**	0.003
1,4,[5],12,- :-	ND	ND	ND	2	0.004	5**	0.01

* Two flocks were positive for both Enteritidis and Typhimurium

** One flock was positive for ST, the ST :- : variant and the ST - :1.2 variant

Table 8. Annual infection rate in fattening turkey flocks in France from 2010 to 2011. Calculation method: the denominator is the sum of all tested flocks in a given year, the numerator is the number of flocks showing a positive sample for that year, for 2010 and 2011

	Number of positive flocks in 2010	Infection rate in 2010 (%) (9 394 locks declared for testing)	Number of positive flocks in 2011	Infection rate in 2011 (%) (8 046 locks declared for testing)
Enteritidis	14	0.15	14	0.17
Typhimurium strict	42	0.45	47	0.58
SE ST strict	**56**	**0.60**	**61**	**0.76**
1,4,[5],12,i :-	3	0.03	8	0.10
1,4,[5],12,- :1,2	2	0.02	0	0.00
1,4,[5],12,- :-	0	0.00	2	0.02

Table 9. Annual infection rate in broiler and fattening turkey flocks in France from 2010 to 2012. Calculation method: the denominator is the sum of all tested flocks in a given year, the numerator is the number of flocks showing a positive sample that year, from 2010 to 2012

	Number of positive flocks in 2010	Infection rate in 2010 (%) (58 418 flocks declared for testing)	Number of positive flocks in 2011	Infection rate in 2011 (%) (65 228 flocks declared for testing)	Number of positive flocks in 2012	Infection rate in 2012 (%) (64 563 flocks declared for testing)
Enteritidis	75	0.13	96	0.15	93	0.14
Typhimurium strict	191	0.33	226	0.35	191	0.30
SE ST strict	**266**	**0.46**	**322**	**0.49**	**284**	**0.44**
1,4,[5],12,i :-	24	0.04	27	0.04	37	0.06
1,4,[5],12,- :1,2	12	0.02	2	0.003	5	0.008
1,4,[5],12,- :-	2	0.003	7	0.01	2	0.003

Fattening turkey flocks (Table 8)

For fattening turkeys, serovar Typhimurium (s.s.) accounted for 77% of the positive cases of Enteritidis and Typhimurium s.s.

The European target was met for fattening turkey flocks with a prevalence of 0.76% (Table 8).

For 2012, results on fattening turkeys were pooled with those for broiler chickens (Table 9).

Broiler chickens and fattening turkey flocks (Table 9)

For 2012, the results for all meat production categories were combined, i.e. results on broiler chickens and fattening turkeys were pooled. To compare these results with those of previous years, the rates of positive cases observed in 2010 and 2011 have been pooled.

The results obtained in 2012 met the European target set for late 2012, i.e. less than 1% (Table 9). The prevalence of Enteritidis and Typhimurium s.s. has varied little since 2010.

Control measures

Control measures have remained unchanged since 2009; they were extended to turkey flocks in 2010.

For broilers, the new Ministerial Order of 24 April 2013 includes the following changes:

- confirmatory sampling in muscles is no longer required in the event of a positive sample detected in the screening programme,
- confirmatory sampling is limited to special cases that will be described in detail in a forthcoming ministerial memorandum
- if confirmatory samples are also positive (i.e. APDI order), the entire flock can be culled shortly thereafter (decision depending on the risk of contamination for exposed holdings),
- implementation of Regulations (EU) No. 200/2012 (regarding broiler chickens) and No. 1190/2012 (regarding fattening turkeys) extending the validity of analysis results to 6 weeks before culling for poultry categories with long fattening periods (i.e. 81 days for broiler chickens, 100 days for turkeys), or for organic poultry production flocks.

Changes in the costs of the screening programme and control measures (Figure 2)

Until 2011 the pullet and laying hen stages for table egg production represented the highest share of the budget in the French control programme for *Salmonella* due to the high number of infected flocks and the number of compensation items paid (slaughter, cleaning and disinfection, official veterinary inspection and other costs). However, the costs have continued to decrease since 2006 (less than €900,000 in 2012). In 2012, the number of cases observed in breeding *Gallus gallus* increased slightly, particularly for pre-adult breeders, and

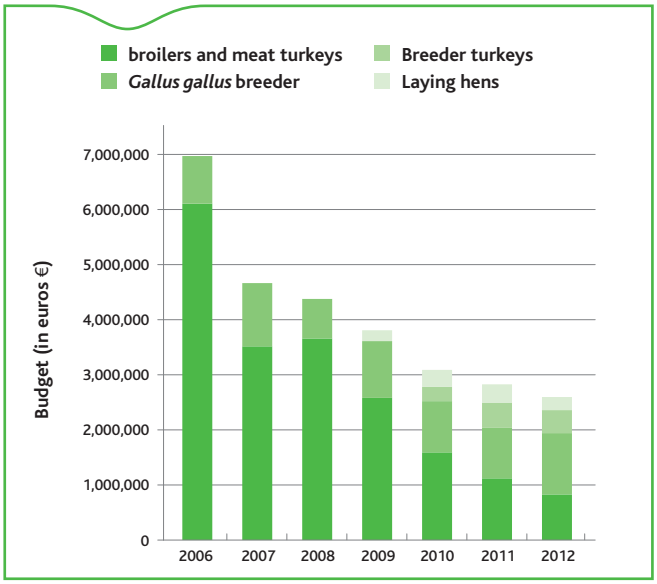

Figure 2. Annual government funds spent for the surveillance programme for *Salmonella* in poultry holdings from 2006 to 2012

the budget allocated to this level also increased, representing the highest budget line in 2012 (more than €1,100,000). The budget for broiler chicken flocks, for which only cleaning and disinfection operations are compensated, appears to be stable (€238,000 in 2012). Likewise, the budget for the control programme on breeder turkey flocks and the compensation paid has been stable since 2011 (€418,000).

Epidemiological investigation of the first Salmonella Kentucky outbreak involving a strain highly resistant to fluoroquinolones detected in a poultry holding in France (Guillon et al., 2013)

An epidemiological investigation was ordered after the identification of a *Salmonella* Kentucky CIP-R strain with multiple resistance to antimicrobials, including ciprofloxacin (a fluoroquinolone), in two fattening turkey holdings in Morbihan (Guillon *et al.*, 2013). The investigations concluded that this strain was introduced after the farmers had visited Morocco, an area in which the strain is endemic. Local surveillance measures were taken to ensure that the S. Kentucky CIP-R strain had been eliminated. This event illustrates:

- the usefulness of continuous surveillance of antimicrobial resistance in isolates collected by the *Salmonella* network, which includes all accredited and authorised laboratories;
- the efficient collaboration between the departmental and national veterinary services and public health organisations (ANSES, National centres of reference or NCRs);
- the relevance of regulations for the surveillance of emerging serovars.

Box. Surveillance and control measures for *Salmonella* infections in poultry

Objectives of the surveillance programme

- To detect, control and eradicate infections by Category 1 health hazard *Salmonella* serovars, as defined by Ministerial Decree No 2012-845 of 30 June 2012.
- To reduce the prevalence of infections by Category 1 health hazard *Salmonella* serovars and the risk that they present to public health.
- To assess the progress made in light of the obtained results.
- To monitor the emergence of any *Salmonella* serovars.

Monitored population

- Flocks of *Gallus gallus* (hens and chickens) and *Meleagris gallopavo* (turkeys), irrespective of their level in the poultry breeding scheme, their geographic location or epidemiological situation, with the exception of «small» flocks (less than 250 birds).
- *Salmonella* serovars classified as Category 1 health hazards (see table below).

Surveillance procedures

- Samples are taken by an mandated veterinarian, by a technician delegated and trained in veterinary sampling techniques by the mandated veterinarian or by DDecPP/DAAF staff technicians:
  - > *in poultry farms and hatcheries, minimum frequencies and the basic sampling programme are set by European regulations; French regulations voluntarily extended these regulations.*
  - > *for other* Salmonella *serovars (Category 2 health hazards), epidemiological surveillance is based on a systematic sampling programme carried out before moving or culling each poultry flock.*

Control measures (for Category 1 health hazard *Salmonella* serovars)

Control measures remain unchanged since 2009; they were extended to turkey flocks in 2010.

- **Suspicions** are based on any positive result from samples taken in the environment of a poultry flock. The suspected flock is then placed under an APMS monitoring order and, to confirm infection, the DDecPP/DAAF orders official sampling at the poultry farm and, if the flock involves production-level layers, on eggs.
- **Confirmation is based on:**
  - > *For breeders or pullets (future table egg-laying hens), mandatory preventive elimination of poultry and waste.*
  - > *For table egg-laying hens, preventive elimination is encouraged by offering compensation to the farm operator, but is not mandatory; however, all eggs from an infected flock are downgraded and can only be sold to the food-processing industry where they undergo heat treatment.*
  - > *At any production level of the poultry breeding scheme, emphasis is placed on the quality of cleaning and disinfection operations, whose effectiveness must be officially validated before repopulation; compensation is contingent upon this inspection.*

	S Enteritidis	*S Hadar*	*S Infantis*	*S Typhimurium*	*S Virchow*
Breeder flocks Gallus gallus	x	x	x	x	x
Breeder flocks Meleagris gallopavo	x			x	
Layer flocks Gallus gallus	x			x	
Meat-producing flocks Gallus gallus *and* Meleagris gallopavo	x			x	

Conclusion

The national control programme for *Salmonella* implemented since 1998 in *Gallus gallus* breeding flocks and layer flocks, since extended to broilers and turkeys, appears to be providing satisfactory results and the overall cost of the programme is decreasing.

At the *Gallus gallus* breeding level, the infection rate increased slightly in 2012, particularly for the pre-adult stage. Although the number of positive flocks at this level was still low (around 10 per year), the public health and economic consequences of these infections are potentially high.

In the layer sector, the number of cases of infection and the associated cost continued to fall.

In the broiler sector, the infection rate remained stable.

At the breeding level for the turkey sector, the number of cases of infection also remained stable. As noted for *Gallus gallus* breeding flocks, the public health and economic consequences of these infections are potentially high.

Although the number of infections has stabilised – or even decreased – and there has been a decrease in the costs for the French government, all poultry sectors must remain vigilant with regard to *Salmonella* infections.

The investigation on a non-regulated strain of *Salmonella* Kentucky CIP-R with multiple drug resistance shows how important it is to follow and apply biosafety rules to avoid introducing and spreading pathogens in holdings. This investigation also demonstrated how the various surveillance programmes work together and the central role of testing laboratories in optimising food safety in the food chain, from the farm level up. The control programme, focused on regulated diseases, is thus also useful for monitoring non-regulated diseases that, once emergent, can become major zoonotic risks, such as *Salmonella* Kentucky CIP-R.

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Overview of surveillance of **equine infectious anaemia (EIA)** in France in 2012: management of two clinical episodes

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Abstract

Equine Infectious Anemia virus (EIAV) belongs to the *Retroviridae* family, genus Lentivirus like the human immunodeficiency virus (HIV). EIAV infects horses, donkeys and mules and has a worldwide distribution. The virus is responsible for a persistent infection associated with clinical signs such as fever, anorexia and anaemia. Non-symptomatic horses are contagious and act as a viral reservoir. Consequently, positive horses need to be isolated before euthanising them. In 2012, 15,691 Coggins tests were performed by accredited French laboratories. Among those analyses, 27 were positive for EIA and came from eight equids. Positive equids came from two different sites: one located in the Vaucluse *département* , declared in February 2012, and the second from the Gard *département*, declared in August 2012. Phylogenetic *analysis* showed that the 2012 isolates are different from those isolated in France since 2007. It is important to reiterate that only laboratory diagnosis can confirm EAI infection.

Keywords

Horse, equine infectious anaemia, persistent infection

Résumé

Bilan de la surveillance de l'anémie infectieuse des équidés (AIE) en France en 2012 : gestion de deux épisodes cliniques

Le virus de l'anémie infectieuse des équidés (AIE) appartient à la famille des Retroviridae, *genre* lentivirus *comme le virus de l'immunodéficience humaine (VIH). Il infecte les chevaux, ânes et mulets et présente une répartition géographique mondiale. Le virus est responsable d'une infection persistante associée à l'apparition de signes cliniques évocateurs tels que de la fièvre, de l'anémie et de l'anorexie. Les équidés infectés asymptomatiques sont contagieux et sont les réservoirs du virus. C'est pourquoi, tout équidé séropositif pour l'AIE doit être isolé avant son euthanasie. En 2012, la surveillance évènementielle clinique et le dépistage réglementaire ont conduit à la réalisation de 15 691 analyses dont 27 étaient positives. Ces analyses positives concernaient huit équidés répartis en deux foyers distincts : le premier situé dans le département du Vaucluse déclaré en février 2012 et le deuxième situé dans le département du Gard, déclaré en août 2012. L'analyse phylogénétique montre que les isolats de 2012 sont différents de ceux identifiés en France depuis 2007. Il est important de rappeler que seul le diagnostic de laboratoire permet de confirmer une infection par l'AIE.*

Mots clés

Cheval, anémie infectieuse, infection persistante

The equine infectious anaemia virus (EIAV) is a member of the genus *Lentivirus* of the family *Retroviridae*. Only equines (horses, donkeys, zebras) can be infected by EIAV. Once infected, the animal is infected for life and remains a source of contagion for other equines, even in the absence of any clinical sign. The virus is bloodborne and transmitted primarily *via* biting flies, but also iatrogenically if non-sterile needles or surgical equipment are used. Insects — mainly horseflies and stable flies — are mechanical vectors: although the virus does not replicate within the insect, the infectious virus can remain in its mouthparts for several hours after biting. Insect transmission of the disease is most effective when horses are in close proximity with other horses because horseflies and stable flies are biting insects that often flit from one animal to another, starting and finishing their meals on different individuals. Equines are not subject to routine testing at any point during their lifetime. To diagnose EIA, the World Organisation for Animal Health (OIE) recommends a serological test, the agar gel immunodiffusion assay (AGID) known as the Coggins test (Coggins *et al.*, 1972). This test is not mandatory except in the case of import/export of equines, horse sales (EIA constitutes a redhibitory defect) and, for stallions, prior to breeding. EIA outbreaks are therefore often detected by a veterinarian upon observation of suggestive clinical signs in a horse. This initial suspicion may lead to tests and discovery of other seropositive horses on the same site or on a site with an epidemiological connection, whether or not they exhibit any symptoms of the disease.

Review of EIA surveillance in 2012

In 2012, the network of accredited laboratories carried out 15,691 serological analyses of which 27 tested positive. The 27 positive analyses involved eight equines in two distinct outbreaks in two French *départements*: Vaucluse and Gard. These two outbreaks were discovered following the appearance of clinical signs in the infected equines that were suggestive of EIA (listlessness, weight loss, anaemia, nasal bleeding, etc.).

Vaucluse outbreak

The primary outbreak was declared on 30 January 2012, after confirmation of the infection in a 16 year-old part-bred Arabian gelding that showed suggestive clinical signs such as fever, nasal bleeding, swelling of the lower abdomen, hind legs and the penile sheath, and pale mucus membranes. The APDI order for the outbreak site was issued on 2 February 2012, requiring restriction of movements and testing of all horses on the site. This site held seven other horses, three of which were also diagnosed with an EIAV infection on 6 February 2012 and all four infected horses were euthanised on 9 February 2012.

The epidemiological surveys covered a six-month period from August 2011 to January 2012 and focused on identifying (1) all horses that had been in 'contact' and at risk for EIA infection and (2) all horses present within a 1 km radius of the outbreak site.

These surveys revealed that some of the four infected horses had participated in an endurance race in September 2011 in Vaucluse. As a result, more than 180 horses were identified as being at risk of EIA infection; no cases of infection were detected, except for the four horses at the primary outbreak site. Despite the high number of at-risk horses exposed during the endurance race in late September 2011, the virus transmission was not demonstrated outside of the outbreak site, and there was no secondary transmission to horses off site.

Gard outbreak

The primary EIA outbreak declared in September 2012 in Gard involved six horses of the Camargue, Pura Raza Española (PRE), Barb and Mérens breeds. The index case, which exhibited fevers (40°C) and low (16%) haematocrit levels, was originally thought to be infected with babesiosis (for which it tested positive), and was treated for this disease. In the absence of subsequent improvement, the veterinarian supposed that the horse was infected with EIA and, noting the poor state of its health (massive weight loss of about 100 kg), the horse was euthanised on 30 August 2012. On 5 September, after the horse had been euthanised, the NRL confirmed that it was positive for EIA.

The epidemiological survey was first carried out on horses present within 500 m of the primary outbreak site as well as on all horses that had been in contact with the infected horses during the four months preceding the confirmation of the outbreak. The surveyed sites and farms, belonging to 25 different owners and involving 63 horses, were placed under APMS surveillance, as was the veterinary clinic where the primary case had been euthanised. The perimeter of the survey was then extended to 2 km around the primary outbreak site. In this extended perimeter, there were 315 additional horses, found on 42 different sites, including three equestrian centres. These new sites were also placed under APMS surveillance. The epidemiological surveys identified 378 at-risk 'contact' horses, among which only 364 could be sampled. Of these 364 horses, 2 belonging to the primary outbreak site tested positive for EIA (NRL confirmation on 11 September 2012). These two new cases were euthanised on 18 September 2012. A third EIA-positive horse was identified on 27 September 2012 and euthanised on 11 October 2012. This horse had an epidemiological connection with the primary outbreak site, as it had been put to pasture on a field adjacent to the primary outbreak site.

It is important to note that these epidemiological surveys were difficult to carry out and regrettably were not exhaustive due to several factors, e.g. (1) some horses could not be included in the survey because their owners had not provided proper information on them; (2) there were problems compiling a detailed registry on the owners of all the horses found in the surveillance zone; (3) the contact information of these horse owners was not always readily available.

Molecular epidemiology

To genotype the EIA strains isolated from the various outbreaks, the gag gene (1400 nucleotides long) was sequenced. EIA outbreaks in 2012 led to the identification of eight seropositive equines. The virus isolates were characterised from five of the eight euthanised horses. The phylogenetic analysis, carried out using the MEGA 5.0 software package, was used to compare and classify the viruses isolated in 2012 with respect to those isolated previously in France and those reported in the literature (Figure 1).

The obtained phylogenetic tree shows that the isolates from the outbreak sites in Vaucluse and Gard were not only different from each other but also from those identified previously in France, in particular those isolated in Var in 2009 (Ponçon *et al.*, 2011). Although there were indications from the epidemiological field survey that there was an epidemiological connection between the Vaucluse and Gard outbreaks *via* the purchase, sale or trade of horses, this hypothesis was not borne out by the phylogenetic analysis. In contrast, the data of this phylogenetic study show that the Vaucluse and Gard outbreaks had two distinct origins because the sequenced virus isolates were not identical. Furthermore, the seropositive horses in the Vaucluse outbreak were infected by two different isolates (Figure 1). The isolate characterised from horse 12D134 shows that it was related to isolates sampled in 2007 on donkeys in Ardèche (Rème *et al.*, 2009). However, no epidemiological connection could be established from the field surveys between horse 12D134 and the Ardèche donkeys euthanised in 2007. Likewise, the viral isolates characterised from horses 12D128 and 12D131 were similar to the virus isolated in 2010 in Dordogne on a trotting horse. Once again, no epidemiological connection was demonstrated between these two outbreaks. Moreover, the isolates characterised from horses 12D899 and 12D903, found in Gard, were

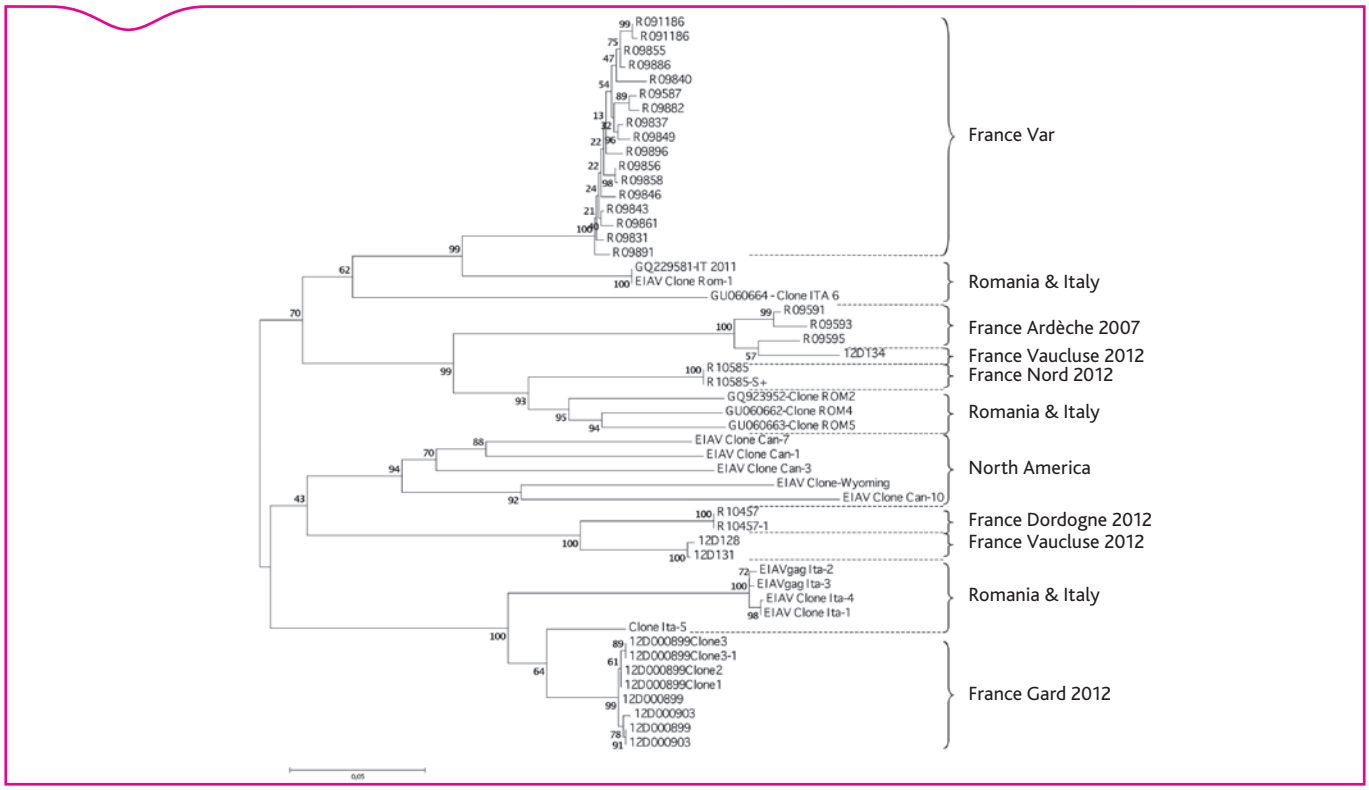

Figure 1. Phylogenetic analysis of 91 strains of the EIA virus. Analysis was done on the complete sequence of the *gag* gene (1400 nucleotides) from viruses isolated in France since 2007

similar to viruses isolated in Italy in the past few years (Hans *et al.*, 2012).

Cost

In 2012, the Vaucluse and Gard DDecPPs spent roughly €34,500 and €31,500 respectively, i.e. nearly €70,000 for the control of EIA. These costs do not include the time staff spent carrying out and following up on the epidemiological surveys. The low prevalence of EIA, euthanasia of only infected equines and the caps on the compensation paid to the owners of the euthanised horses helped to limit the costs.

EIA control measures

These measures are generally implemented when an infection has been confirmed, since, in accordance with current regulations, the state veterinary services are only informed of the outbreak when it has been confirmed. Control measures mainly consist of movement restrictions placed on the outbreak site and decontamination by euthanasia of the infected horses, the disinfection of facilities and materials, and monthly serological testing on the horses remaining at the outbreak site. The outbreak is considered to have been eliminated and the restrictive measures are lifted when the remaining horses have tested negative at least twice, three months apart.

At the same time, the horses at risk of infection (i.e. those that had been in any kind of contact with the infected animal and those present within 200 m) are identified and placed under surveillance (restriction of movements and regular serological testing to ensure there is no seroconversion 90 days after contact with the infected horse).

EIA is transmitted primarily by blood *via* biting insects (mainly tabanid species), or iatrogenically (use of contaminated syringes or needles). Epidemiological surveys show that, more often than not, virus transmission from an asymptomatic horse is low within an equine population. However, it is nonetheless essential to implement good animal health practices and use disposable, sterile materials.

Furthermore, better recording of property transfers and horse holding facilities, and requiring prompt updating of this information in the central SIRE database would help to facilitate the epidemiological surveys and improve equine health surveillance.

Lastly, although the prevalence of EIA in France is certainly very low, the potential impact of this disease should not be underestimated, particularly because there is no curative treatment and no way to manage the disease. Given that many infected equines do not show any symptoms, voluntary testing by horse owners would be an efficient surveillance measure, especially when horses are introduced into a new farm or during the sale, purchase or trade of horses, particularly considering that the EIA is a redhibitory defect. Moreover, given the attenuated and discreet clinical signs of EIA, screening for EIAV should also be considered on a more regular basis when symptoms lead veterinarians to suspect babesiosis.

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Overview of the monitoring of category 1 health hazards for fish in 2012: Viral Haemorrhagic Septicaemia (VHS), Infectious Haematopoietic Necrosis (IHN), and Koi Herpes Virus disease (KHV)

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Abstract
The concomitant intensification of single-species breeding in the aquaculture industry and trade of fish and eggs has complicated farm health management by encouraging the emergence in production areas of pathogens such as rhabdoviruses, responsible for Viral Haemorrhagic Septicemia (VHS) and Infectious Haematopoietic Necrosis (IHN). Appropriate monitoring was set up in the 1990s in order to evaluate the health situation. Surveillance applies to salmonid farming (the number one French fish production sector), as well as to pond fish farming, mainly with regard to the presence of pike, a typical species carrier of VHSV. In 2012, monitoring results confirmed maintenance of a stable and controlled health situation in France for these two rhabdovirus diseases. A herpesvirus outbreak in carp (HCV) was detected, confirming the contamination of our national carp livestock with this recently imported disease.

Keywords
Fish, viral diseases, health hazard, VHS, IHN, KHV, ISA

Résumé
Bilan pour l'année 2012 de la surveillance des principaux dangers sanitaires de première catégorie pour les poissons : septicémie hémorragique virale (SHV), nécrose hématopoïétique infectieuse (NHI) et herpèsvirose de la carpe (HVC)
La généralisation de l'élevage mono-spécifique intensif dans la filière piscicole et l'intensification des échanges de poissons et d'œufs a complexifié la gestion sanitaire des élevages en favorisant l'apparition et la diffusion d'agents pathogènes tels que les rhabdovirus, responsables de septicémie hémorragique virale (SHV), ou de nécrose hématopoïétique infectieuse (NHI) dans les bassins de production. Une surveillance appropriée a été mise en place à partir des années 1990 pour tenter de mieux appréhender cette situation sanitaire. Cette surveillance concerne en premier lieu les élevages de salmonidés (qui occupent la première place dans la production piscicole française) mais également la pisciculture d'étang, en raison de la présence d'une espèce typique de ce biotope, sensible au virus de la SHV : le brochet. Les résultats de la surveillance en 2012 confirment le maintien d'une situation sanitaire stable et maîtrisée sur le territoire vis-à-vis de ces deux maladies. Un foyer d'herpèsvirose de la carpe (HVC) a été détecté, confirmant la contamination de notre cheptel national de carpes par cette maladie importée récemment.

Mots clés
Poissons, maladies virales, dangers sanitaires, SHV, NHI, HVC, AIS

In fish, four non-exotic diseases previously classified as notifiable diseases with compulsory control measures (NDCCM) are henceforth defined as category 1 health hazards, under the terms of Decree 2012-845 of 30 June 2012 (Table 1). Three of these diseases are endemic in France. Koi Herpes Virus (KHV) had been detected only sporadically in France since 2001 but has become more frequent since 2011 (Papin *et al.*, 2012). Viral Haemorrhagic Septicaemia (VHS) and Infectious Haematopoietic Necrosis (IHN) are the two main diseases found in France. France is officially free of Infectious Salmon Anaemia (ISA).

These regulated diseases have been subject to surveillance since the entry into force of Directive 91/67/EEC, followed by Directive 2006/88/EC, in response to the health requirements set by European Regulations, to protect fish farms and to facilitate trade.

Surveillance is based on a dual system: a mandatory programme (outbreak and programmed surveillance), and a voluntary scheme (authorisation programmes) (see box).

Results of surveillance in 2012

Clinical surveillance

Clinical surveillance of VHS

One outbreak of VHS was reported in 2012 in a fish farm in Côte d'Or, a region that had not been declared as disease-free, following suspect mortalities of rainbow trout observed by a health service veterinarian. An epidemiological investigation carried out by the DDecPP of Côte d'Or and the aquaculture resource person for the region, based at the DDecPP of the Jura *département*, failed to identify the precise source of the outbreak.

Clinical surveillance of IHN

No cases of clinical suspicion of IHN were reported in 2012.

Clinical surveillance of KHV

One outbreak of KHV was reported in 2012 in a recreational fishing

Table 1. Classification of regulated fish diseases, their pathogens and health situation on 31 December 2012

Disease	Agent	Regulatory status	Health situation on 31 December 2012
Viral Haemorrhagic Septicaemia (VHS)	Rhabdovirus	Category 1 health hazard (formerly notifiable disease with compulsory control measures)	Present
Infectious Haematopoietic Necrosis (IHN)			Present
Koi Herpes Virus disease (KHV)	Herpesvirus		Present
Infectious Salmon Anaemia (ISA)	Orthomyxovirus		Absent

Box. Surveillance and control measures for regulated diseases in fish

Objectives of surveillance
- Early detection of any outbreak of regulated disease.
- To verify France's status as a Member State officially free of Infectious Salmon Anaemia.
- To grant "disease-free" status to aquaculture areas and farms (fish farms, pond-based aquaculture) in order to protect farms (from VHS, IHN, KHV) and facilitate trade.

Monitored population
Farmed and ornamental fish.

Surveillance procedures
- Outbreak surveillance
  > *Declaration of any suspicion or confirmation to the DDecPP (or the DDAAF in French overseas departments), in the event of abnormal mortality or observation of clinical signs.*
  > *Samples taken for first-line analysis by one of the seven accredited laboratories and, if necessary, confirmation analysis by the NRL at ANSES Ploufragan-Plouzané (identification of the virus by cellular or molecular methods).*
- Programmed surveillance

Since 2011, outbreak surveillance has been supplemented by the implementation of an animal health authorisation for aquaculture farms. This authorisation, which is mandatory for fish farms, is granted by the relevant local authority (DDecPP or DDAAF). It requires that the person responsible for the aquaculture farm undertake a risk analysis and draw up a corresponding health surveillance scheme for the regulated diseases. Clinical inspections by an accredited veterinarian and audits by the relevant authority are scheduled at a frequency depending on the level of risk of the aquaculture farm. Samples are taken in the event of suspicion.

- Programmes to achieve disease-free status for fish-farming zones and compartments (voluntary)

A programme for obtaining "disease-free" status by a single farm or a larger area including several farms and natural fishing areas can be set up voluntarily by professionals on the basis of the provisions of the EU regulations. The farmer may choose either a short programme with intense sampling (two clinical inspections and two batches of samples of 150 individuals each per year for two years), or a longer programme with less intense sampling (two clinical inspections and two batches of samples of 30 individuals each per year for four years). Currently, in France, these programmes only concern VHS and IHN. The list of aquaculture zones and compartments declared free of VHS and/or IHN can be consulted on the website of the MAAF (http://agriculture.gouv.fr/maladies-des-animaux-aquatiques).

Animal health rules
If an outbreak of a regulated disease is detected, health control measures are implemented (in compliance with Directive 2006/88/EC, transposed into French law by the Order of 4 November 2008). In the event of suspicion, the DDecPP or the DDAAF issues an APMS (prefectural monitoring order). If infection is confirmed by the accredited laboratory and/or the NRL, the infected fish farm is placed under an APDI (prefectural declaration of infection), with measures for eliminating dead fish, slaughtering animals presenting clinical signs, and draining, cleaning and disinfecting ponds. An epidemiological investigation is also carried out.

Regulations
- Council Directive 2006/88/EC of 24 October 2006 on the animal health conditions applicable to animals and products in the aquaculture sector and the prevention of certain diseases in aquatic animals and the measures for combating these diseases.
- Order of 4 November 2008, on the health control measures applicable to animals and products in the aquaculture sector and the prevention of certain diseases in aquatic animals and the measures for combating these diseases.
- Order of 8 June 2006 amended, on the animal health qualification or authorisation of primary production units, or businesses placing on the market products of animal origin or foodstuffs containing products of animal origin.

pond in the Pas-de-Calais *département*, following the observation of abnormal mortality in carp. The epidemiological investigation carried out by the DDecPP concluded on the hypothesis of a contamination by koi carp imported from Belgium.

Origin of the clinical suspicions
Both outbreaks of regulated diseases came to light as a result of outbreak surveillance. Abnormal mortality was observed by the veterinary practitioner in the case of VHS and by the fish farmer in the case of KHV.

Declaration of fish farms as "VHS and IHN free"

No additional fish farms were declared as disease-free in 2012. On 31 December 2012, 385 fish farms were free of VHS and IHN, out of a total of 621 freshwater aquaculture sites identified in 2008 (Agreste, 2011), to which should be added a large number of ponds (several tens of thousands).

Cost

In the 81 *départements* for which data are available, €1,511 were spent in 2012 under the surveillance scheme to finance visits related to an outbreak (veterinarians' fees and analysis costs) and €8,701 to finance visits related to achieving and maintaining the disease-free status of fish farms, including €7,095 in analysis costs. The cost of health control measures reached a total of €41,116 (compensation for slaughter, disinfection and rendering). The total cost to the national budget of all these operations was €51,327 in 2012.

Discussion

The number of outbreaks of regulated fish diseases reported has declined since 2001 (Figure 1) regarding VHS and IHN. This positive trend is probably the result of control measures associated with the implementation of programmes to achieve or maintain disease-free status over the last 15 years and the introduction of aquaculture production business authorisation. It is nonetheless probable that some outbreaks went unreported. IHN, in particular, can be difficult to detect as it is frequenlty associated to low mortality affecting only juveniles. The small number of veterinary practitioners specialising in aquaculture, the lack of awareness among certain fish professionals and amateurs, as well as the lack of compensations for the value of lost fish for farmers not participating in a programme to achieve disease-free status tend to amplify the under-reporting of outbreaks. The generalisation of aquaculture production business authorisation and the deployment of the corresponding inspection schemes should progressively improve detection rates when regulated diseases are suspected.

KHV had been detected sporadically in France in 2001 and 2002, but recurrent outbreaks have been reported since 2008, suggesting either that the virus has become established in France or, and this is more likely, that infected animals have been introduced repeatedly. This disease should henceforth be monitored more closely.

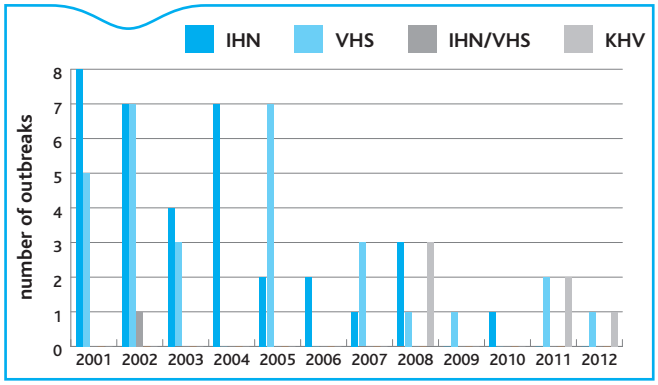

Figure 1. Changes in number of outbreaks of regulated fish diseases reported since 2001

OBITUARY

Pierre de Kinkelin passed away on 10 May 2013.

He was Secretary General of the Permanent Commission of the OIE for fish diseases, Director of the INRA's Ichthyopathology Laboratory in Jouy-en-Josas, supervised multiple PhD students, and held many other titles. He was the author of Précis de Pathologie des Poissons, the standard reference book for all those working in aquaculture, and never ceased updating and republishing it as a legacy to future generations. Over the course of his career, he trained several generations of researchers in the field of fish diseases, in which he was long a leading figure, and remained so even after retiring.

It is now up to us to continue in the path he mapped out for us with such energy and enthusiasm.

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Report of surveillance of **bee diseases** and **disorders** in 2012

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Abstract

Surveillance of regulated bee diseases concerns diseases present in France such as American foulbrood, nosemosis caused by *Nosema apis*, and varroasis, and two exotic pathogens, *Tropilaelaps spp.* and *Aethina tumida*. It is closely linked to the surveillance scheme for bee disorders set up in 2002 to deal with cases of acute bee mortality where poisoning by plant protection products is suspected. Despite the numerous limitations of the various surveillance components, the results confirm previous results when it comes to the strong suspicion that American foulbrood, and varroasis are enzootic forms, and also show that *Tropilaelaps spp.* and *Aethina tumida* are absent in France.

Keywords

American foulbrood, Nosemosis, *Tropilaelaps*, *Aethina*, mortality, depopulation, bees, surveillance

Résumé

Bilan de la surveillance des maladies et troubles des abeilles sur l'année 2012

La surveillance des maladies réglementées des abeilles concerne des maladies présentes en France telles que la loque américaine, la nosémose à* Nosema apis*, la varroose, ainsi que les deux agents pathogènes exotiques que sont* Tropilaelaps *spp. et* Aethina tumida*. Elle est étroitement liée au dispositif de surveillance des troubles des abeilles mis en place en 2002 pour traiter les cas de mortalités aiguës d'abeilles avec suspicion d'intoxication phytosanitaire. Malgré plusieurs limites aux différentes modalités de surveillance, les résultats confortent ceux des années précédentes concernant la forte suspicion de circulation sous forme enzootique de la loque américaine et de la varroose, et confirment l'absence de* Tropilaelaps *spp. et* Aethina tumida *sur le territoire.

Mots clés

Loque américaine, nosémose,* Tropilaelaps, Aethina, *mortalité, dépopulation, abeilles, surveillance

The surveillance of bee diseases involves four diseases formerly designated as «notifiable animal diseases with compulsory control measures»: American foulbrood, nosemosis (caused by Nosema apis), the small hive beetle (Aethina tumida) and Tropilaelaps spp. mites. These diseases are currently classified as Category 1 health hazards, in accordance with Decree No 2012-845 of 30 June 2012. Varroasis, formerly defined as a notifiable disease (ND), is now considered a Category 2 health hazard. American foulbrood and two exotic pathogens (A. tumida and Tropilaelaps spp.) are regulated diseases in the European Union also, as defined in Commission Regulation (EU) No 206/2010 and Directive 92/65/EEC (Table 1).

Surveillance programme (Box)

Surveillance programme for regulated diseases

Any clinical suspicion for one of the five regulated diseases must be notified to the DDecPP which will confirm or reject this suspicion and, if necessary, place the apiary under an APMS monitoring order until laboratory results are received. If the apiary has been confirmed to be infected with one of the regulated diseases, it is placed under APDI surveillance, in accordance with Ministerial Order of 11 August 1980 regarding the control of contagious bee diseases as amended by Ministerial Order of 23 December 2009.

Table 1. List of regulated bee diseases and their main features

Disease	Pathogen	Classification	Regulations	Status in mainland France
Varroasis	*Varroa destructor*	Mite	Category 2 health hazard	Present
Nosemosis	*Nosema apis*	Microsporidian	Category 1 health hazard	Present
American foulbrood	*Paenibacillus larvae*	Bacterium	Category 1 health hazard Directive 92/65/EEC	Present
Small hive beetle	*Aethina tumida*	Insect	Category 1 health hazard Directive 92/65/EEC	Absent
Tropilaelaps spp. mites	*Tropilaelaps spp.*	Mite	Category 1 health hazard Directive 92/65/EEC	Absent

The various field visits to apiaries as part of the surveillance programme or in compliance with animal health rules are carried out by DDecPP staff or bee health inspectors appointed by prefectural order and authorised to carry out specific surveillance missions.

The effectiveness of the surveillance programme depends strongly on the active participation of local beekeepers in the clinical surveillance of regulated diseases. Therefore, the results vary greatly by *département* and it is difficult to compare them at national level.

DGAL-accredited laboratories participate by carrying out analyses when there is a suspected case of an outbreak of a regulated disease. In October 2012, a network of eight accredited departmental laboratories was set up to diagnose American foulbrood and nosemosis (Memorandum DGAL/SDPRAT/N2012-8199 of 10 October 2012). This network supplements the network of laboratories accredited in 2011 for determining the risk of introducing the small hive beetle and *Tropilaelaps* mites *via* imported queen bees or drones (Memorandum DGAL/SDPRAT/N2011-8128 of 8 June 2011). Moreover, six laboratories have also been accredited to carry out analyses specifically for the European pilot programme for bee epidemiological surveillance that started in the autumn of 2012 (see description of EPILOBEE below).

The official analytical methods for diagnosis of regulated diseases were defined during the accreditation process. The NRL for bee diseases is in charge of organising and coordinating the various laboratory networks and training sessions. It also carries out second-line analyses and, in particular, identifies any suspected exotic pests.

Surveillance programme for massive bee deaths for which pesticide poisoning is suspected

This surveillance is carried out as part of the national network for surveillance of bee disorders set up in 2002 following the reported increase in colony collapse and massive bee losses (Memorandum DGAL/SDQPV/SDSPA/N2002-8110 of 2 August 2002). Accordingly, beekeepers are urged to report any problem or incident observed in their bee colonies to the DDecPP. Upon notification by a beekeeper, an epidemiological investigation is conducted by the staff of the DDecPP, sometimes in conjunction with the SRAL, the regional body in charge of plant protection, as well as the BNEVP.

Analyses to screen for pesticide residues are carried out in various laboratories, according to their testing capacities. The surveillance protocol does not specify which diagnosis methods to use to screen for toxic substances and, to date, there are no officially accredited laboratories for pesticide screening.

Pilot network for bee epidemiological surveillance (Resabeille/EPILOBEE)

In 2011, the global increase in massive bee deaths and colony collapse led the European Commission to launch a call for applications to set up a network for the surveillance of bee diseases and colony loss in Member States. Thus, the pilot bee epidemiological surveillance programme (Resabeille) was set up in 2012 in six French *départements*: Cantal, Drôme, Haut-Rhin, Bouches-du-Rhône, Indre-et-Loire and Finistère.

Locally coordinated by the respective DDecPPs, it involves bee health protection associations (GDSA), animal health protection Farmers' organization(GDS), departmental testing laboratories, bee health inspectors and beekeepers.

In each *département*, 66 randomly selected apiaries are monitored by trained health protection technicians. These apiaries are visited three times: before and after overwintering, and during the beekeeping season. At each visit, a random sample of colonies is examined and samples are taken to assess the level of colony infestation with *V. destructor*, to screen for *Nosema* spp. and to diagnose the main bee diseases in colonies that show symptoms. The results of this pilot programme were due to be compiled in 2013 and will be published in an upcoming issue of the Bulletin Épidémiologique. The review of the bee health surveillance given in this article therefore does not include these results.

Annual apiary registration

In 2010, annual apiary registration was reactivated in accordance with Ministerial Order of 11 August 1980 as amended. In 2011, the "Téléruchers" application was set up on the Ministry of Agriculture, Food and Forestry (MAAF)'s website to allow beekeepers to declare their apiaries on-line. Alternatively, beekeepers who so desire can submit their paper registration form to their local GDS office, who will enter the data in the TéléRuchers database. In 2011, 30,416 registrations were received for 59,493 apiaries and 814,750 hives. Among these registrations, 10% (n = 3013) were filed on-line by the beekeeper and 90% (n = 27,242) were entered by the local GDSs. In 2012, 30,542 registrations were received for a total of 61,024 apiaries and 899,886 hives. Although slightly more apiaries and hives were declared in 2012 than in 2011, there is still under-reporting due in part to the difficulty in setting up the new electronic database and, for certain beekeepers, in using the Téléruchers application.

In parallel, the DDecPPs continue to oversee bee health using the data compiled in the SIGAL database. Thus, APMS and APDI surveillance orders and the review of health inspections are based on the official data recorded in SIGAL.

Results

The results given below were compiled from the data sent by 80 DDecPPs and DAAFs, out of the 100 who were contacted.

Health inspections

The *départements* reported 932 active bee health inspectors, or approximately 11 inspectors for each *département*. This average masks a large discrepancy among *départements*, because 15 of them do not have any inspectors, whereas others have up to 80 bee health inspectors. In the *départements* who responded, bee health inspectors carried out 2,294 health inspections. DDecPP staff carried out 252 visits.

Results from clinical surveillance of American foulbrood and nosemosis

American foulbrood surveillance.

In the 80 *départements* that provided data, there were 232 clinical suspicions of American foulbrood in 2012. An APMS order for the apiary was issued for 28 of these cases, i.e. 12%.

Of the suspected cases, 97 (i.e. 42% of suspicions) new outbreaks of American foulbrood were confirmed, of which almost all were confirmed by laboratory testing (n=96 and one confirmation based on symptoms), and APDI orders were issued by the respective DDecPPs (Table 2).

Table 2. Annual number of suspected cases and confirmed outbreaks of American foulbrood since 2010

American foulbrood	2010	2011	2012
Clinical suspicions	348	290	232
Confirmed outbreaks (APDI)	95	121	97

The number of outbreaks confirmed by an APDI (97) was much greater than the number of outbreaks for which an APMS order was issued (28). This difference was attributed to the fact that many apiaries were directly placed under an APDI order without a prior APMS order.

Overall, 227 APDI orders were in effect on 1 January 2012 and 253 were in effect on 31 December 2012.

Detection of suspected cases of American foulbrood

Information on how the suspected case was reported is available for 194 cases out of 232. In total, 22% (n=43) of the clinical suspicions were based on notifications by beekeepers and 72% (n=140) were notified following random inspections carried out by bee health inspectors mandated by the DDecPPs (Figure 1). Lastly, 6% (n=11) were notified upon other visits to apiaries, e.g. visits related to health certification, visits following notification of massive bee deaths or colony collapse, or visits in the 3 km protection zone around an American foulbrood or nosemosis outbreak.

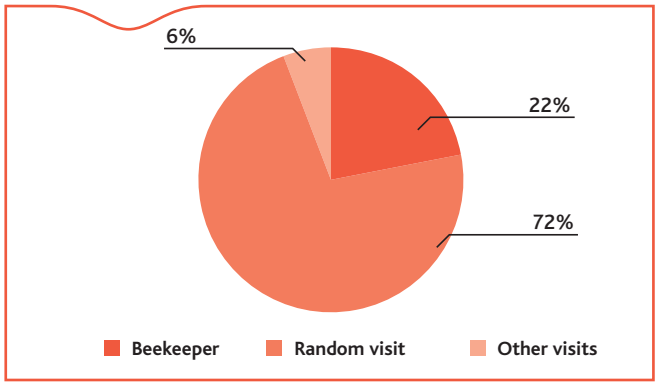

Figure 1. Detection of suspected cases of American foulbrood in 2012

Of the 97 confirmed outbreaks of American foulbrood, one-quarter were notified by beekeepers based on clinical suspicion (n=24), the other outbreaks were detected during visits mandated by DDecPPs, including visits in the surveillance zone following confirmation of a disease outbreak (Figure 2).

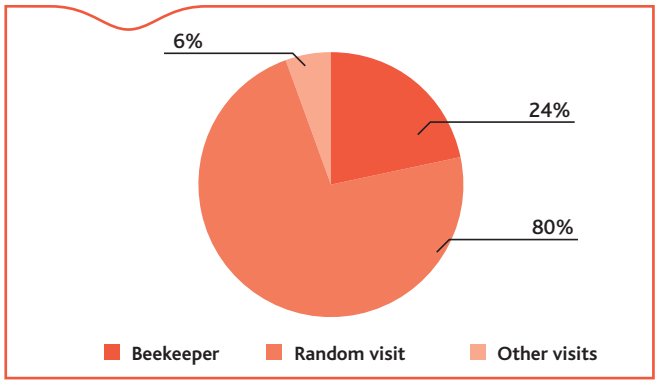

Figure 2. Origin of confirmed American foulbrood outbreaks in 2012

Nosemosis (caused by N. apis) surveillance

For the 80 *départements* for which information was available, 25 clinical suspicions of *N. apis* nosemosis were reported in 2012. Only one apiary was placed under APMS.

During 2012, two *N. apis* nosemosis outbreaks were confirmed, i.e. 8% of the 25 initial suspected cases (Table 3). Two APDI orders were issued by DDecPPs following confirmatory analyses undertaken in a laboratory.

Table 3. Annual number of suspected cases and confirmed outbreaks of nosemosis since 2010

Nosémose à *N. apis*	2010	2011	2012
Clinical suspicions	64	43	25
Confirmed outbreaks (APDI)	7	5	2

Detection of suspected clinical cases of *N. apis* nosemosis

Based on the available information, suspicions of nosemosis were reported equally by beekeepers (n=5) and by mandated bee health inspectors (n=4), or during investigations in surveillance zones or during visits for obtaining a health certificate (n=2).

Results from surveillance of the small hive beetle (A. tumida) and Tropilaelaps mites

Surveillance of these two exotic pests is carried out on two levels: on queen bees imported from non-EU countries and on any suspected case in local apiaries.

During 2012, as part of regulatory surveillance for queen bees imported from non-EU countries, the NRL received a sample suspected to be infected with the small hive beetle for identification. The analysis of the specimen was negative.

In addition, the departmental laboratories were requested to detect specimens of suspect insects and mites using the analyses commonly carried out for diagnosing bee diseases and for the DGAL epidemiological surveillance programmes, on brood samples or bees from French apiaries. Five suspicious specimens were examined (one larva, two beetles, one mite, one non-identified parasite). The parasite identifications of *A. tumida* and *Tropilaelaps* spp. carried out as second-line tests by the NRL for bee diseases were all negative.

Results from clinical surveillance of varroasis In all, 63 *départements* did not declare any varroasis outbreaks and 17 *départements* declared at least one outbreak. Of a total of 1,426 declared outbreaks, there was significant variation among *départements*, ranging from 1 to 1270 declared outbreaks. These figures are lower than those of 2011 (with 2809 outbreaks) and highlight under-reporting of the disease by beekeepers, given that the *V. destructor* parasite is now widespread throughout France.

Results from surveillance of massive bee deaths

In 2012, 105 notifications of massive bee deaths were recorded in 36 *départements*. Although these figures come from passive surveillance, they nevertheless indicate that some beekeepers are aware of the usefulness of reporting massive bee deaths that may be linked to plant protection products.

Investigations of these 105 cases showed that some required activating the whole bee health network (SRAL, DDecPP, BNEVP) and other cases were handled locally by the DDecPP when there was no clear link with pesticide poisoning. Thus, the SRAL in conjunction with interministerial departmental services, worked on 33 cases, of which 23 called for pesticide testing. The BNEVP was involved in only 11 cases.

Toxicological analysis results were positive in 13 cases, i.e. 12% of the investigated cases. In all, 18 different chemical substances were identified (17 pesticides and 1 herbicide). Nevertheless, the link

between the presence of these substances and massive bee deaths was clearly established for only seven cases, i.e. just over 6% of the suspected cases.

The chemical substances identified as the potential cause of poisoning in these seven cases were acetamiprid and cyprodinil, glyphosate, parathion-methyl, carbaryl, triazole and a pyrethroid.

Of these cases, one prohibited substance (carbaryl) was detected. Legal proceedings are currently in progress for this case.

The outcome of the investigations for the other suspected cases of pesticide poisoning was related to either poor beekeeping practices (improper use of anti-varroa treatments, cold brood, famine, over-wintering deaths, etc.), or to the presence of pathogens that could account for the high mortality rate in the visited colonies. The confirmed diseases observed include chronic bee paralysis (disease caused by the chronic bee paralysis virus, CBPV), high *Varroa* infestation with the concomitant presence of CBPV and one case of American foulbrood. In addition, for nine cases, investigators could not identify any specific cause.

Regarding colony collapse, 47 notifications were recorded in 2012 in the 79 *départements* for which data are available. A colony collapse is defined as the abrupt disappearance of bees from the colony, not associated with any dead bees near the hive. This type of observation was more common in the first to second quarter (19 and 20 cases, respectively) compared to the third (n=9) and fourth quarters (n=1) of the year.

Cost

In 2012, 29 *départements* invested funds in bee health inspections mandated following notification by beekeepers, for a total of €5,870.

In addition, *départements* spent funds on random or targeted visits ordered by the DDecPPs. The total cost of bee health inspections nationally was €106,315 and the average cost per *département* was €1,346 (ranging from €9 to €45,000 according to *département*).

The analyses carried out as part of these inspections cost €34,828 (for an average of €1,088 per *département*, n=32) and €3,165 (for an average of €395 per *département*, n=8) for pathological and toxicological analyses, respectively.

The total expenditure (€150,135) for bee health management and monitoring (visits, certificates, analyses) was higher than in 2011.

Discussion

The analysis of the results from the surveillance programme for bee diseases and disorders obtained in 2012 must be considered taking into account some of the limits and particularities of the surveillance programme.

The passive and active surveillance programmes for bee diseases and disorders are based on clinical surveillance. They cannot detect apiaries that are asymptomatically infected. The apiaries visited as part of the active surveillance programme are not pre-selected, ensuring representativeness of all the apiaries in a given *département* (e.g. chosen randomly). In addition to the difficulties involving the registration of beekeepers and their apiaries (into a separate database, Télérucher, not linked with SIGAL), the number and frequency of these 'random' visits also vary by *département*.

Regarding American foulbrood, nearly all outbreaks were confirmed by the surveillance programme in 2012 (96 out of the 97 suspected cases), continuing the improvement noted in 2011 (96%), compared to 52% in 2010. Nevertheless, there has been a decrease in the number of suspected cases over time (348 in 2010, 290 in 2011 and 232 in 2012), despite the fact that the total number of confirmed outbreaks was stable over these three years. Moreover, the surveillance programme results compiled since 2006 show a discrepancy in the reporting of American foulbrood outbreaks among *départements* (Bronner *et al.*, 2011, Papin *et al.*, 2012). Given the enzootic nature of the suspected disease in most regions, it would be helpful to establish the factors that favour the detection of outbreaks only in certain parts of the country.

This observation suggests that the likely causes behind these differences include the absence of a local bee health network; low frequency of beekeeper registrations and bee health inspections; difficulties encountered by certain DDecPPs to obtain and sustain human resources (available, trained personnel, motivation of those involved in the network); the level of financial compensation for outbreaks and diseases; inappropriate animal health rules for certain epidemiological situations; bias in declarations where only 'real' suspicions (confirmed outbreaks) are recorded.

The second lesson that can be drawn from these results that confirm under-reporting is that only around 20% of suspected cases are reported by beekeepers, whereas 60% are reported after visits carried out during epidemiological investigations (e.g. in the surveillance zone set up following an outbreak).

Regarding (*N. apis*) nosemosis, both outbreaks were confirmed by a laboratory in 2012. This diagnosis is necessary because the clinical signs of nosemosis are not specific and can be confused with those of other diseases. Likewise, as part of the surveillance programme set up in Drôme, regular sampling of 60 bees was carried out just after winter on 283 randomly chosen colonies. They were analysed to detect and identify *Nosema* spores. The results show that *N. apis* was not prevalent in apiaries (no colony was deemed infected). The infection rate of apiaries by N. ceranae (another *Nosema* species that is not regulated) was high (between 32% and 64% of apiaries, 95% confidence interval) (Dominguez *et al.*, 2013). This pathogen has been present for several years in many European countries, particularly in Southern Europe (Chauzat *et al.*, 2007; Fries, 2010), but its pathogenicity is not well known. Scientific studies have shown its effect in terms of bee deaths and colony weakening, possibly in synergy with other factors (Botias, 2013; Fries, 2010; Higes *et al.*, 2008, 2013; Botias *et al.*, 2013). A more detailed analysis of the infection status could help to guide nosemosis surveillance strategy in France (i.e. particularly in terms of improving efficacy and in adapting the surveillance programme).

Likewise, the number of reported cases of varroasis is probably much lower than the number of actual cases. There has been a decrease in reports between 2011 and 2012 (2809 compared to 1426 cases). Since it was introduced in 1982, *V. destructor* has now spread throughout France and causes (direct and indirect) damage to bee colonies. Cases of varroasis, very frequent, are rarely notified to the DDecPPs. This under-reporting can be attributed to the absence of any specific individual or collective measures taken after notification. The expected advantages of notification and management of this disease by the public authorities should be evaluated and updated, in regard to the many voluntary control measures set up in different *départements* and sponsored by animal health programmes (PSE) for many years without any conclusive results.

Regarding the exotic pests *Tropilaelaps* mites and *A. tumida*, as in 2011, no official suspected infestation was reported in 2012 *via* the DDecPPs. There were some rare suspected cases, but they were not confirmed after laboratory analyses and no warnings were issued. This observation suggests that the capacity for early detection of these two exotic pathogens is inadequate. Only a targeted inspection of imported queen bees is currently in effect when they are brought into France, which is insufficient for early detection. The absence of any outbreak of *Tropilaelaps* mites or *A. tumida* lends support to the disease-free status of France with respect to these two pathogens, although vigilance must be maintained.

Notification of massive bee deaths and colony collapse

The results of notifications of massive bee deaths or colony collapse also need to be interpreted with caution, given the absence of mandatory notification and no clear definition of these two types of

disorders, the lack of resources deployed and of practical opportunities to study and better understand this disorder and analyse its causes and, lastly, the inherent biases in the notification process. Reports are generally sporadic, disparate and irregular. They depend on the weather and beekeeping conditions of the moment, the availability of beekeepers, the frequency of apiary visits, and the time and resources invested in field visits. The investigations conducted generally show the concomitant presence of chemical contaminants and pathogens in apiaries, although it is not possible to conclude, given the current state of knowledge, as to the causes of this disorder and any direct causality relationship between chemical contaminants, pathogens and bee deaths. In any case, the results obtained following field investigations must be tested in an appropriate experimental setting to study and confirm the effects objectively.

The analysis of the 2012 data shows that the surveillance network for bee deaths is better known and operates more effectively, as attested by the increasing reports to the network.

Outlook

The limits of the French passive surveillance programme have already been described by EFSA in 2009. Its expert report "Bee mortality and bee surveillance in Europe" indicates that, generally speaking, the surveillance systems set up in the European Union are not very effective and that the data available in individual Member States and comparable data across the EU are insufficient (EFSA, 2009).

Given this observation of an overall increase in massive bee deaths and colony collapses and the weaknesses of the established surveillance programmes, the European Commission decided to set up a surveillance programme in 17 Member States (EPILOBEE programme). This harmonised programme, based on the appropriate bee health techniques and co-funded by the Commission, was deployed in 2012.

This surveillance programme will help to estimate the prevalence of the main bee diseases and disorders and in particular estimate the over-wintering and in-season mortality rates of colonies. The protocol designed for application across Europe and adapted, to a certain extent by each Member State will make it possible to compare surveillance results among MSs on a national level and across Europe by ensuring standardisation and harmonisation of the various data collection steps in the different French *départements* and Member States. The results on the bee colony losses will be compiled for 2013 in an upcoming issue of the Bulletin Épidémiologique.

This surveillance programme can be enhanced by further involvement and awareness on the part of all involved: beekeepers, breeders who import queen bees or drones, animal health organisations, veterinary organisations, public authorities, etc., to encourage beekeepers to register their apiaries, to declare suspicions to the DDecPPs and to train and motivate officials who inspect imported bees and carry out apiary visits.

To ensure the quality of the obtained results, the surveillance programmes must rely on DGAL-accredited laboratories that all use the same officially defined and validated diagnostic methods. In 2011 and 2012, several laboratory networks were set up in conjunction with the NRL for bee diseases, to help to enhance this part of bee health surveillance. A similar approach would be relevant for screening for chemical residues.

Lastly, active surveillance is currently limited and should be revamped and harmonised on the national level so that collected data can be compared to determine the prevalence and evolution of regulated diseases. This weakness could be overcome by the improved animal health governance plan slated for 2013. In parallel, a risk analysis would help to define and improve the national strategy on health hazards (review of disease classification and categories, animal health measures, monitoring tools, etc.).

Box. Surveillance programme and control measures for regulated bee diseases and disorders

Objectives of the surveillance programme
- To detect, as early as possible outbreaks of American foulbrood and *Nosema apis* nosemosis to prevent the spread of these two pathogens in France.
- To ensure early detection of any introduction of two exotic pathogens, *Aethina tumida* and *Tropilaelaps* spp., in France and guarantee the country's pest-free status for trade purposes.
- To monitor changes in prevalence of varroasis in the French bee population.
- To identify any abnormal or massive bee deaths related to the use of plant protection products.
- To evaluate the over-wintering mortality rate and the prevalence rate of major bee diseases as part of the pilot network in place in six French *départements* in 2012.

Monitored population
Honeybee (*Apis mellifera*) colonies throughout France.

Surveillance procedures
- Surveillance événementielle

Regulated diseases
> *Mandatory notification by beekeepers to the local DDecPP of any clinical suspicion of one of the five regulated diseases (American foulbrood, N. apis nosemosis, varroasis, as well as two exotic pathogens: Tropilaelaps spp. and A. tumida).*
> *Sampling for confirmatory laboratory analyses.*

Colony mortalities
> *Reports by beekeepers to the DDecPP of any incident (mortality, weakening) in an apiary.*
> *The DDecPP focuses its action on cases of "massive bee deaths that occur in spring, summer and autumn" and carries out investigations coordinated with DRAAFs (via the SRAL, the regional body in charge of plant protection), and, sometimes, the BNEVP.*
> Programmed surveillance

Random national surveillance
Random inspections planned for the département by each DDecPP. The scheduling of random visits (number, how apiaries are chosen, etc.) is done by each DDecPP. Therefore, the number and the frequency of these 'random' visits can vary by *département*.

Targeted surveillance
For queen bees and drones imported from non-EU countries, surveillance involves thorough laboratory examination of transport cages and the accompanying bees to detect the A. tumida hive beetle and *Tropilaelaps* mites in accordance with Regulation (EU) No 206/2010.

Pilot network for bee epidemiological surveillance (Memorandum DGAl/SDSPA/N2012-8211)
> *programme initiated in the Drôme département in 2011 and extended to five other départements in the autumn of 2012.*
> *In each of the six départements of the pilot network, 66 apiaries were monitored, totalling 396 apiaries.*
> *Three visits were scheduled in this programme (before over-wintering, after over-wintering and during the beekeeping season): clinical examination of a random sample of colonies, sampling to assess the level of Varroa infestation, screening for N. apis and Nosema ceranae and diagnosis of the main bee diseases on symptomatic colonies. These visits also serve to estimate the over-winter and production season colony mortality rates.*

Animal health control
> *Clinical suspicions: apiary placed under APMS monitoring order.*
> *Implementation of an epidemiological investigation.*
> *Laboratory confirmation: apiary placed under APDI surveillance with, according to the case, implementation of measures such as containment, destruction of infected colonies, destruction or disinfection of material*

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Regulations

Commission Regulation (EU) No 206/2010 of 12 March 2010 laying down lists of third countries, territories or parts thereof authorised for the introduction into the European Union of certain animals and fresh meat and the veterinary certification requirements

Council Directive 92/65/EEC of 13 July 1992 laying down animal health requirements governing trade in and imports into the Community of animals, semen, ova and embryos not subject to animal health requirements laid down in specific Community rules referred to in Annex A(I) to Directive 90/425/EEC

Ministerial Order of 11 August 1980 regarding the control of contagious bee diseases amended by Ministerial Order of 23 December 2009

Commission Implementing Decision of 4 July 2012 concerning a financial contribution by the Union to certain Member States to support voluntary surveillance studies on honeybee colony losses. Memorandum DGAL/SDSPA/SDQPV/N2011-8101 of 26 April 2011 regarding the surveillance network for bee disorders.

Summary Table of regulated diseases in France

	Disease	Farms (animals) investigated	Farms (animals) with non-negative results	Clinical/ Pathological suspicions	Incident outbreaks 2012* (%)	Evolution of incidence 2012-2011	Prevalence 2012 * (%)
Cattle	Bovine tuberculosis	14,722 (678,085)	1,345 (2,531)	171	116 (0.05%)	20	169 (0.075%)
	Bovine brucellosis	127,499	233	70,853	2 (<0.001%)	2	2 (<0.001%)
	Enzootic bovine leucosis	46,638	158	3	2 (<0.001%)	2	2 (<0.001%)
	Infectious bovine rhinotracheitis	177,502	19,011	0	3,465 (1.9%)	+ 2,157	19,011 (10.7%)
	Hypodermosis (Warble flies)	9,513	8	0	0	0	0
	Bovine Spongiform Encephalopathy	(1,240,941)	(5)		1 (atypical L)	-2	(0 for BSE C)
	Anthrax	NA	NA	12	0	0	Undetermined
	Foot-and-mouth disease	NA	NA	4	0	0	Undetermined
	Bluetongue	(99,572)	(173)	101	0	0	0
Ruminants	Brucellosis in small ruminants	47,970 (1,591,114)	620 (18,498)	4,643	0	0	0
	Classical scrapie in sheep	(11,923 animals slaughterhouse) (40,988 animals rendering)	"1 sheep slaughterhouse,	0	4	2	0% slaughterhouse 0.009% rendering
	Classical scrapie in goats	(10,470 animals slaughterhouse) (55,909 animals rendering)	0 goat slaughterhouse, 2 goats rendering	0	2	2	0% slaughterhouse, 0.004% rendering
	Atypical scrapie in sheep	(11,923 animals slaughterhouse) (40,988 animals rendering)	4 sheep slaughterhouse, 18 sheep rendering	0	22	-2	0.034% slaughterhouse, 0.044% rendering
	Atypical scrapie in goats	(10,470 animals slaughterhouse) (55,909 animals rendering)	1 goat slaughterhouse, 4 goats rendering	0	5	-1	0.01% slaughterhouse, 0.007% rendering
Pigs	Classical swine fever	2,236 (18,165)	17 (152)	1	0	0	0
	Aujeszky's Disease	1,639	18	1	0	0	0
	Porcine brucellosis	NA	NA				

* Herds for which a Prefectural declaration of infection (APDI) was issued in 2012
NA: Not Applicable
1: Number of pools of rectal swabs positive in PCR M
2: Number of wild birds found dead and analysed
3: No outbreak of Newcastle disease in poultry but 10 cases of paramyxovirus of pigeons in captive pigeons

	Disease	Farms (animals) investigated	Farms (animals) with non-negative results	Clinical/ Pathological suspicions	Incident outbreaks 2012* (%)	Evolution of incidence 2012-2011	Prevalence 2012 * (%)
Poultry	Avian influenza in poultry	898	17	2	0	0	0
	HP avian influenza in wild birds	NA	NA	49 (1)	0	0	0
	HP avian influenza in decoy ducks	NA	NA	0	0	0	0
	Newcastle disease in poultry	NA	NA	16	[0]	0	0
	Pullorum disease and fowl typhoid	NA	NA	0	0	-4	0
	Salmonella in breeding flocks of broilers and layers in *Gallus gallus* (during the rearing and laying periods)	4,034	11	NA	11	0.08%	0.27%
	Salmonella in flocks of *Gallus gallus* laying eggs for consumption (during the rearing and laying periods)	7,182	61	NA	61	-0.16%	0.85%
	Salmonella in breeding flocks of *Meleagris gallopavo* (during the rearing and laying periods)	1,517	5	NA	5	-0.11%	0.33%
	Salmonella in broiler flocks of Gallus gallus	64,563	328	NA	328	-0.11%	0.51%
Horses	Equine infectious anaemia	15,331	8	2	2	8	Undetermined
	Contagious equine metritis	Undetermined	2	0	NA	0	Undetermined
	Equine viral arteritis	578	3	0	3	3	Undetermined
Fish	IHN	NA	NA	0	0	0	
	VHS	NA	NA	1	1	-1	
	KHV	NA	NA	1	1	-1	
Bees	American foulbrood	NA	NA	232	97	-24 (-20%)	
	Nosemosis	NA	NA	25	2	-3 (-60%)	
Other animals	Rabies in carnivores	NA	NA		0		Undetermined
	Rabies in bats	NA	NA		5		Undetermined

* Herds for which a Prefectural declaration of infection (APDI) was issued in 2012
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1: Number of pools of rectal swabs positive in PCR M
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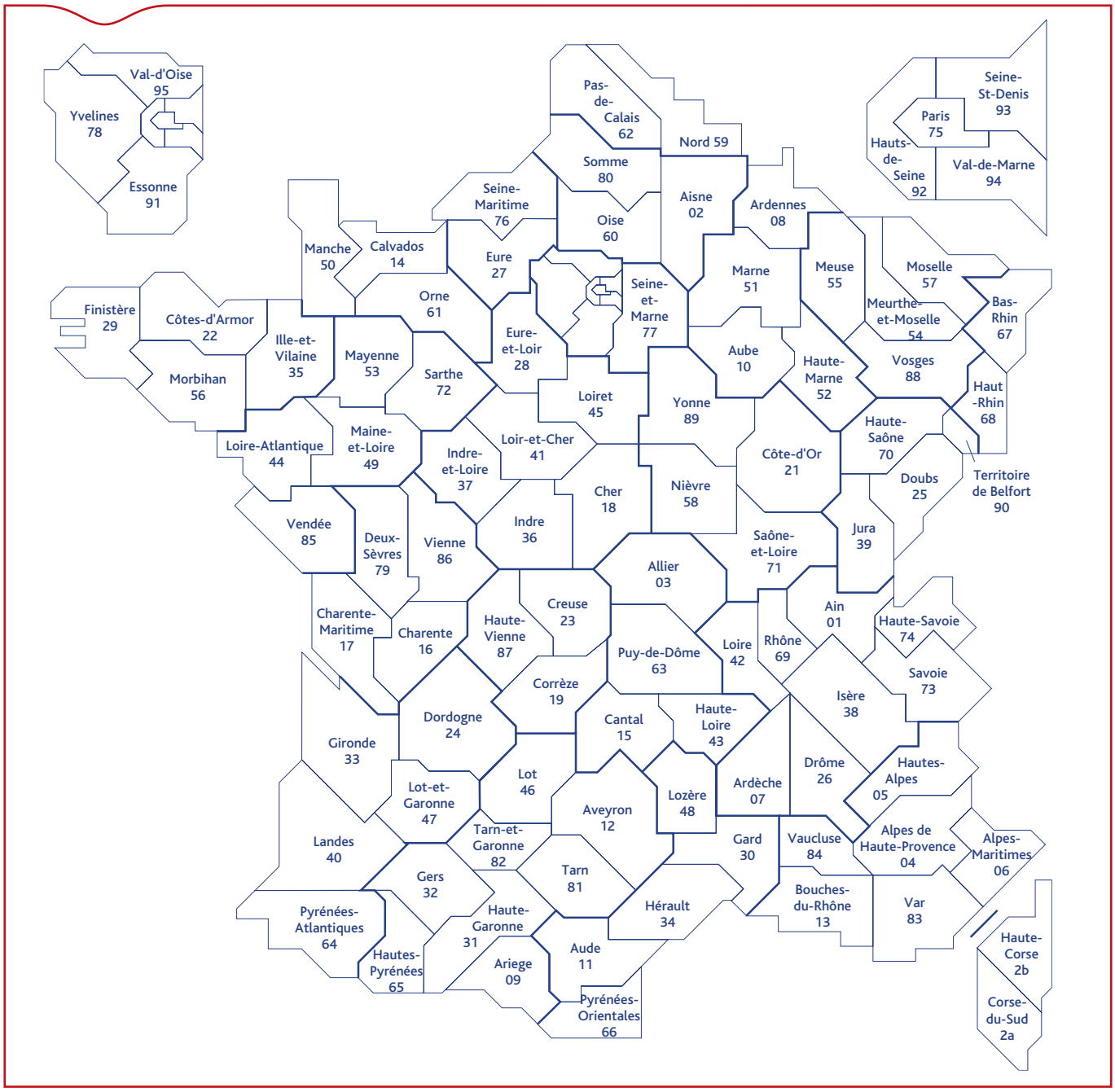
Reference map

The following map identifies the *départements* referred to, either by name or by number, in the different articles.



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