Report on animal rabies surveillance in France: 3 serotine bat cases detected in 2014

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Abstract
Since France was officially declared rabies-free in 2001, the disease continues to be reported in mainland France in illegally imported pets (dogs and cats) incubating rabies when entering the country, as well as in bats. Currently, the rabies surveillance network mainly concentrates on pets and bats. In 2014, no positive pets were reported. However, three new rabies cases were identified in serotine bats, bringing the total number of rabies cases detected in Chiroptera to 57 since 2001. The discovery of novel species of lyssavirus and the regular detection of rabid bats each year highlight the need to maintain and reinforce rabies surveillance in France.

Keywords
Surveillance, Rabies, Pets, Bats

Rabies is a viral zoonosis causing acute encephalomyelitis. It is caused by a virus of the Rabdoviridae family, Lyssavirus genus, which is currently thought to include fourteen species (ICTV, 2012). Found in the saliva of infected animals in the final phases of the disease, the virus is generally transmitted to another animal or to humans through biting. Rabies causes more than 55,000 human deaths each year around the world, according to estimates by the WHO (WHO, 2013). Different species of pets (mainly dogs, especially in Africa and Asia) or wild animals (for example foxes and bats) can maintain and transmit the lyssaviruses responsible for the disease. In France, rabies is a notifiable disease that must be reported to the OIE (OIE, 2012). It is recognised as a Category 1 health hazard (Ministerial Order of 29 July 2009). Metropolitan France has been officially declared rabies-free since 2001 (Ministerial Order of 30 April 2001), except for the period from February 2008 to February 2010 following the import of a rabid dog which led to secondary cases (Dacheux et al., 2008). The outbreak surveillance of rabies remains a topical issue in France, because of regular imports of pets incubating rabies and of rabid bats each year.

Results from Outbreak surveillance
In 2014, 1,839 animals were sent to the two laboratories for rabies diagnosis (Box). Of them, 29% (n=495) had no known history of human contamination and were sent to the ANSES NRL in Nancy. The other samples, i.e. 73% (n=1,344), were sent to the National Reference Centre for Rabies (NRC) at the Institut Pasteur in Paris (IPP). As every year, dogs and cats made up the majority of animal species diagnosed, with respectively 34% and 36% of the total (Table 1). Foxes accounted for only 2.3% (n=42) of the samples received by the two laboratories in 2014. The epidemiological surveillance network for rabies in bats, which was extended in 2000, continues to prove its worth, with Chiroptera representing a significant share (25%) of the animal species received for diagnosis of rabies and constituting, at nearly 88%, a large majority of the wild species investigated.

The geographical distribution (Figure 1) of animals received for diagnosis of rabies remains fairly homogeneous in metropolitan France, and also in the overseas départements (French Guiana, Reunion Island, Guadeloupe and Martinique).

Ninety-eight percent of the samples received (n=1,803) were analysed: One thousand eight hundred were diagnosed as negative and three were diagnosed as positive for rabies. These three cases of rabies were all detected in serotine bats in the Cher, Loir-et-Cher and Haute-Vienne départements.

Case of indigenous rabies in a bat in the Cher département
On 4 June 2014, a bat identified as a serotine bat was diagnosed by the NRL as positive for rabies using immunofluorescence. The diagnosis was confirmed on 6 June by cell infection and molecular biology techniques. Typing of the virus, through partial nucleoprotein gene and polymerase gene sequencing, showed that it was a lyssavirus of the European bat lyssavirus type 1 (EBLV-1) species, subtype b, very similar to the EBLV-1b viruses previously isolated in the Centre region. This virus has 99.2% homology with a viral strain previously isolated in Bourges (Cher département) in 2009.
Surveillance and health control measures concerning rabies

The French network for epidemiological surveillance of animal rabies was set up following the discovery of the first case of rabies in a fox on 28 March 1968.

**Objectives**
The primary objective of this network for outbreak surveillance is to enable early detection of the presence of a rabies infection by carrying out a diagnosis of any animals that are suspect (clinical signs suggestive of rabies, human contamination by a bite, scratch or licking on mucous membranes or damaged skin) or found dead without reason, so as to rule out rabies.

**Players in the surveillance programme**
The partners in the surveillance network call on specialists from the fields of health (coordinated by the Directorate General for Health), agriculture (coordinated by the General Directorate for Food), and the environment (coordinated by the Ministry of Ecology, Sustainable Development and Energy). The Chiroptera Group of the French Society for the Study and Protection of Mammals (SFEPM) plays a vital role in the collection of bat specimens (Picard-Meyer et al., 2013b).

**The population monitored**
As France is rabies-free, but nonetheless exposed due to the regular introduction of cases of imported rabies and the presence of rabies in bats, the primary objective of the epidemiological surveillance network is to monitor for rabies in pets (particularly biting dogs and cats) and wild animals (especially bats).

**Surveillance procedures**
**Pets:** This surveillance depends primarily on the presentation to the veterinary practitioner of animals suspected of rabies or animals that bite or scratch. A biting or scratching animal is defined as an “animal susceptible to rabies that, irrespective of where the incident occurred, has bitten or scratched someone” (Article R.223-25-5° of the CRPM) and must be placed under the supervision of a mandated veterinarian (Ministerial Order of 21 April 1997). Even if it has been properly vaccinated against rabies, a biting or scratching animal must be placed under veterinary surveillance, because while the protection conferred by anti-rabies vaccination is extremely high, it is not absolute. The surveillance period is statutorily set at fifteen days for biting or scratching pets and thirty days for wild animals that have been tamed or kept in captivity, taking into account the longer pre-symptomatic carrying period sometimes observed in certain species (Ministerial Order of 21 April 1997). During the surveillance period, the animal must be presented three times to the same mandated veterinarian. During the surveillance period, the animal may not be euthanised (except with the agreement of the veterinary services or in cases of force majeure), nor may it be vaccinated against rabies. In the event of the death or euthanasia of a biting or scratching animal during this period, a diagnosis of rabies must be carried out by the NRC.

**Wild carnivores:** It is recommended that anyone finding a wild animal dead, injured or sick should not handle it and should contact the veterinary services of the département concerned. The system for monitoring rabies in bats is based on an epidemiological surveillance network coordinated by the Nancy Laboratory for Rabies and Wildlife (ANSES) in partnership with the Chiroptera Group of the SFEPM, consisting of volunteers and veterinary practitioners. This network, which was strengthened in 2000, is an adaptation of the existing organisation for the epidemiological surveillance of animal rabies. The surveillance of rabbits in bats is based on the diagnosis of rabies in the corpses of bats found, most often, in an environment close to humans. Approximately 70% of the bats are sent by the network of chiropterologists, directly or via members of the public who contact the volunteers by calling their bat-rescue service (“SOS chauves-souris”), or the SFEPM’s Chiroptera Group (http://www.sfepm.org/groupeChiropteres.htm). Bats are a protected species in metropolitan France, so they may neither be killed, nor handled, nor transported, even after death, without official authorisation granted by the Ministry of Ecology.

**Diagnosis**
The French surveillance network sends samples to two laboratories. The NRC of the IPP is mobilised when human contamination is suspected, i.e. if at least one of the four following conditions is met:
- a bite resulting in broken skin,
- scratching,
- licking of damaged skin (broken or scratched skin),
- projection of saliva on mucous membranes.

If this is not the case, the samples are sent to the Nancy Laboratory for Rabies and Wildlife (ANSES), the NRL for rabies.

These two laboratories use the reference techniques recommended by the OIE (OIE, 2012, Rabies chapter) and the WHO (Meslin et al., 1996) and undertake phylogenetic identification of the virus strain in the event of positive diagnosis, providing information about the species and the type of virus (canine or from bats) and its geographical origin, which is of use for epidemiological investigations and for the implementation of management measures, especially in cases where rabies has been imported.

**Health control measures**
Rabies management is based on the management of animals that have been in contact with a rabid animal or one suspected to have rabies. The conditions and characteristics of contact are defined by the provisions of the CRPM, which specifically describes the identification of infected and potentially infected animals.

The classification of carnivorous animals as infected or potentially infected depends on the probability of contact between the carnivore and an animal known to be rabid, and this probability of contact is assessed by the DDecPP.

The management of infected animals is based on the Ministerial Order of 9 August 2011, which stipulates that infected animals not properly vaccinated at the time of infection must be euthanised.

The management of possibly infected animals is based on Article R. 223-34 of the CRPM. Appropriate measures determined by the Director of the DDecPP are taken with consideration for the species of lyssavirus infecting the animal recognised as rabid, and the vaccination status of the potentially infected animals.

**Regulatory References**

Ministerial Order of 21 April 1997 on the surveillance of biting and scratching animals as defined in Article R. 223-25 of the CRPM. Official Journal, 4p.

Ministerial Order of 1 March 2002 on approval of the National Centre for Veterinary and Food Studies, Nancy, for the diagnosis of animal rabies. Official Journal, 1108.

Ministerial Order of 1 March 2002 laying down the list of organisations responsible for examinations for the diagnosis of rabies in animals suspected of being at the origin of human contamination. Official Journal, 4389.


Ministerial Order of 9 August 2011 relating to specific measures to combat rabies applicable in the area of movement of a dog or cat recognised as rabid. Official Journal, 4p.

Case of indigenous rabies in a bat in the Loir-et-Cher département

The NRL diagnosed a second serotine bat as infected with a lyssavirus on 24 June 2014. This diagnosis using immunofluorescence was confirmed on 25 June using molecular biology techniques and on 26 June using cell infection. Typing of the virus, through partial nucleoprotein gene and polymerase gene sequencing, showed it was a genotype 5 (EBLV-1), subtype b lyssavirus, with 98.8% homology with a strain isolated in a bat from Spain and 98.2% homology with two EBLV-1b strains previously isolated in the Doubs and Meuse départements.

Case of indigenous rabies in a bat in the Haute-Vienne département

On 12 September 2014, a serotine bat received at the NRL was diagnosed as positive for rabies using immunofluorescence. The diagnosis was confirmed in the days that followed using cell infection and molecular biology techniques. Typing of the virus, through partial nucleoprotein gene and polymerase gene sequencing, showed it was a genotype 5 (EBLV-1), subtype b lyssavirus, with 98.7% homology with the EBLV-1b strain isolated three months prior from a serotine bat in the Loir-et-Cher département.

Discussion

Currently, most recorded cases of animal rabies in metropolitan France involve bats (57 cases since 2001). Nonetheless, cases of illegally imported infected pets (ten cases since 2001) are reported on a regular basis despite the implementation of strict regulations. Rabies thus remains a significant ongoing threat to animals in France and more broadly in Europe (Cliquet et al., 2014); since 2001, 22 alerts have been recorded in Europe, including twelve from Morocco. In this context, a one-day event entitled “Rabies, a highly topical disease” was jointly organised on 9 October 2014 by the Ministry of Agriculture, Food and Forestry and ANSES as part of World Rabies Day. This day of awareness-raising on the health risks of the disease and the risk of its introduction into France brought together over a hundred scientists, stakeholders involved in the surveillance and management of carnivorous animal movements, health professionals, veterinarians, and air and maritime transport operators.

Table 1. Regional distribution of animal species examined for diagnosis of rabies in France in 2014

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Figure 1. Geographical distribution of positive and negative diagnoses of rabies in metropolitan France for 2014

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The number of suspected cases and therefore analyses undertaken for the detection of rabies is high, and similar from one year to another for each animal category (Servat et al., 2014), which reflects a good level of vigilance among the parties involved. Moreover, the geographical distribution of these suspected cases remains fairly homogeneous, suggesting satisfactory coverage of the entire national territory.

The cases of rabies recorded each year in bats demonstrate the need to maintain a high level of information, prevention and vigilance on the part of the population and of mandated veterinarians regarding the risk related to these specific epidemiological cycles. Since 1989, 67 bats have been found to be infected with lyssaviruses in France. The serotine bat, the principal species infected with EBLV-1 in Europe, accounts for 64 of these 67 cases of rabies recorded in France. The recent discovery in Europe of the new BBLV (Bokeloh bat lyssavirus) (Picard-Meyer et al., 2013b; Dacheux et al., not published) and LLBV (Lleida bat lyssavirus) lyssaviruses, combined with the annual detection of infected bats, underlines the need to maintain and strengthen epidemiological surveillance in all regions for effective management and to raise the awareness of at-risk individuals. It is therefore well worth intensifying the collection of bats for diagnosis and in particular of target species such as serotine bats (carriers of EBLV-1), Natterer’s bats (supposed carriers of BBLV), bent-wing bats (carriers of LLBV) and Daubenton’s bats (carriers of EBLV-2).

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References