

Maisons-Alfort, 22 November 2006

OPINION

of the French Food Safety Agency (Afssa) on the risk of bluetongue developing over the coming months in the north of Europe and the conceivable measures for controlling this disease

Terms of the mandate

Further to the emergence of bluetongue outbreaks in mainland France, the Agence française de sécurité sanitaire des aliments (Afssa) [French Food Safety Agency] decided a self-mandate on 06 September 2006. The joint emergency expert group on "Bluetongue", created at the proposal of the specialist expert committee "Animal Health" at the session of 06 September 2006, was charged with conducting an expert review on bluetongue containing a description of the situation of this disease in northern Europe and its recent development in time and space, a risk analysis of its possible development over the coming months, conceivable control measures and lastly recommendations on desirable research that would enable a better control of the disease.

Opinion of the joint emergency expert group on "Bluetongue"

The joint emergency expert group on "Bluetongue", appointed in a decision dated 09 September 2006, met on 24 October, 07, 14 and 20 November 2006 and has issued the following opinion:

"Context"

- *The identification, in mid-August 2006, of the presence of bluetongue serotype 8 in the provinces of Dutch and Belgian Limbourg (Maastricht zone), then in North Rhine Westphalia in Germany and in the départements of Ardennes and northern France, has acknowledged the emergence of a real epizootic in a region that, in theory, seemed to be under less of a threat than others situated further south in the European Union. Moreover, this emergence has appeared particularly atypical in view of the serotype identified (entirely exotic in Europe and very poorly known worldwide), of the confirmed absence in the region of the main Bluetongue vector in Europe (*Culicoides imicola*), of clinical signs identified in a species that is traditionally of low sensitivity (cattle) even if its susceptibility to bluetongue is well-known, of very low mortality and morbidity rates in sheep, usually a highly sensitive species, and, lastly, low seroprevalence rates are frequently observed in herds where cases are identified. This emergence has prompted the implementation, in the affected zone, of control measures defined by European Union law, mainly in Directive 2000/75/EC and Decision 2005/393/EC and its various amendments, particularly of 18 August (OJEU of 1/08/2006), 1 September (OJEU of 02/09/2006) and 15 September 2006 (OJEU of 21/09/2006).*
- *Decision 2005/393/EC has more particularly defined a restricted zone F, encompassing the restricted zones (protection and surveillance zones) in the four Member States (Germany, Belgium, France, the Netherlands) where bluetongue*

serotype 8 has been identified, as well as Luxembourg. The distinction between protection and surveillance zone (in accordance with Directive 2000/75/EC) is only evident for France in the last amendment of Decision 2005/393/EC.

- The amendment to Decision 2005/393/EC of 15 September 2006 stipulates that inside restricted zone F (except for standstill zones – cf. the following paragraphs – including for movements between Member States, as provided for in Decision 2006/693/EC of 13/10/2006 amending Decision 2005/393/EC), movements of live ruminants and their sperm, ova and embryos are authorised, but that “in Member States which have established a surveillance zone (France, cf. above), movements in this zone may only take place after obtaining authorisation from the competent authority in the final destination”.
- Furthermore, with regard to animals (domestic ruminants) located in the 20 km radius standstill zones around the outbreaks (included in the protection zone), irrespective of the restricted zone (A, B, C, D, E or F), their movements are free when they are being transported directly to a slaughterhouse located in the corresponding restricted zone or a holding located in this same restricted zone when it meets certain conditions¹. For movements from a restricted zone to a bluetongue-free zone inside a Member State, there are also derogatory conditions laid down by Decision 2005/393/EC, which are valid irrespective of the restricted zone (A to F).
- It could therefore be considered that, on the basis of current law concerning restricted zone F, it is possible to receive ruminants from French protection and surveillance zones in restricted Belgian, Dutch or German zones. If, during the period of vector inactivity, the standstill zones, protection and surveillance zones were gathered into a single restricted zone in France (as for other Member States), ruminants from restricted zone F of other Member States could be received in the French part of restricted zone F. This possibility and any possible consequences must be assessed with regard to the epidemiological situation in France and in other Member States, both during the period of vector(s) inactivity and through the spring and autumn of 2007. If the epidemiological situation remained highly heterogeneous inside restricted zone F (as it did in autumn 2006), it would probably be necessary to adapt Community legislation for a better control of bluetongue in this zone and in particular to protect zones with a low prevalence of infection, as it is the case in north-eastern France at present.

¹ This must be situated (i) in a standstill zone or (ii) outside a standstill zone, and have received:

- either approval from the competent veterinary authorities in the places of origin and destination under the animal health conditions that they lay down concerning measures to control the spread of bluetongue and protection against any vector attacks,
- or a negative result to a bluetongue identification test on a sample obtained 48 hours prior to an animal's departure which must, in addition, be protected from vector attacks at least over the whole of the period since the sample and not leave the holding of destination, except to be slaughtered after direct transport.

Questions

The joint emergency expert group on “Bluetongue” is charged with:

1. Providing an exact description of this disease in northern Europe, and of its recent development in time and space;
2. Analysing the risk of its possible development over the coming months;
3. Presenting conceivable control measures, based on the objective adopted for controlling this disease and hypotheses of how the disease may develop;
4. Laying down recommendations on desirable research that would enable a better control of the disease.

Method

Following the meeting of 24 October 2006, the joint emergency expert group on “Bluetongue” produced a draft opinion which was discussed on 07, 14 and 20 November 2006 and validated using telematic facilities on 20 November 2006.

The review was conducted based on the following documents:

- Commission Decision 2005/393/EC and its subsequent amendments (the most recent dated 13 October 2006),
- the available data on the epidemiological situation in the Netherlands, Belgium, Germany (EFSA BTV working group, data updated on 20 November 2006, DEFRA on 17 November 2006, European Commission ADNS notification system on 17 November 2006),
- the available data on the epidemiological situation in France on 20 November 2006,
- OIE alerts on 20 November 2006,
- ProMED dispatches dated 20 November 2006,
- Purse B., Mellor P., Rogers D., Samuel A., Mertens P., Baylis M. (2005). Climate change and the recent emergence of bluetongue in Europe. *Nature reviews*, 3:171-181,
- Mullens B., Gerry A., Velten R. (2001). Failure of a Permethrin Regime to Protect Cattle Against Bluetongue Virus, *J. Med. Entomol.* 38(5): 760-762.

Examination of the issues

1. Description of the situation

Given the available data, (number and location of outbreaks identified since mid-August, species affected), particularly those issued by the working group of the European Food Safety Authority (EFSA) since 23 October 2006, the situation as at 20 November 2006 may be characterised by the following points:

- Over 1700 “cases” have been identified in the three most affected Member States (Belgium, Germany, the Netherlands). The two other Member States included in restricted zone F, Luxembourg and France, have identified no cases and six cases respectively (as at 20 November 2006).
- Two incidence peaks are detectable, one in the last week of August and first week of September (24/08/2006 to 07/09/2006), the other, more significant, running from the last week of September (21/09/2006 onwards) through to the present. The contribution of sheep seems to be considerably higher during the second peak, although it is still in the minority in one of the Member States (Germany) [cf. Annex 1A].
- In August, the majority of cases were identified within the Belgian, Dutch and German borders, in a 100 km radius around Maastricht, the epicentre of the epizootic. The

second incidence peak coincided with a very significant increase in the number of cases identified in north-western Belgium and in the area of North Rhine- Westphalia situated on the right bank of the Rhine, at the same time as a gradual reduction in the number of cases arising in the epicentre of the epizootic; the southern (Luxembourg, Belgian Ardennes) and northern zones (Netherlands above the Rhine) still have few or no cases.

- The cases identified to date therefore seem to have arisen, for the most part, within an irregular pentagon shape, the tips of which could be the towns of Dordrecht (Netherlands), Munster and Koblenz (Germany), Mons and Ostende (Belgium) [cf. Annex 1 B], with three zones presenting a much higher number of cases: the Maastricht region (epicentre of the epizootic), the area of North Rhine Westphalia on the right bank of the Rhine (in the districts of Düsseldorf, Cologne and Arnsberg) and the north-western region of Belgium. This pentagon, where a large proportion of the outbreaks identified have occurred, may be identified as an “epidemiologically active zone” (EAZ). The EAZ appears more limited in its extension than restricted zone F.
- Without additional data on outbreaks (serological prevalence in ruminants present at the moment of an outbreak identification, and one or two months later) and on the zones affected (serological prevalence in standstill zones and protection zones in most of the Member States affected), it is difficult to interpret the observations in terms of the cause of the spread of the infection (movement of infected animals and/or vector(s) activity). This difficulty, intensified by persisting uncertainty surrounding the identification of vector(s), is also faced when analysing the risk of possible development over the coming months and in 2007.

2. Analysis of the risk of bluetongue developing over the coming months and in 2007

2.1 Main determining factors

- Over the next few months, winter, corresponding to a period of vector inactivity, should lead to a reduction and then a halt in the emergence of new cases, more or less quickly depending on the weather evolution in the zone concerned, where it may be uniform (cf. temperature variations observed in early November between Dunkerque and Strasbourg).
- Through 2007, development will be linked, on the one hand, to a resumption in vector(s) activity after the winter season (April to June) and in summer-autumn (August to October) and, on the other hand, to the possible presence of a virus source whose origin and determinism are the source of many as yet unanswered questions.
- In northern Europe in 2007, it can be considered that there will be one or more vectors with the necessary competence to transmit bluetongue serotype 8 in zone F. Its (their) vector capacity will depend on environmental (and climatic) factors enabling sufficient development for it (them) to become (an) effective vector(s)."

Moreover, the access of this (these) vector(s) to a source of the bluetongue virus will be influenced to a large extent by the possibility of the virus persisting, whatever the mechanism. This is traditionally associated, on the one hand, with a persisting viremia observed in infected cattle over a period of sixty to one hundred days (maximum) and, on the other hand, with a series of hypotheses put forward to explain periods of virus persistence beyond winter (“overwintering”) that last longer than one hundred days; these are based on the preservation (and then the reactivation by vectors) of the virus in certain immune cells of the vertebrate host, on the survival of infected adult culicoides in certain biotopes, or on the vertical transmission of the virus in the vector culicoide. The possibility of access to a viral source will therefore be associated with the prevalence of the infection at the start of the vector inactivity period and throughout this period (the shorter the period, the higher the prevalence, the greater the probability of the virus overwintering, and vice versa).

- *The emergence or non-emergence of bluetongue in 2007 will therefore be associated, in view of the demonstrated presence of one or more competent vectors and the current location of bluetongue serotype 8, with the combination of probabilities of the virus overwintering and the development of a sufficient vector capacity. A number of facts, as yet unavailable, need to be gathered (see chapter 4 research) to be combined with information obtained from bluetongue surveillance procedures in zone F (culicoides population dynamics, level of infection of sensitive ruminant populations as the vector activity period comes to an end in the various restricted zones) and climatological monitoring (temperature in these zones) so that an assessment can be made of the risk of re-emergence, or not, of the epizootic in 2007. If cases were to be re-identified, these elements must enable the assessment, at least in the form of scenarios, of the possibilities of spatial extension and temporal development in, or outside, the epidemiologically active zone (EAZ) in 2006. Should development be observed and/or anticipated in the EAZ, control measures inside this EAZ and surveillance measures on its border may be adapted depending on the scenarios adopted.*

2.2 Hypotheses and possible scenarios

- *In theory, two major hypotheses concerning the development of the disease in zone F after the 2006-2007 winter can be distinguished, depending on the overwintering of the virus.*
 - *In the first case, the virus does not persist sufficiently to circulate; the virus and corresponding disease disappear.*
 - *In the second case, the virus, having survived, starts to circulate again by provoking or not provoking (cf. below) associated cases of the disease.*

The expert group considered that, if serotype 8 and the associated vector(s) present a comparable behaviour to that which was characterised in the previous episodes of bluetongue (for different vectors and serotypes), the probability of the second hypothesis arising is low (on a scale containing the levels "nil", "negligible", "low", "moderate" and "high"). Indeed, if the period during which winter temperatures prevent vector activity lasts longer than the maximum timespans (60 to 100 days) of viremia in cattle, then there is no more virus available for these vectors once they resume activity.

Should the opposite occur, the expert group believes that it does not have enough facts at its disposal to estimate the probability of either of these hypotheses arising.

- *In the context of the second hypothesis (virus circulation), three scenarios can be envisaged:*
 - Aborted re-emergence or disappearance of the disease but with virus circulation: in this case, the conditions which enabled the emergence and epizootic development of the disease in 2006 (presence of virus, vector competence) would not be gathered any more in 2007. Some cases may nonetheless be identified, particularly in the "epidemiologically active zone", without epizootic development.*
 - Re-emergence and persistence of the disease in similar conditions to those in 2006 with the regular presence of epizootic waves in the EAZ.*
 - Re-emergence and spreading of the disease to other regions, with development of epizootic waves that spread to surrounding areas, as it is the case for zone E (serotype 4) in the Iberian peninsula.*
- *To summarise, without knowing the possible survival mechanisms of the virus through the winter, or the duration or intensity of the winter months to come, it is*

difficult to make an exact estimation in November 2006 of the possibility of serotype 8 re-surfacing in northern Europe in 2007.

Nevertheless, this possibility is estimated as being low, if the 2006-2007 winter bears the usual characteristics (in terms of duration, intensity of cold temperatures). A new estimation in late March-early April may be more exact with account taken of the characteristics of the 2006-2007 winter.

If the disease does re-emerge in 2007, the degree of its development is hardly predictable in November 2006. It can only be considered that it would risk being more serious should it re-emerge early in the year.

A choice cannot therefore be made between these scenarios on the basis of current knowledge and available data concerning zone F. It should be conceivable in spring 2007, on the basis of (i) the outcome of research currently under-way, (ii) the availability of data required for characterising the situation as the current vector activity period comes to an end, and (iii) the results of the surveillance protocol implemented under the authority of the European Commission (opinion of 19 October 2006²).

3. Conceivable control measures based on the objective adopted for controlling bluetongue

Conceivable measures include, firstly, those to be implemented during the 2006/2007 winter and, secondly, those which may be applied as from spring 2007 after re-evaluation of the situation in late March-early April. They will mainly be considered for France, in view of the experts' better knowledge of the epidemiological situation in this country. Their principles are applicable in the other Member States affected, incorporating a necessary adaptation to their particular situation.

a) during the period of inactivity of culicoides 2006-2007

By taking account of the aforementioned estimation, it can be considered that:

- The possibility of the disease re-emerging somewhere in zone F in 2007 is low,
- If the disease were to re-emerge in 2007, it would probably do so in the zones that were significantly affected in 2006 (EAZ) rather than the zones located on its outskirts,
- The hypothesis of this re-emergence in the highly affected zone in 2006 cannot therefore be discounted,

It is possible to recommend:

- Continuing specific measures for standstill zones (20 km around outbreaks), noting the fact that the removal of ruminants from these zones depends on proof that these animals are not affected,
- Merging protection and surveillance zones and allowing ruminants to leave this extended zone for a bluetongue-free zone for slaughter and breeding during the period of vector inactivity, without any specific requirements. Given the information available on culicoides in France, this period is estimated to run (cf. opinion³ of 15 November 2006), depending on the zone for France:
 - o Mediterranean and Atlantic zones: from early January to mid March
 - o Continental zone: from early December to end of March
- The trapping in each of these zones over the coming months should provide information to confirm or otherwise the periods proposed for vector inactivity.

² Opinion 2006-SA-0279 of the French Food Safety Agency on a draft protocol for monitoring bluetongue in the European Union.

³ Opinion 2006-SA-0306 of the French Food Safety Agency on vector inactivity within the context of bluetongue.

- This period should also be turned to good account to check the earlier non-propagation of the infection outside of the standstill zones (serological or other surveys).

b) after resumption of culicoides activity and for the rest of 2007

It is recommended to continue checking for an absence of viremia in ruminants in the standstill zone due to be transported to a bluetongue-free zone or restricted zone.

- In the absence of any bluetongue serotype 8 outbreak identified in northern Europe

As long as re-emergence of the disease has not been demonstrated, it could be envisaged to allow free circulation in and from former protection and surveillance zones.

- In the event of bluetongue serotype 8 outbreak(s) emerging in northern Europe

In the event of bluetongue serotype 8 outbreak(s) emerging in northern Europe in 2007, the zoning measures provided for by European regulations shall be applied. Consideration may be given to the possible proposal of (an) alternative strategies (y) to current regulations. The study of the analytical epidemiology of bluetongue and corresponding control measures (cf. annex III) shows the diversity of obstacles encountered (arthropod vector, frequency of inapparent infection, etc.) and the difficulties in controlling this disease.

- In the absence of a vaccine, it is possible to partially limit the disease's spread, mainly by prohibiting ruminant movements in infected zones and limiting them in threatened zones.

The slaughter of animals who prove to be infected may also contribute to limiting the spread of a bluetongue epizootic, but may only be envisaged practically if the disease appears late (October?) and in a moderate manner in northern Europe. This would also reduce the probability of the disease re-emerging in the following year.

In fact, the method of control that would effectively modify the development of the disease and prevent a bluetongue epizootic from developing is a vaccine. In the hypothesis of a possible re-emergence of serotype 8 in northern Europe in 2007, the development of such a vaccine is of top priority.

It seems very unlikely that an effective and inoffensive vaccine against serotype 8 will be available to protect domestic ruminants in northern Europe in 2007. It is more likely for the year after.

- Should a vaccine be available in 2007:

If bluetongue were to re-emerge in 2007, the possibility and degree of use of vaccination of domestic ruminants in northern Europe would be considered for the 2007-2008 winter with a view to preventing another re-emergence in 2008.

Accordingly, at this stage, in addition to the recommendation to develop a vaccine against serotype 8, it is only possible to envisage, as a desirable modification of current European regulation on controlling bluetongue, the possibility of slaughtering infected animals if the disease re-emerges late in 2007, and in a limited manner.

4. Recommendations on desirable research enabling a better control of the disease

The joint emergency expert group on "Bluetongue" that met on 24 October, 07, 14 and 20 November 2006 at Afssa and using telematic facilities, has issued the following recommendations:

- *Research must be conducted first and foremost on four main areas:*
 - a) *Developing an inactivated virus vaccine against serotype 8, as well as tools (differential diagnostic tests) for implementing a DIVA strategy enabling the circulation of uninfected vaccinated ruminants between restricted and bluetongue-free zones;*
 - b) *Evaluating the effectiveness of protection methods that are currently recommended (pyrethrinoids) or conceivable (ivermectin), not only in holdings but also for transporting, based on the disinsectisation of susceptible animals and developing anti-vector control/prevention methods based on the physical layout of surroundings and/or reasoned use of “clean” larvicides (IGR, Bti, etc.);*
 - c) *Generating knowledge necessary for:*
 - i. *Understanding the dynamics of the infection within populations and individuals with regard both to receptive ruminants and vectors,*
 - ii. *Understanding the immunological and physiopathological characteristics of the bluetongue serotype 8 virus (infectivity and pathogenicity in susceptible ruminants),*
 - iii. *Identifying vectors, factors determining their vector capacity and competence (particularly checking the likelihood of vertical transmission of bluetongue serotype 8 in Culicoides (epidemiological importance in terms of the virus overwintering), and factors determining their geographical distribution (particularly the characterisation of larval biotopes of vector species),*
 - d) *Compiling a freely accessible database and, if required, producing data that can be used directly for risk assessment, therefore the choice between the hypothetical scenarios stated in 3., particularly:*
 - i. *Data revealing the rates of seroprevalence, morbidity and lethality in each of the outbreaks identified for each of the affected species,*
 - ii. *Data gathering together the available demographic information on the number and distribution of holdings of the three susceptible ruminant species in each of the affected regions, in order to appreciate the standard rates enabling an epidemiological description of the epizootic,*
 - iii. *Data revealing the geographical origin of the cases identified so as to distinguish the animals from the zones recognised as infected (“transplanted”, both inside a Member State, as with case No. 5 in France, and between Member states, as with the four other cases identified in France on animals from other affected Member States) [cf. annex 2] from those who were infected in their original holding (“native” as with the four cases identified in the French départements of Ardennes and Nord). This information should allow a better understanding of the respective role of viremic ruminant movements and of vector movements in the spatial and temporal spread of the epizootic.*
In this regard, it would probably be useful to clearly state (and define) the notions of infected (ruminants with no clinical signs), of cases (infected ruminants presenting clinical signs) and outbreaks (herds in which at least one case has been identified with virus circulation).
- *As a lesser priority for the immediate future, it would be desirable to have available for spring 2007, elements of analysis on:*
 - i. *The origin and ways of introduction of bluetongue serotype 8,*

- ii. *The possible role of wild ruminants in the “epidemiologically active zone” as reservoirs of the bluetongue virus or any other (related) orbivirus,*
- iii. *The role of possible co-infections with other pathogens in the clinical expression of the disease, particularly in cattle.*

Key words: *Bluetongue, cattle, sheep, culicoides”*

Opinion of the Agence française de sécurité sanitaire des aliments

These are the points of analysis which Afssa is in a position to provide in response to its self-mandate on the risk of bluetongue developing over the coming months and on the conceivable measures for controlling this disease in northern Europe where it emerged in mid August 2006.

The Director General of the Agence française de
sécurité sanitaire des aliments

Pascale BRIAND

Annex I , II and III

Annexe I A: Data on the temporal distribution of bluetongue serotype 8 in Europe

Table I. Number of declared cases of bluetongue serotype 8 per country and per animal species

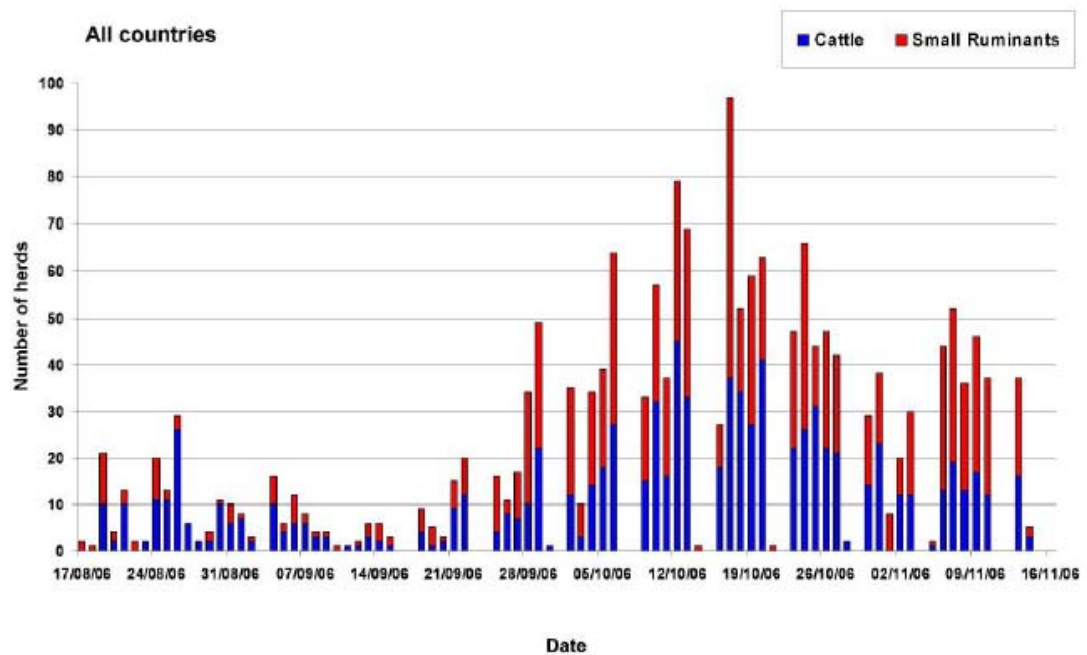
(Source: EFSA, Newsletter of 20 November 2006. Data updated on 15 November 2006, and France data updated on 20 November 2006).

Species	Germany	France	Netherlands	Belgium	Total
cattle	430	6	162	247	844
sheep	254	0	243	370	867
wild ruminants	5	0	0	0	5
mixed holdings*	8	0	25	2	35
total	697	6	430	619	1752

*herds with more than one of the aforementioned species.

Figure 1: Daily incidence of bluetongue serotype 8 cases based on the confirmation date and species (all countries)

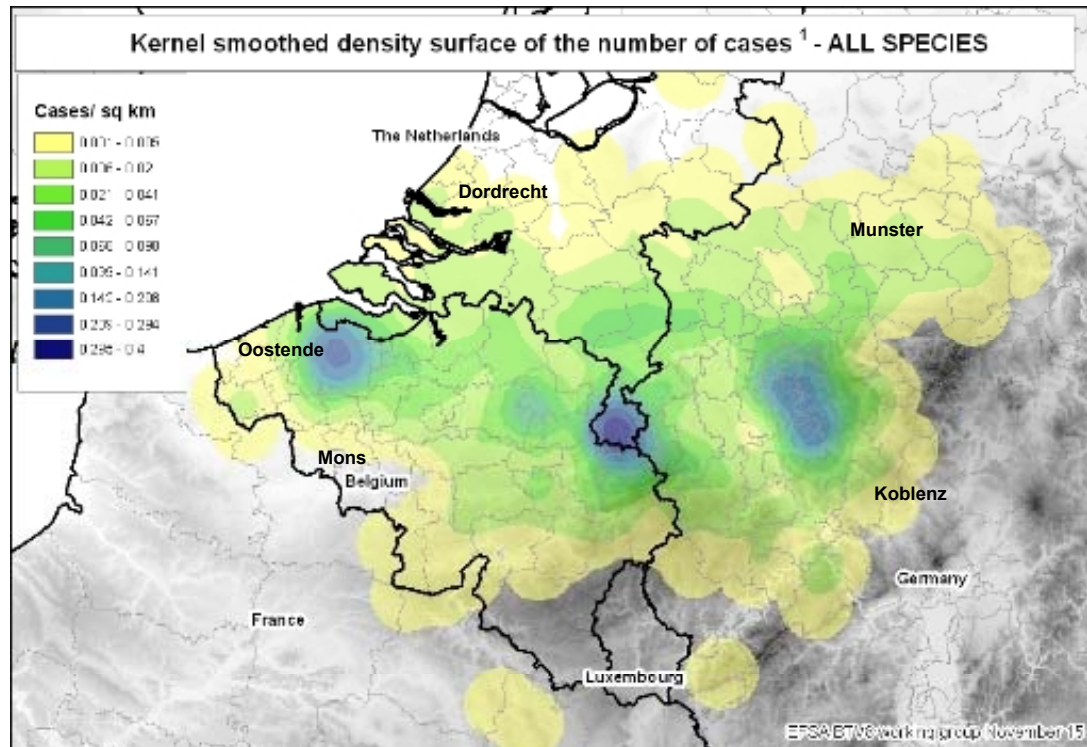
(Source: EFSA, Newsletter of 20 November 2006. Data updated on 15 November 2006).



Annex I B: Data on the geographical distribution of bluetongue serotype 8 in Europe

Figure 2: Geographical distribution and density of bluetongue serotype 8 cases in Europe, all species included

(Source: EFSA, Newsletter of 20 November 2006. Data updated on 15 November 2006)



Annex II: Data on bluetongue serotype 8 in France

Clinically expressed cases (chronologically)

Since 30 August (first French case), there have been around a hundred clinical suspicions reported (94 clinical suspicions, 2 confirmed, 93 cleared, 1 pending). The PCRs conducted from these samples allowed the detection of two cases (cases 1 and 4).

- **Case 1: a head of cattle (heifer) in Brognon in the Ardennes département (08)**
 - Suspicion following clinical signs (slight limp, slight oral inflammation on 28 August 2006), serology (ELISA+) and virological analysis (PCR+) confirmation on 30 August 2006.
 - Only one positive out of 30 animals tested (of a livestock of 96 cows).
- **Case 4: a head of cattle (bull-calf) in Taily in the Ardennes département (08)**
 - Suspicion following clinical signs (salivation, nasal discharge, watering of the eyes, oral congestion, hyperthermia on 1 September 2006), serology (ELISA+) and virological analysis (PCR+), confirmation on 06 September 2006.
 - A serology + and 3 PCR+ (4 in total)/out of 22 animals tested (of a livestock of 200 cows).

Prevalence surveys(chronologically)

A serological surveillance of cattle livestock (dairy cows, or bull-calves for fattening) was organised along the *départements* bordering Belgium and Germany from 25 to 31 August 2006.

60 cattle livestock were selected (targeting⁴ the proximity of holdings to the border and presence of wetlands: i.e. 8 holdings in the 02 *département*, 20 in 08, 8 in 54, 8 in 55, 8 in 57 and 13 in 59) and 30 animals per holding (actual average: 28.7), or a total sampling of 1,800 cows.

- 2 sera turned out to be positive by competitive ELISA (CIRAD-EMVT).
- The same two animals also proved positive by PCR (Afssa-LERPAZ).
- **Case 2: a cow in Beaurieux in the Nord département (59)**
 - Identified by serology (ELISA+ on 30 August 2006) then virological analysis (PCR+).
 - Only one positive cow out of 37 animals analysed (of a livestock of 143 cows).
- **Case 3: a cow in Hierges in the Ardennes département (08)**
 - Identified by serology (ELISA+ on 30 August 2006) then virological analysis (PCR+).
 - Only one positive cow out of 92 animals tested (of a livestock of 517 cows).
- **Case 6: a cow in Bondues in the Nord département (59)**
 - Identified by serology (ELISA + on 13 November 2006).
 - Only one positive cow out of 39 animals tested (on 14/11/2006).

⁴ Memorandum DGAI/SDSPA/N2006-8211 of 23 August 2006: Bluetongue – surveillance measures related to the epizootic in Germany, Belgium and the Netherlands.

In addition, two questionable serologies and one positive serologies, unconfirmed by a second serology and by PCR, have been recorded in 55.

Investigations related to moved animals

Investigations have been conducted on animals (mostly cattle) imported from infected regions in Belgium, Germany and the Netherlands since 1 June. As at 05 October, 3,674 animals (mainly cattle), had been analysed by serology.

Five sera turned out to be positive:

- 2 cows aged 3 to 7 years, imported from Belgium on 22 July to a holding in Seine-Maritime and found to be ELISA + on 30/08/2006 in *département* 76.
- a cow imported from Belgium in a holding in *département* 79, found to be ELISA+ on 13/09/2006.
- a calf imported from Germany in *département* 49, found to be ELISA+ on 02/10/2006.
- a calf imported from Germany in *département* 35, found to be ELISA+ on 10/10/2006.

The two animals from *département* 76 were PCR negative and were slaughtered on 5/09/2006 at the breeder's request. The cow from *département* 79 was a low positive after the ELISA test and PCR+: it was slaughtered on 19/09/2006 on DSV orders. The calf introduced into *département* 49 was ELISA positive and then found to be PCR+: It was euthanased on 06/10/2006. The calf introduced into *département* 35 was found to be PCR positive and euthanased on 12/10/2006.

In addition, animals who had been moved from standstill zones around the French outbreaks were traced.

- **Case 5: a cow (dairy cow) in Sivry-sur-Meuse in the Meuse *département* (55)**

The cow detected had left the holding on 05 September for another holding located in the Meuse.

- Identified by serology (ELISA+ on 6 October 2006) then virological analysis (PCR+).
- Only one positive cow out of 44 animals analysed (of a livestock of 44 cows). The 5 other cows tested in the livestock of destination are seronegative. No clinical signs of bluetongue were observed in the livestock of origin or destination.

Additional surveys related to a clinical or serological suspicion

Following the discovery of seropositive animals, serological surveys were conducted from 29/08/2006 to 08/09/2006 in holdings neighbouring the suspected holdings, in three *départements* (60, 08, 55)

- Around case 2 (DSV 59): 9 holdings and 36 animals per holding
- Around case 3 (DSV 08): 8 holdings and 28 animals per holding
- Around a suspicion, subsequently unconfirmed (DSV 55): 8 holdings and 28 animals per holding

1,417 cattle sera have been analysed (accredited ELISA tests), all of which have turned out to be negative.

Annex III: Analytical epidemiology of bluetongue and resulting control measures.

If bluetongue emerges in a region, the multiplication, spread and conservation of the virus are ensured, firstly, by domestic and wild ruminants and, secondly, by arthropods (various species of culicoides).

As regards the viral multiplication:

- *Cattle, sheep and goats (as well as wild ruminants) without any previous contact with the serotype or corresponding vaccine may act as virus amplifiers . Four situations, natural or which depend on human management, may influence this amplifying role.*
 - i) *Ruminants who were previously infected by the same serotype can play a greater amplifying role should new contamination by the same serotype occur. As a result, if in a given year a serotype managed to become very widespread in a region, the proportion of receptive subjects the following year is inversely proportional to the intensity of the incidence of the previous year (at the very least, the only animals to be receptive are the year's youngsters after colostral anti-bodies have been eliminated),*
 - ii) *Attempts could be made to protect ruminants from culicoide bites by bringing them inside or by using insecticides. The effectiveness of this action remains very limited, however, since:*
 - *It is not conceivable to bring inside every ruminant over an extensive geographical area every night;*
 - *Culicoides can be found inside buildings;*
 - *Protection by insecticide can prove ineffective: a study by Mullens et al. (2001) did not prove any significant difference between control cattle and cattle treated with permethrin every two weeks, as regards the frequency of serological conversion for bluetongue in summer.*
 - iii) *The question is raised as to whether it is conceivable to apply a slaughtering policy to control bluetongue, in the same way as is applied for non-vector diseases such as foot and mouth disease, swine fevers, highly pathogenic avian influenza, etc. In comparison with these diseases, two major obstacles are encountered in bluetongue:*
 - *The multiplication and spread of the virus by arthropods, which means that even a widespread slaughtering of domestic ruminants would not stamp out the source of the virus;*
 - *A frequently high proportion of ruminants that, despite infection, show no clinical signs of the disease; this means that slaughtering of clinically affected animals would only stamp out some of the viral sources in vertebrates.*

The slaughtering of animals whose infection (viremia) is identified would succeed in stamping out for good their role as a potential viral source for culicoides, and in possibly conserving the virus over the cold season.

However, such a slaughtering would, firstly, only stamp out a probably minor fraction of the viral sources and, secondly, be difficult to envisage in the event of a large-scale epizootic.

Nevertheless, particularly in the absence of available vaccines, it would be conceivable in the event of a late and limited emergence of the disease (in the period of culicoide activity).
 - iv) *When there is a vaccine available, it will be possible to immunise animals. This involves the making and availability of an effective and inoffensive vaccine . Moreover, the amplifying role of ruminants can only really be reduced depending on the proportion of receptive subjects vaccinated; this means envisaging the*

vaccination of three species: cattle, sheep and goats, particularly when cattle are frequently affected as is the case with serotype 8.

As a result, provided it is applied extensively to ruminants in the threatened region, vaccination is certainly the most effective means of limiting the virus amplifier role played by domestic ruminants. Of course, this leaves wild ruminants,, whose vaccination is impossible for the time being. It all depends on the respective weight of domestic and wild ruminants in the amplification of the virus in the region. It can be presumed that, for the time being in northern Europe, domestic ruminants have played a much greater amplifying role than wild ruminants.

- *Culicoides* form the other amplifying element of the bluetongue virus in an infected region. It is certainly interesting to find out the identity of the one or more species who play this role, particularly with regards to the geographical distribution of this (these) vector(s). However, whatever the species, it is very difficult, in practice, to prevent the bluetongue virus amplification within a region by the presently existing competent *culicoides* .

As regards the spread of the virus:

- Movements of cattle, sheep and goats in infected zones may be prohibited (cf. European regulations); and if they are necessary, screening of the infection in moved animals is conducted. The same type of action is not possible in wild ruminants.
- Movements of *culicoides* cannot be controlled. This has actually been checked through 2006, which saw bluetongue spread particularly over an East-West axis.

- As regards the conservation of the virus:

The more or less long conservation of the bluetongue virus may occur either in ruminants or in *culicoides*. In geographical zones where the climate favours *culicoide* activity and reproduction all year round, conservation of the virus is ensured by both over a continuous cycle.

In zones where there is a cold spell:

- The possibility of winter conservation of the virus (by viremia) is inversely proportional to the timespan of *culicoide* inactivity (cf. previous development).

Northern Europe is one of the northernmost regions to be affected by bluetongue. It is this aspect that suggests that the probability of the virus re-emerging next year is low.

- The other mechanisms of conservation in ruminants remain uncertain.
- Vertical transmission of the virus in *culicoides* also remains a poorly known mechanism as regards its frequency, and therefore possible role.