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The surveillance and control programme for bovine spongiform encephalopathy (BSE) in Norway

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# Surveillance and control programmes for terrestrial and aquatic animals in Norway

#### Title

The surveillance and control programme for bovine spongiform encephalopathy (BSE) in Norway

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# Introduction

All 20,143 samples, originating from 9,692 herds, were tested negative for BSE in 2008.

The BSE surveillance programme was initially based on passive surveillance (1998-2000), with active surveillance introduced in May 2000. In the period 1998-2000 the samples were investigated by histopathological examination. From 2001 onwards the samples were examined by an Enzyme-Linked ImmunoSorbent Assay (ELISA) method for detection of resistant prion protein (PrP<sup>Sc</sup>). In addition, clinically suspected animals were investigated by histopathological examination according to the protocol of the Office International des Epizooties (OIE) (1). The number of samples examined in each category in the period 1998-2008 is presented in Table 1. BSE has never been detected in any of the examined animals.

# Aim

The aim of the surveillance programme is to document that the Norwegian cattle population is free from BSE.

# Surveillance programme

#### Programme outline

For 2008 the surveillance programme was in accordance with the European Commission Regulations (EC) No 999/2001 Annex III with amendments. The programme included examination of the following categories:

- clinically suspected cattle irrespective of age
- all cattle older than 24 months of age, which have died or been culled, but not slaughtered for human consumption (fallen stock)
- all emergency slaughtered cattle older than 24 months
- all cattle older than 24 months, with abnormal findings at ante-mortem examination, rejected for human consumption, or which died at the abattoir or during transport (referred to as ante-mortem animals)
- all slaughtered cattle with unknown age or origin irrespective of age
- all slaughtered imported cattle from any country irrespective of age
- 10,000 randomly selected healthy routinely slaughtered cattle older than 30 months

#### Implementation

The farmers were requested to report all cases of clinically suspected cattle irrespective of age, fallen stock older than 24 months and when delivering an imported cattle to slaughter to the Norwegian Food Safety Authority. The brain or head from clinically suspected cattle or a spoon sample from the medulla oblongata from fallen stock, were submitted and analysed at the National Veterinary Institute, Oslo. Inspectors from the Norwegian Food Safety Authority collected the spoon samples of the medulla oblongata from the other categories at the abattoirs and sent them within 24 hours in a cool insulated container to the National Veterinary Institute in Oslo or Harstad.

# Laboratory methods

### Clinically suspected animals

The usual protocol followed for the clinically suspected animals is that the whole brain is divided midsagittally into equal halves. One half is formalin-fixed and processed according to a standard routine protocol, embedded in paraffin, sectioned at 5  $\mu$ m and stained with haematoxylin eosin (HE). Immunohistochemical staining for detection of PrP<sup>Sc</sup> is performed on selected sections using a monoclonal anti-PrP antibody (SAF 84, courtesy of J. Grassi, CEA, France). From the non-fixed half, tissue from the obex area <u>is</u> analysed by ELISA for detection of PrP<sup>Sc</sup> (TeSeE®, Bio-Rad) as described by the manufacturer. Table 1. Number of samples from cattle collected for BSE examination by the Norwegian surveillance programme according to categories from 1998-2008

| Reason for submission to the laboratory | 1998-<br>2000 | 2001   | 2002   | 2003   | 2004   | 2005   | 2006   | 2007   | 2008   |
|---|---------------|--------|--------|--------|--------|--------|--------|--------|--------|
| Clinically suspected animals            | 78            | 14     | 2      | 2      | 3      | 1      | 0      | 0      | 0      |
| Fallen stock                            |               | 1,352  | 1,482  | 1,872  | 2,145  | 2,318  | 2,364  | 2,213  | 2,391  |
| Emergency slaughtered                   |               | 7,073  | 7,246  | 7,322  | 9,217  | 8,462  | 8,177  | 7,304  | 8,358  |
| Ante-mortem animals                     |               | 2,612  | 3,562  | 4,102  | 1,355  | 102    | 36     | 48     | 16     |
| Imported slaughtered<br>animals         | 19*           | 88     | 39     | 39     | 24     | 10     | 4      | 9      | 5      |
| Healthy slaughtered<br>animals          |               | 2,400  | 9,907  | 10,726 | 10,443 | 10,486 | 10,455 | 10,000 | 9,373  |
| Total                                   | 97            | 13,539 | 22,238 | 24,063 | 23,187 | 21,379 | 21,036 | 19,574 | 20,143 |

\* All the samples were examined in 2000.

#### Risk population and routine slaughtered animals

Non-fixed brain tissue from the obex area was analysed by ELISA for detection of  $PrP^{Sc}$  (TeSeE®, Bio-Rad) as described by the manufacturer. In cases with positive or inconclusive test results, the remaining half obex was fixed in 10 % neutral buffered formalin, embedded in paraffin, sectioned at 5 µm, and stained with HE. Subsequently, the sections were analysed by immunohistochemical detection of  $PrP^{Sc}$  using the same protocol as for specimens from clinical suspects.

Brain samples were evaluated as unsuitable for examination when they were severely autolysed, the dorsal part of the obex area was cut obliquely, the obex was not present, or the medullar anatomy was not recognisable.

# **Results and discussion**

The National Veterinary Institute received samples from 20,143 cattle. Of these, 111 (0.6%) samples were unsuitable for examination (83 from fallen stock, 7 from emergency slaughter and 21 from healthy slaughtered animals).

For 319 samples (1.6%) the herd of origin was not reported. However, it is important to note that in case of a positive test result from such a herd, the identity could be traced via the carcass number. The remaining 19,824 samples originated from 8,309 dairy cattle herds and 1,383 beef cattle herds. The mean number of examined animals per herd was 2.0.

#### Clinically suspected animals (passive surveillance)

In 2008, no animals were investigated as clinical suspects. Improved methods for clinical examination to distinguish between real suspected BSE cases and cases with central nervous disease of other causes has probably resulted in few clinical suspected cases in later years. It is likely that animals with diseases related to the central nervous system have been examined either as fallen stock, emergency slaughtered animals or ante-mortem animals, and thus included in these categories.

### Surveillance of slaughtered animals and fallen stock (active surveillance)

Fallen stock older than 24 months comprises approximately 0.96% of the adult population (National Cattle Registry (Husdyrregisteret), per 31.12.2007), i.e. approximately 3,600 animals. The majority of samples from fallen stock were collected on farm. The difference between the examined number and the expected number of fallen stock is partly explained by the fact that many cattle herds are located in remote areas where sampling is considered too time consuming and cumbersome. In addition, a proportion of the cattle is grazing on mountain and forest pastures where sampling of dead animals may be too difficult.

Norwegian cows are slaughtered at a low age, the mean age is 54 months for dairy cows and 74 months for suckling cows (suckling cows constitute only 17,5 % of the cattle population older than 24 months) (National Production Recording Scheme 2007, Norwegian Beef Herd recording System 2007). The low age at culling implies that 43% of the samples are originated from cattle younger than 4 years. The age distributions of cattle sampled in different categories are shown in Table 2.

The geographical distribution of the cattle population and the animals of different categories tested are presented in table 3. There is a relatively good correlation between the collection of samples for fallen stock, emergency slaughter and healthy slaughtered animals from different regions and the distribution of the cattle population in the regions, but corresponding figures for the ante-mortem population show considerable variation between regions.

| Age groups<br>(months)  | Total<br>population<br>(%) | Fallen stock<br>(%) | Emergency<br>slaughter<br>(%) | Ante mortem<br>animals<br>(%) | Healthy slaugh-<br>tered animals<br>(%) | Total tested<br>(%) |
|-------------------------|----------------------------|---------------------|-------------------------------|-------------------------------|---|---------------------|
| < 24                    | 59.6                       | 1.2                 | 3.3                           | 0.0                           | 1.5                                     | 2.2                 |
| 24-29                   | 7.9                        | 11.7                | 14.9                          | 0.0                           | 7.8                                     | 10.8                |
| 30-35                   | 6.0                        | 10.3                | 8.4                           | 43.8                          | 11.2                                    | 9.7                 |
| 36-47                   | 9.9                        | 20.2                | 16.2                          | 18.8                          | 23.8                                    | 20.2                |
| 48-59                   | 6.8                        | 18.1                | 17.7                          | 12.5                          | 20.9                                    | 19.2                |
| 60-71                   | 4.1                        | 15.3                | 15.4                          | 6.3                           | 15.6                                    | 15.5                |
| 72-83                   | 2.4                        | 11.2                | 11.3                          | 6.3                           | 9.4                                     | 10.4                |
| 84-95                   | 1.4                        | 5.9                 | 6.4                           | 6.3                           | 5.1                                     | 5.8                 |
| 96-107                  | 0.8                        | 3.1                 | 3.3                           | 0.0                           | 2.6                                     | 2.9                 |
| 108-119                 | 0.4                        | 1.1                 | 1.5                           | 0.0                           | 1.0                                     | 1.2                 |
| 120-131                 | 0.2                        | 0.6                 | 0.7                           | 0.0                           | 0.5                                     | 0.6                 |
| 132-143                 | 0.1                        | 0.8                 | 0.3                           | 0.0                           | 0.2                                     | 0.3                 |
| 144-155                 | 0.1                        | 0.3                 | 0.2                           | 0.0                           | 0.2                                     | 0.2                 |
| ≥ 156                   | 0.1                        | 0.3                 | 0.3                           | 6.3                           | 0.1                                     | 0.2                 |
| Total number of animals |                            | 2,391               | 8,358                         | 16                            | 9,373                                   | 20,027*             |

Table 2. Age distribution of cattle from which samples were received in 2008. There were 848 samples (4.2%) from cattle with unknown age. The age of these cattle are assumed to be distributed like the age distribution of the cattle with known age within each target group.

\* Five imported animals not included

Table 3. Regional distribution of Norwegian cattle and the cattle tested for PrP<sup>Sc</sup> in 2008. There were 319 samples (1.6%) from cattle with unknown region. These samples are assumed to be distributed following the regional distribution of the cattle from known region within each target group.

| Region                         | Total<br>population<br>(%) | Fallen stock<br>(%) | Emergency<br>slaughter<br>(%) | Ante mortem<br>animals<br>(%) | Healthy<br>slaughtered<br>animals<br>(%) | Total<br>(%) |
|--------------------------------|----------------------------|---------------------|-------------------------------|-------------------------------|--|--------------|
| Oslo, Akershus, Østfold        | 4.5                        | 5.5                 | 6.3                           | 0.0                           | 3.9                                      | 5.1          |
| Hedmark, Oppland               | 18.2                       | 11.1                | 17.5                          | 0.0                           | 11.8                                     | 14.1         |
| Buskerud, Vestfold og Telemark | 6.3                        | 6.8                 | 5.0                           | 7.7                           | 9.4                                      | 7.3          |
| Rogaland og Agder              | 20.8                       | 24.0                | 19.5                          | 7.7                           | 26.4                                     | 23.2         |
| Hordaland, Sogn og fjordane.   | 10.3                       | 8.4                 | 6.8                           | 15.4                          | 9.5                                      | 8.3          |
| Trøndelag, Møre og Romsdal     | 29.7                       | 35.3                | 36.1                          | 69.2                          | 30.3                                     | 33.4         |
| Nordland                       | 7.3                        | 5.5                 | 6.3                           | 0.0                           | 6.4                                      | 6.2          |
| Troms og Finnmark              | 2.8                        | 3.5                 | 2.4                           | 0.0                           | 2.3                                      | 2.5          |
| Total number of animals        |                            | 2,308               | 8,351                         | 16                            | 9,352                                    | 20,027*      |

\* Five imported animals not included

# Conclusion

It is considered probable that the Norwegian cattle population never has been infected with BSE-agent due to few imports to Norway of cattle and products potentially infected with the BSE-agent, limited use of meat and bone meal in concentrates intended for ruminants, and the use of high temperature and pressure in the domestic production of meat and bone meal (2). This is supported by a recent quantitative risk assessment for BSE in Norway (3) and by the compiled results from the surveillance and control programme for BSE in the years 2001 to 2008 (4) with approximately 165,000 negative samples.

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