Surveillance and control programmes for terrestrial and aquatic animals in Norway

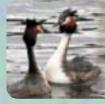


















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Surveillance and control programmes for terrestrial and aquatic animals in Norway

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# Contents

| Preface  | 7   |
|--|-----|
| Introduction   |     |
| Main results from the surveillance and control programmes in 2006  | 10  |
| The livestock and aquaculture populations in Norway  | 13  |
| The surveillance and control programmes for <i>Salmonella</i> in live animals, eggs and meat in Norway   | 21  |
| Residues of prohibited substances in live animals and of veterinary drugs, prohibited substances and environmental contaminants in animal products in Norway |     |
| The surveillance and control programmes for paratuberculosis in Norway   |     |
| The surveillance and control programme for bovine spongiform encephalopathy (BSE) in Norway  | 41  |
| The surveillance and control programme for infectious bovine rhinotracheitis (IBR) and infectious pustular vulvovaginitis (IPV) in Norway                    | 49  |
| The surveillance and control programme for enzootic bovine leukosis (EBL) in Norway  | 55  |
| The surveillance and control programme for <i>Brucella abortus</i> in cattle in Norway   | 61  |
| The surveillance and control programme for bovine virus diarrhoea (BVD) in Norway  |     |
| The surveillance and control programme for bovine tuberculosis in Norway   |     |
| Survey of specific serogroups of <i>E. coli</i> in sheep in Norway   |     |
| The surveillance and control programme for maedi in Norway   |     |
| The surveillance and control programme for Brucella melitensis infection in sheep flocks in Norway   |     |
| The surveillance and control programme for scrapie in Norway   |     |
| The surveillance and control programme for specific virus infections in swine herds in Norway  |     |
| The surveillance and control programme for chronic wasting disease (CWD) in wild and captive cervids in Norway   | 109 |
| The surveillance and control programme for avian influenza (AI) in wild birds in Norway  | 115 |
| The surveillance and control programme for avian influenza (AI) in poultry in Norway   | 119 |
| The surveillance and control programme for infectious laryngotracheitis (ILT) and avian rhinotracheitis (ART) in poultry flocks in Norway                    | 123 |
| The surveillance and control programme for <i>Campylobacter</i> in broiler flocks in Norway  |     |
| The surveillance and control programme for viral haemorrhagic septicaemia (VHS) and infectious haematopoietic necrosis (IHN) in Norway                       | 133 |
| The surveillance and control programme for <i>Gyrodactylus salaris</i> in Atlantic salmon and rainbow trout in Norway  | 141 |
| The surveillance and control programme for bacterial kidney disease (BKD) in Norway  | 147 |
| The surveillance and control programme for bonamiosis and marteiliosis in European flat oysters ( <i>Ostrea edulis</i> L.) in Norway                         | 153 |

# Preface

We are pleased to present the results of the surveillance programmes for terrestrial and aquatic diseases in Norway in 2006. These results, together with the animal disease report to the World Organization for Animal Health (OIE), confirm that Norway continues to ensure a very good situation concerning diseases in terrestrial and aquatic animals.

We continue to be free of all relevant serious contagious diseases in aquaculture, with the exception of ISA.

A very real threat that Highly Pathogenic Avian Influenza might be found for the first time in Norway, required much attention and resources in 2006. Despite increased awareness and active surveillance we are very happy to report that no cases were found in either wild or domesticated birds.

The last cattle herds with bovine virus diarrhoea were declared free of the disease and therefore, after 14 years of intensive collaboration between the industry and the authorities to combat this disease, we have declared the disease eradicated in Norway.

A serious outbreak of *E. coli* O103 in humans led to tracings back to sheep meat used in sausage production. This has unleashed an aftermath of discussions concerning the possible control of EHEC/VTEC in live animals. A survey in sheep was initiated in 2006 and will be concluded in 2007.

We hope the reader will find relevant and interesting information in this report. For more information in English concerning the Norwegian Food Safety Authority we refer to our web page at www.mattilsynet.no.

Oslo, June 2007,

Joakim Lystad Director general, Norwegian Food Safety Authority

# Introduction

This report contains information on the official surveillance programmes for diseases in aquatic and terrestrial animals in Norway in 2006. These programmes are run by the Norwegian Food Safety Authority and are planned and coordinated by the National Veterinary Institute.

Since 1994 Norway has had a free trade agreement (European Economic Area agreement or EEA) with the EU that has among other fields encompassed most of our veterinary legislation. This agreement includes Norway's commitment to follow harmonised legislation concerning surveillance and control of animal diseases. The surveillance programmes in Norway are therefore categorized according to their legal basis as programmes implementing EEA directives, programmes related to additional guarantees within the EEA region and programmes based solely on national requirements.

# Surveillance programmes for documentation and control

# Programmes implementing EEA-directives and regulations

Bovine brucellosis was eradicated in Norway over 50 years ago and the last case of tuberculosis was recorded in 1986. Based on this information a freedom of disease status was approved by the EFTA Surveillance Authority (ESA) in 1994 on historical data. In order to maintain the free-status a surveillance programme was established in 2000. The status of enzootic bovine leucosis (EBL) has been documented and the few infected animals have been eliminated. On this basis, Norway has applied for free-status for enzootic bovine leucosis. This status was approved by ESA early in 2007.

Surveillance for bovine spongiform encephalopathy (BSE) in cattle and scrapie in sheep and goats is performed according to the requirements of the EU regulations. Norway has never had a case of BSE and in later years all scrapie cases have been of the NOR98 variant. Early in 2007 Norway received additional guarantees for classical scrapie based on our ongoing national surveillance and control programme.

As part of the EEA-agreement in 1994, Norway achieved the status of freedom from *Brucella melitensis* in small ruminants based on historical data. In order to maintain this position, a surveillance and control programme was established in 2004.

In poultry, programmes for Newcastle disease, *Mycoplasma* and *Salmonella* were established according to EU-directives. In the autumn of 2005 the threat of global avian influenza increased substantially. A surveillance programme on avian influenza in wild birds was initiated as part of the preparedness for preventing introduction into commercial poultry flocks. In 2006 a surveillance programme in commercial flocks was also initiated.

This report also contains information on the programme for control of residues in live animals and animal products of ruminants, pigs and poultry.

The programmes for aquatic animals are of paramount importance for the intensive and export oriented aquaculture industry in Norway. The purpose is twofold, combining prevention of spread of diseases through trade from infected premises or regions, and the documentation of a free-status to benefit the export of aquaculture products. The surveillance for viral haemorrhagic septicaemia (VHS) and infectious haematopoietic necrosis (IHN) was initially based on the recognition of free-status for these diseases on historical data. In 2004 the entire coastline of Norway was recognized as an approved zone with regard to *Bonamia ostreae* and *Marteilia refringens*. The decision is based on the results of the surveillance and control programmes for bonamiosis and marteiliosis which were initiated in the autumn of 1995.

# Programmes related to additional guarantees within the EEA region

Some diseases are not regulated by common EEA rules. However, countries may apply for additional guarantees based on their documented status. In 1994, additional guarantees for infectious bovine rhinotracheitis (IBR) in cattle and Aujeszky's disease (AD) in pigs were granted to Norway.

The favourable *Salmonella* situation in Norway was recognized by the ESA in 1994. The additional guarantees were based on the national surveillance and control programmes for cattle, pigs and poultry.

# Other national surveillance and control programmes

Several diseases of great national significance have no legal basis in the EU legislation. Norwegian authorities and industries have for years used great efforts and resources to control and eradicate diseases such as bovine virus diarrhoea (BVD) in cattle, and maedi in small ruminants.

# Responsibilities for the programmes

The surveillance and control programmes are part of the legislation for terrestrial and aquatic animal health and food in Norway. This legislation is decided by the Ministry of Agriculture and Food, the Ministry of Fisheries and Coastal Affairs and the Ministry of Health and Care Services jointly as regulations under the Norwegian Food Law. The Norwegian Food Safety Authority is responsible for implementation of all measures related to this legislation. The National Veterinary Institute ensures the scientific quality of the programmes with regard to epidemiological design, testing and analysing with approved methods and by presenting and interpreting the results according to accepted standards. Sampling is performed by or under the supervision of official inspectors in the Norwegian Food Safety Authority.

Ongoing programmes for terrestrial and aquatic animals in 2006 (the year of initiation in parentheses)

| Animal category | Programmes according to EU-directives and regulations  | Programmes approved by ESA                 | Other national surveillance and<br>control programmes                |
|-----------------|--|--|--|
| Cattle          | BSE (1998)<br>Residual substances (1999)<br>EBL (1994)<br>Tuberculosis (2000)<br>Brucellosis (2000)              | IBR/IPV (1992)<br><i>Salmonella</i> (1995) | Paratuberculosis (1996)<br>BVD (1992)                                |
| Swine           | Residual substances (1999)   | AD (1994)<br><i>Salmonella</i> (1995)      | TGE (1995)<br>PRRS (1995)<br>Swine influenza (1997)                  |
| Small ruminants | Scrapie (1997)<br>Brucellosis (2004)   |  | Maedi (1997)<br><i>E. coli</i> (2006)                                |
| Poultry         | Residual substances (1999)<br>Newcastle disease<br><i>Mycoplasma</i><br><i>Salmonella</i> (1995-breeding flocks) | Salmonella (1995-96)                       | ILT (1997)<br>ART (1997)<br><i>Campylobacter</i> (2001)<br>AI (2005) |
| Farmed deer     | Tuberculosis (2000)  |  | CWD (2005)   |
| Llama           |  |  | Paratuberculosis (2000)  |
| Fish            | VHS/IHN (1994)   |  | <i>Gyrodactylus salaris</i> (2000)<br>BKD (2006)                     |
| Shellfish       | Bonamia/Marteilia (1995)   |  |  |

BSE=bovine spongiform encephalopathy, EBL=enzootic bovine leukosis, IBR=infectious bovine rhinotracheitis, IPV=infectious pustular vulvovaginitis, BVD=bovine virus diarrhoea, AD=Aujeszky's disease, TGE=transmissible gastroenteritis, PRRS=porcine reproductive and respiratory syndrome, ILT=infectious laryngotracheitis, ART=avian rhinotracheitis, Al=avian influensa, HPAI=highly pathogenic avian influenza, CWD=chronic wasting disease, VHS=viral haemorrhagic septicaemia, IHN=infectious haematopoietic necrosis, BKD= Bacterial kidney disease.

The economic funding for the programmes is agreed between the Norwegian Food Safety Authority and the National Veterinary Institute as part of the annual steering agreement between these institutions.

# Impact of the programmes

The programmes serve several purposes for Norwegian authorities and for the agriculture and aquaculture industries. The scientific documentation shows that Norway complies with legal commitments in relation to international agreements. The programmes contribute also to decreasing the risk associated with trade of animals and animal products and in the case of zoonotic diseases the programmes constitute a scientific documentation with great significance for food safety. Finally, the documentation provided is important for industries exporting aquatic and terrestrial animals and products.

addung

Roar Gudding Director general, National Veternary Institute

Keren Bar-Yaacov Chief Veterinary Officer, Norwegian Food Safety Authority

# Main results from the surveillance and control programmes

The surveillance and control programme for bovine virus diarrhoea (BVD) started in December 1992. Its aim to eradicate the disease from the cattle population has been achieved as the restrictions of the last herd has been lifted and no new herd was restricted due to BVD in 2006.

From 2000 to 2006, more than 120,000 bovines have been investigated for BSE. All samples have been negative. Classical scrapie was detected in one sheep flock while scrapie Nor98 was diagnosed in eight sheep and one goat coming from nine different flocks. Chronic Wasting Disease (CWD) was not detected in any of the cervids tested in 2006.

Maedi was discovered in a number of sheep flocks in 2002 and a few seropositives have since been detected annually by the surveillance programme. No positives were however, detected in 2006.

The surveillance for Aujeszky's disease, swine influenza, transmissible gastroenteritis, and porcine respiratory and reproductive syndrome in pigs was negative in 2006, giving additional documentation of freedom from these specific virus infections in the Norwegian swine population. This status is currently unique in an international context.

Influenza A was found in 85 out of 1,274 wild birds tested, but none of them were positive for high pathogenic strains. All the 3,199 farmed birds (306 flocks) were tested negative for high pathogenic influenza strains. All samples tested negative to antibodies against avian rhinotracheitis (ART) in turkey and to infectious laryngotracheitis (ILT) in broiler and layers.

The annual prevalence for poultry flocks positive for *Campylobacter* sp., decreased from 7.7 % in 2001 to 3.3 % in 2004. But for the last two years, there seems to be an increasing trend 3.6 % in 2005 and 4.9 % in 2006.

The Norwegian *Salmonella* programmes document that the Norwegian cattle, swine, sheep, and poultry populations are only sporadically infected with *Salmonella* sp.

Due to an outbreak of *Escherichia coli* in humans in 2006, a surveillance programme was established to investigate possible geographical variation and risk factors for the occurrence of different human pathogenic strains of *Escherichia coli* in sheep. Samples taken in 2006 will be analysed together with samples taken 2007.

A total of 69 samples (1.9%) from animals and primary animal products that were classified as non-compliant, containing substances from groups: A2 Thyrostats, A3 Steroids, B2b Coccidiostats and B3c Heavy metals.

*Mycobacterium avium* subsp *paratuberculosis* is endemic in the six counties that contain half of the goat population in Norway. Two goat herds tested positive as well as two sheep herds as a result of a follow up surveillance on positive goat herds. No cattle herds were found positive during 2006.

The surveillance and control programme for bacterial kidney disease (BKD) in salmonids 2005 and 2006 show that BKD is present in feral salmonids, but was not discovered in farmed brood stock fish. Norway has a disease free status for viral haemorrhagic septicaemia (VHS), infectious haematopoietic necrosis (IHN), bonamiosis and marteiliosis. The results from 2006 support the free status for these infections in Norwegian populations of aquatic animals. *Gyrodactylus salaris* was detected in two rivers in 2006, one of which had been rotenone treated in 2003. In the other river, the parasite was observed for the first time. Hitherto, *G. salaris* has been detected in 46 Norwegian rivers.

| Species         | Infection                     | Start | Extent of program   |
|-----------------|-------------------------------|-------|---|
| Cattle          | IBR/IPV                       | 1992  | 10 % of dairy cattle<br>10 % of beef cattle   |
|                 | Brucella abortus              | 2000  | In cases of abortic   |
|                 | BVD                           | 1992  | All diary cattle her<br>20% of beef cattle  |
|                 | EBL                           | 1994  | 10 % of dairy cattle<br>10 % of beef cattle   |
|                 | Bovine tuberculosis           | 2000  | Inspection of carca of suspected lesio  |
|                 | BSE                           | 1998  | Investigation of cli  |
|                 |                               | 2000  | Testing of importe  |
|                 |                               | 2001  | Testing of fallen stoc<br>Testing of animals<br>Testing of random   |
| Swine           | AD                            | 1994  | All breeding herds,<br>a selection of integ   |
|                 | TGE                           | 1994  | a selection of integ  |
|                 | PRRS                          | 1995  |   |
|                 | Swine influenza               | 1997  |   |
| Poultry         | Avian influenza in wild birds | 2005  | Cloacal and trached during the hunting  |
|                 | ILT                           | 1997  | All chicken (broile   |
|                 | ART                           | 1997  | All turkey breeder<br>turkey broiler floc   |
|                 | Campylobacter                 | 2001  | All broiler flocks  |
| Small ruminants | Scrapie                       | 1997  | Testing of clinicall  |
|                 |                               | 2002  | Testing of fallen st  |
|                 |                               | 1997  | Random sampling   |
|                 |                               | 1997  | Testing of primary  |
|                 | Maedi                         | 1997  | All breeding flocks 2003-2005   |
|                 | Brucella melitensis           | 2004  | All breeding flocks 2004-2005   |
|                 | E. coli                       | 2006  | 100 randomly sele   |
| Several species | Salmonellosis                 | 1995  | Cattle: 3,000 lymp<br>Swine: 3,000 lymp<br>from all breeding<br>Poultry: faecal san<br>or >250 layers/bre |
|                 | Paratuberculosis              | 1996  | Testing of clinicall<br>Testing of all llam<br>randomly selected  |
| Fish            | VHS/IHN                       | 1994  | Sampling 50 % of a farms tested in th   |
|                 | BKD                           | 2005  | Sampling 50 % of a farms only produc course of a two-ye   |
|                 | Gyrodactylus salaris          | 2000  | Sampling ~50 % of<br>trout farms. Samp<br>parr/smolts from a  |
| Oyster          | Bonamiosis                    | 1995  | Sampling of select annually   |
|                 | Marteiliosis                  | 1995  | Sampling of select annually   |

| mes in 2006   | Number of samples examined in 2006  | Positive samples in 2006  | Previous positive results   |  |
|---|---|---|---|--|
| herds<br>herds  | 1,673 bulk milk samples<br>4,624 blood samples from 479 herds   | None<br>None  | 1992: 1 positive herd   |  |
| ns  | 11 foetuses from 11 herds<br>36 blood samples from 20 cows (14 herds)   | None None   |   |  |
| ds<br>herds   | 14,620 bulk milk samples<br>997 pooled blood samples<br>113 individual blood samples from 28 herds  | 2004: restrictions lifted in a 2005: restrictions lifted in a     | ed in 1097 herds and imposed on 413 herds<br>4 herds and imposed on 4 herds<br>4 herds and imposed on 2 herds<br>1 herd and not imposed in any herd   |  |
| herds<br>herds  | 1,673 bulk milk samples<br>4,624 blood samples from 479 herds   | None<br>None  | 1995-1996: 7 positive herds<br>2002: 1 positive herd  |  |
| isses at slaughter, submission<br>is for testing  | Organs from 3 individuals   | None  | 1984: 1 positive herd<br>1986: 1 positive herd  |  |
| nically suspect animals   | 1 sample  | None  | None  |  |
| d animals and their progeny   | 10 samples  | None  | None  |  |
| and emergency slaughtered animals   | 10,541 samples  | None  | None  |  |
| selected at <i>ante mortem</i> control  | 36 samples  | None  | None  |  |
| y selected slaughtered animals  | 10,455 samples  | None  | None  |  |
| all nucleus herds of the sow pools and ated and fattening herds are tested                                  | 4,555 samples from 457 herds  | None  | None  |  |
|   |   |   |   |  |
| ×   | 4,542 samples from 457 herds  | None  | None  |  |
| «   | 4,559 samples from 457 herds  | None  | None  |  |
| *   | 4,552 samples from 457 herds  | None  | 1998: 1 positive herd (H3N2)  |  |
| al swabs from healthy birds shot season.  | 1,274 birds from 5 counties   | 85 positive for AI; no HPAI,<br>10 LPAI H5 and no H/.             | 2005: 80 positive birds; no HPAI, 2 LPAI<br>H5N2 and no H/.   |  |
| and layer) breeder flocks   | 3,811 samples from 72 holdings (127 flocks)   | None  | 2005: 1 seropositive flock  |  |
| flocks and randomly selected<br>s   | 869 samples from 28 holdings (29 flocks)  | 1 seropositive flock  | 2003: 2 positive flocks (1 holding)<br>2004: 2 positive flocks (1 holding)<br>2005: 1 seropositive flock  |  |
|   | Samples from 3,908 flocks   | 190 (4.9 %) positive flocks                                       | 2001: 7.7 % positive flocks<br>2002: 6.3 % positive flocks<br>2003: 4.9 % positive flocks<br>2004: 3.3 % positive flocks<br>2005: 3.6 % positive flocks   |  |
| / suspect animals   | 28 samples  | None  | 1997-2004: 24 positive individuals<br>2005: 1 positive individual   |  |
| ock   | 4,941 samples   | 6 positive individuals  | 2002-2004: 15 positive individuals<br>2005: 1 positive individual   |  |
| of slaughtered animals  | 15,613 samples  | 4 positive individuals  | 2001-2004: 16 positive individuals<br>2005: 2 positive individuals  |  |
| and secondary flocks  | 473 samples   | 1 flock (6 individuals)   | 2003-2004: 2 positive flocks<br>2005: None  |  |
| of sheep once during the period   | 27,846 samples from 911 flocks  | None  | 1998-2004: 4 positive flocks<br>2005: 2 positive flocks   |  |
| of sheep once during the period   | 27,812 samples from 911 flocks  | None  | None  |  |
| cted sheep flocks   | -   | -   | -   |  |
| h node samples<br>n node samples, faecal samples<br>ierds<br>nples from all flocks of >50 broilers<br>eders | 2,317 lymph node samples<br>3,484 lymph node samples and 2,438<br>faecal samples from 143 herds<br>6,964 faecal samples from 1,310 holdings | None<br>None<br>None  | 1995-2002: Only a few positive samples<br>each year, 2003: 5 positive (2 cattle, 2<br>swine, 1 broiler), 2004: 3 positive samples<br>(2 cattle, 1 swine), 2005: 3 positive<br>samples (2 cattle, 1 swine) |  |
| suspect animals<br>s older than 48 months and<br>cattle, goat and sheep herds                               | Organ and faecal samples from 524 cattle,<br>1,132 goats, 214 sheep and 81 llamas   | 2 goat herds<br>2 sheep herds                                     | 1997: 4 cattle herds (imported animals)<br>1998-2004: 5 cattle herds, 17 goat herds<br>and 2 sheep flocks, 2005: 14 goat herds  |  |
| I salmonid and turbot farms (all course of a two-year period)   | 12,390 samples from 392 sites   | None  | None  |  |
| I salmonid farms except sea-water<br>ng food fish (all farms tested in the<br>ar period)                    | 2,755 samples from 99 sites   | None  | None  |  |
| all fresh water salmon and rainbow<br>ing of Atlantic salmon fingerlings/<br>pproximately 106 rivers        | 1,862 fish from 57 salmonid farms<br>3,082 fish from 94 rivers  | No positive salmonid farms<br>2 positive rivers<br>(1 reinfected) | 1975-2005 39 positive salmonid farms, last<br>time 2002 (3 hatcheries)<br>1975-2005: 45 positive rivers   |  |
| ed farms and wild populations twice   | 270 oysters from 6 sampling points  | None  | None  |  |
| ed farms and wild populations twice   | 270 oysters from 6 sampling points  | None  | None  |  |
|   |   |   |   |  |



# The livestock and aquaculture populations in Norway





Gry Grøneng Petter Hopp

# The livestock population

Norway covers an area of 323,895 square km and has a population of about 4.7 million people of which about 0.8 million live in or in the vicinity of the capital Oslo. The livestock production is targeted for the national market. Table 1 gives an overview of the livestock population and the number of animals slaughtered in 2006.

Until 1994 there was a general ban on the import of live animals and animal products to Norway. Live animals could only be imported if derogation was given by the Veterinary Authorities. Consequently, there have been very few imports of live animals to Norway. Table 2 shows the number of live animals and animal products imported to Norway in 2005 and 2006.

As a consequence of the European Economic Area (EEA) agreement which was implemented in 1994, the trade of certain animals and animal products within the area was regulated through EU harmonised directives, and the general ban on import of these animals and products to Norway was lifted. There was a general increase in the interest to import live animals during that decade. The authorities encouraged beef production, and the need for suckling cows was met by import of live animals.

# The cattle population

Approximately 14,800 dairy herds were registered in Norway in 2006 of which approximately 1,300 also kept suckling cows. The average number of dairy cows per herd was 17.6. The number of specialized beef herds with at least one suckling cow was about 5,400 with a mean number of 11.0 suckling cows per herd. Overall, the number of Norwegian dairy herds has decreased over the last 15 years (Figure 1).

From 1980 to 1986, approximately 560 cattle were imported. There were no imports from 1987 to 1990. The European Economic Agreement in 1994 allowed more imports of live cattle. Nevertheless, as seen in Figure 2, the number of imports has been limited and most im-ported animals came from Sweden and Denmark. Close to 100 % of the imports have been beef cattle. In 2006, 8 live cattle were imported to Norway (Table 2).

# The swine population

The population consists of approximately 62,200 breeding swine aged more than six months. Approximately 160 approved elite and multiplier breeding herds house 5 % of the live sows in the population, while more than 95 % of the sows purchased on the national market are raised in these herds. About 50 % of the swine production is located in the counties of Hedmark, Oppland, Rogaland and Nord-Trøndelag. In 2006, 1 live swine was imported to Norway (Figure 3). In 2006, about 1.5 million swine were slaughtered.

|   | No. of  |
|---|---|
| Table 1. The livestock population in Norv | and the number of slaughtered animals in 2006 |

|                                      |                       | No. of                 |                         |
|--------------------------------------|-----------------------|------------------------|-------------------------|
| Animal category                      | herds*                | animals*               | slaughtered animals*    |
| Cattle                               | 20,500 <sup>1</sup>   | 918,200 <sup>1</sup>   | 332,100 <sup>2</sup>    |
| Dairy cows only**                    | 13,500 <sup>1</sup>   | 233,700 <sup>1</sup>   | -                       |
| Suckling cow only**                  | 4,100 <sup>1</sup>    | 50,800 <sup>1</sup>    | -                       |
| Combined production (cow)**          | 1,300 <sup>1</sup>    | 33,300 <sup>1</sup>    | -                       |
| Goat                                 | 1,300 <sup>1</sup>    | 72,100 <sup>1</sup>    | 21,100 <sup>2</sup>     |
| Dairy goat**                         | 510 <sup>1</sup>      | 42,500 <sup>1</sup>    | -                       |
| Sheep                                | 16,000 <sup>1</sup>   | 2,334,200 <sup>1</sup> | 1,211,300 <sup>2</sup>  |
| Breeding sheep > 1 year**            | 15,800 <sup>1</sup>   | 894,100 <sup>1</sup>   | -                       |
| Swine                                | 3,000 <sup>1</sup>    | 813,800 <sup>1</sup>   | 1,527,500 <sup>2</sup>  |
| Breeding animal > 6 months**         | 1,800 <sup>1</sup>    | 62,200 <sup>1</sup>    | -                       |
| Fattening pig for slaughter          | 2,700 <sup>1</sup>    | 432,000 <sup>1</sup>   | -                       |
| Poultry                              |                       |                        |                         |
| Egg laying hen (> 20 weeks of age)   | 2,000 <sup>1</sup>    | 3,262,700 <sup>1</sup> | 1,764,300 <sup>2</sup>  |
| Flocks > 250 birds**                 | 740 <sup>1</sup>      | 3,225,800              | -                       |
| Broiler                              | 520 <sup>2</sup>      | -                      | 49,167,500 <sup>2</sup> |
| Turkey, duck and goose for slaughter | 100 <sup>1</sup>      | 250,700 <sup>1</sup>   | 1,025,200 <sup>2</sup>  |
| Flocks > 25 birds**                  | 51 <sup>1</sup>       | 250,400                | -                       |
| Ostrich                              | <b>9</b> <sup>1</sup> | 81 <sup>1</sup>        | -                       |

<sup>1</sup> Register of Production Subsidies as of 31 July, 2006, <sup>2</sup> Register of Slaughtered Animals.

\* Numbers >100 rounded to the nearest ten, numbers > 1000 rounded to the nearest hundred, \*\* Included in above total.

| Table 2. Import of live animals and animal products to Norway in 2005 and 2006 |  |
|--|--|
|--|--|

|                 |                            | 20                  | 2005                          |                     | 06                            |
|-----------------|----------------------------|---------------------|-------------------------------|---------------------|-------------------------------|
| Species         | Imported product           | No. of consignments | No. of animals<br>or products | No. of consignments | No. of animals<br>or products |
| Cattle          | Live animals               | -                   | 0 <sup>1</sup>                | 1                   | 8 <sup>1</sup>                |
|                 | Semen (doses)              | С                   | 39,265 <sup>1</sup>           | С                   | 35,4041                       |
|                 | Embryos                    | 7                   | 63 <sup>1</sup>               | 3                   | 50 <sup>1</sup>               |
| Swine           | Live animals               | 1                   | 49 <sup>1</sup>               | 1                   | 1 <sup>1</sup>                |
|                 | Semen (doses)              | С                   | 394 <sup>1</sup>              | 12                  | 170 <sup>1</sup>              |
| Sheep           | Live animals               | 2                   | 39 <sup>1</sup>               | 4                   | 71 <sup>1</sup>               |
|                 | Embryos                    | 2                   | 339 <sup>1</sup>              | -                   | 0 <sup>1</sup>                |
|                 | Semen (doses)              | 3                   | 500 <sup>1</sup>              | 1                   | 24 <sup>1</sup>               |
| Goat            | Live animals               | 2                   | 53 <sup>1</sup>               | 1                   | 20 <sup>1</sup>               |
|                 | Semen (doses)              | 1                   | 100 <sup>1</sup>              | -                   | 0 <sup>1</sup>                |
| Reindeer        | Live animals for slaughter | 1                   | 2 <sup>2</sup>                | 2                   | 150 <sup>2</sup>              |
| Fur animal      | Live animals               | 38                  | 4,631 <sup>2</sup>            | 42                  | 16,361 <sup>2</sup>           |
| Poultry         | Day-old chicks             | 18                  | 133,155 <sup>1</sup>          | 10*                 | 97,499 <sup>*1</sup>          |
|                 | Fertilised eggs            | 51                  | 2,313,130 <sup>1</sup>        | 126*                | 5,587,650*1                   |
| Turkey          | Day-old chicks             | 4                   | 8,757 <sup>1</sup>            | 4*                  | 8,050*1                       |
| Duck and goose  | Live birds                 | 3                   | 1,505 <sup>1</sup>            | 2*                  | 1,345*1                       |
| Halibut         | Live fish                  | -                   | 0 <sup>2</sup>                | NA                  | NA                            |
| Turbot          | Live fish                  | 7                   | 181,820 <sup>2</sup>          | 8                   | 187,000 <sup>2</sup>          |
| Atlantic salmon | Live fish                  | -                   | 0 <sup>2</sup>                | 1                   | 286,000 <sup>2</sup>          |

<sup>1</sup> Data from Norwegian Livestockindustry's Biosecurity Unit (KOORIMP), <sup>2</sup> Data from the Norwegian Food Safety Authority. \*Only commercial imports, hobby imports are not registered. C=Continuous import, not possible to differentiate consignments. NA= Not available.

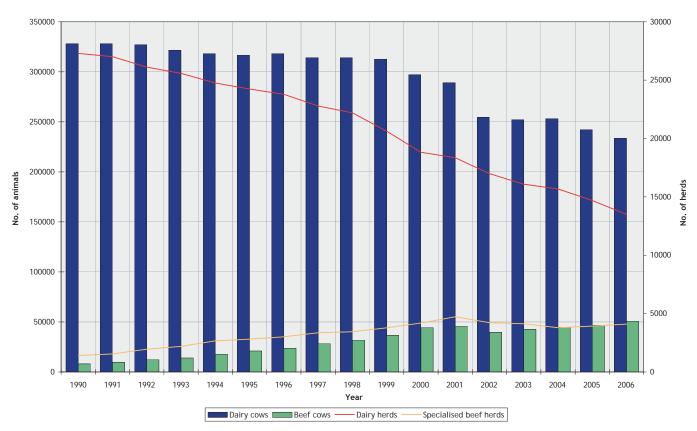


Figure 1. The number of dairy and beef cows in holdings with specialized dairy and beef production during the time period 1990-2006 (Statistics Norway and Register of production subsidies (RPS) for 2006).

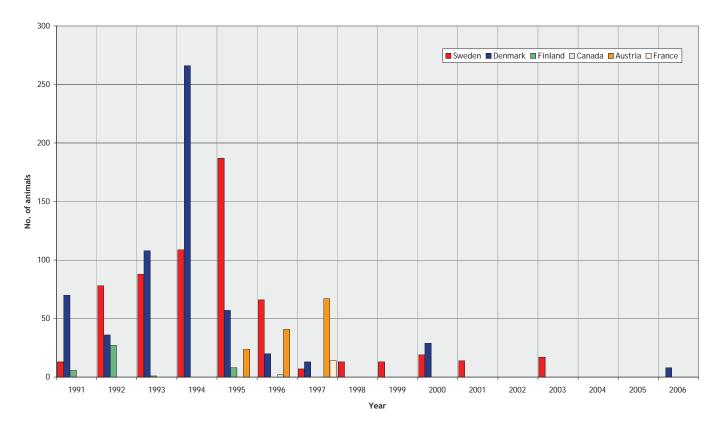


Figure 2. Imports of live cattle to Norway during the time period 1991-2006.

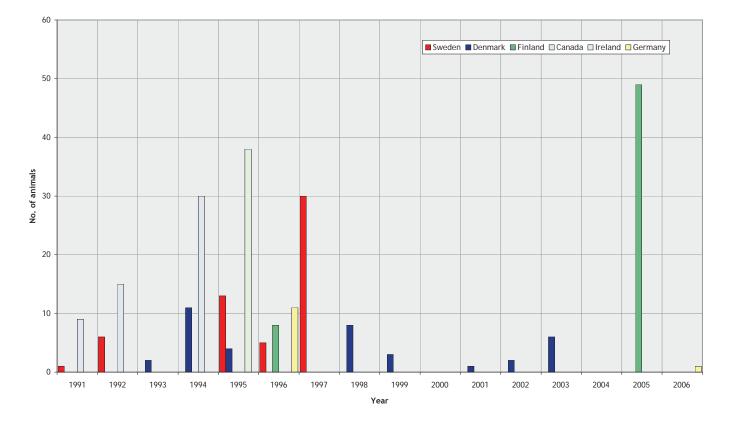


Figure 3. Import of live swine to Norway during the time period 1991-2006.

### The sheep population

The Norwegian sheep population consists of approxi-mately 894,100 sheep above one year of age. The sheep flocks are widely distributed over the country, with the biggest population found in the south-west. The sheep population consists of combined meat and wool pro-ducing breeds, with the breeds dala, spæl, steigar and rygja predominating. Each year about 1.2 million sheep are slaughtered and approved for human consumption. In 2006, 71 live animals were imported.

# The goat population

The Norwegian goat population is comprised of approximately 42,500 dairy goats and is principally composed of one Norwegian breed. The goat flocks are located in mountainous regions in the southern part of the country, in the fjord districts of the western part, and in the counties of Nordland and Troms in northern Norway. The main product is milk used for cheese production. About 21,100 goats are slaughtered and approved for human consumption each year. 20 live goats were imported in 2006.

# The poultry population

The Norwegian poultry production is strictly regulated and the population has a hierarchical structure. Egg and broiler meat production are the most important branches, but the production and consumption of turkey is increasing slightly. Figure 4A shows the location and structure of the Norwegian layer population comprising two hatcheries, 18 pullet rearing farms and about 800 commercial layer farms. The layer population consists of two white layer strains (Lohmann white and Shaver white).

The commercial broiler production takes place in three hatcheries with one strain (Ross), about 70 breeding farms with parent holdings and about 520 commercial broiler flocks. None of these farms are located in the northern part of Norway, as shown in Figure 4B.

The layer and broiler industry import day-old grand parent flocks mainly from Sweden.

# The population of farmed fish and shellfish

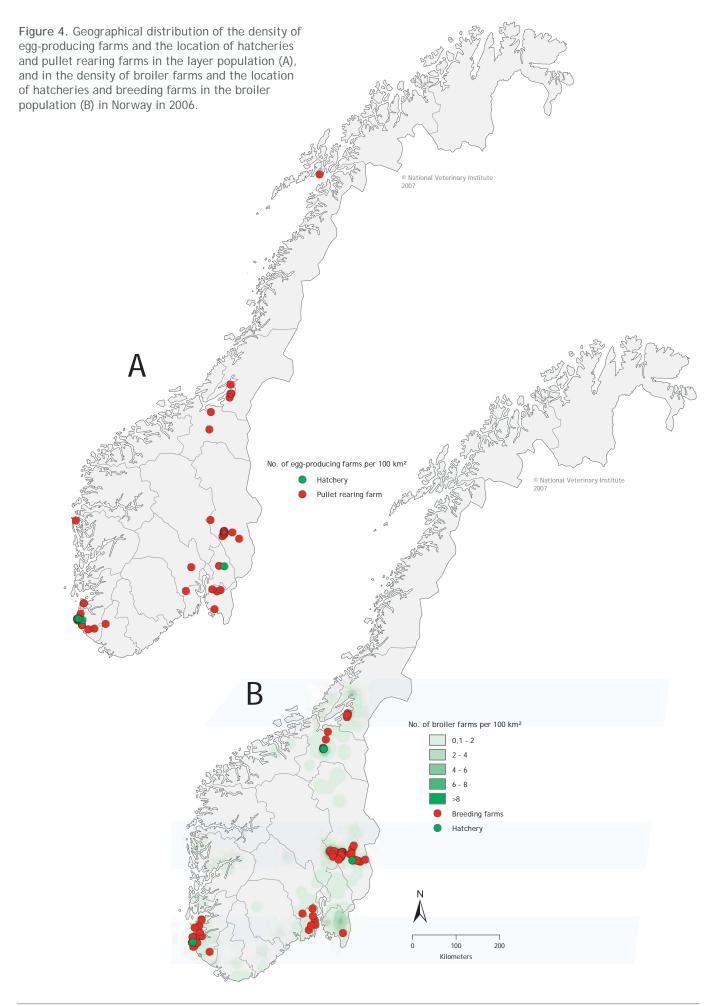
Atlantic salmon is the most important species in the fish and shellfish farming industry. The counties of Hordaland and Nordland are the major counties for seawater farms producing Atlantic salmon. The production volume of Atlantic salmon increased with 3 % from 2004 to 2005. A small reduction was observed in the volume of rainbow trout production in 2003, 2004, 2005 and 2006 (Table 3).

The import of live fish in 2006 consisted of eight consignments of turbot (Table 2).

| Year | Atlantic<br>salmon<br>(tons) | Rainbow<br>trout<br>(tons) | Cod<br>(tons) | Arctic char<br>(tons) | Halibut<br>(tons) | Blue mussels<br>(tons) | Scallops <sup>2</sup><br>(tons) | Oysters<br>(tons) |
|------|------------------------------|----------------------------|---------------|-----------------------|-------------------|------------------------|---------------------------------|-------------------|
| 1992 | 141,000                      | -                          | -             | -                     | -                 | -                      | -                               | -                 |
| 1993 | 170,000                      | -                          | -             | -                     | -                 | -                      | -                               | -                 |
| 1994 | 204,686                      | 14,571                     | 569           | 262                   | 63                | 542                    | -                               | -                 |
| 1995 | 261,522                      | 14,704                     | 284           | 273                   | 134               | 388                    | -                               | -                 |
| 1996 | 297,557                      | 22,966                     | 191           | 221                   | 138               | 184                    | -                               | -                 |
| 1997 | 332,581                      | 33,295                     | 304           | 350                   | 113               | 502                    | -                               | -                 |
| 1998 | 361,879                      | 48,431                     | 203           | 200                   | 291               | 309                    | -                               | -                 |
| 1999 | 425,154                      | 48,692                     | 157           | 498                   | 451               | 662                    | 67.1                            | 40.6              |
| 2000 | 440,861                      | 48,778                     | 169           | 129                   | 548               | 851                    | 37.6                            | 7.6               |
| 2001 | 436,103                      | 71,764                     | 864           | 318                   | 377               | 920                    | 22.3                            | 2.5               |
| 2002 | 462,495                      | 83,560                     | 1,258         | 319                   | 424               | 2,557                  | 5.0                             | 1.7               |
| 2003 | 509,544                      | 68,931                     | 2,185         | 272                   | 426               | 1,829                  | 1.2                             | 1.6               |
| 2004 | 563,815                      | 63,401                     | 3,165         | 350                   | 649               | 3,747                  | 45.5                            | 3.3               |
| 2005 | 586,512                      | 58,875                     | 7,409         | 352                   | 1,197             | 4,885                  | 3.0                             | 2.0               |
| 2006 | 626,382                      | 62,707                     | 11,087        | 881                   | 1,185             | 3,705                  | 4.0                             | 1.0               |

Table 3. Production volume of the most important species in Norwegian aquaculture during the time period 1992-2006<sup>1</sup>.

<sup>1</sup> Data from The Directorate of Fisheries, <sup>2</sup> From the wild population.



The surveillance and control programmes for *Salmonella* in live animals, eggs and meat in Norway



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# Introduction

The Salmonella surveillance programme in 2006, documents that the Norwegian population of cattle, swine, sheep, and poultry is just sporadically infected. The estimated prevalence is below 0.2 %.

The occurrence of *Salmonella* in Norwegian production animals and animal products is very low compared to most other countries, and has been so during the last decades.

The recorded incidence of human salmonellosis has increased in Norway during the last three decades. However, the overall situation seems to have been stable the last five years. In the majority of cases of salmonellosis (approximately 80 %), the patients had acquired the disease abroad (1). Meat produced in Norway is not considered a source of indigenous human salmonellosis.

As it is very important to maintain this favourable situation in Norway, in connection with the Norwegian negotiations for membership in the European Union, the Norwegian *Salmonella* control programme was established (2). In 1995, the programme was launched simultaneously with comparable programmes in Sweden and Finland (3, 4).

The Norwegian *Salmonella* control programmes for live animals, eggs and meat, consists of two main parts; surveillance and control. The surveillance covers live animals (pigs, cattle and poultry), fresh meat (pigs, cattle and sheep) and poultry meat (2). When *Salmonella* is isolated, action is taken to eliminate the infection, prevent transmission, and prevent contamination of food products. The programme is approved by the EU Commission (EFTA Surveillance Authority Decision No. 68/95/COL of 19.06.1995), allowing Norway to require additional guarantees regarding *Salmonella* when importing live animals, feed and food products of animal origin from the European Union.

The surveillance programmes for live animals, fresh meat and poultry meat are based on bacteriological examination for *Salmonella*. Isolation of *Salmonella*, irrespective of serovar, is notified to the Norwegian Food Safety Authority which maintains overall responsibility for the Salmonella surveillance and control programmes. The National Veterinary Institute coordinates the surveillance programmes, examines the faecal samples and publishes the results in monthly and annual reports. Private laboratories perform the examination of samples collected at slaughterhouses and cold stores.

# Aims

The aims of the programmes are to ensure that Norwegian food-producing animals and food products of animal origin are virtually free from *Salmonella*, to provide reliable documentation of the prevalence of *Salmonella* in the livestock populations and their products, and to prevent an increased occurrence of *Salmonella* in Norway.

# Materials and methods

The *Salmonella* surveillance and control programme for live animals includes examination of faecal samples from swine and poultry, and lymph node samples from cattle and swine (at least five ileo-caecal lymph nodes from each animal). The *Salmonella* surveillance and control programme for fresh meat and poultry meat includes examination of swab samples from cattle, swine and sheep carcasses, neck skin samples from poultry and samples of crushed meat from slaughterhouses and cold stores.

The number of samples examined in the different parts of the programmes is sufficient to detect at least one *Salmonella*-positive sample if the prevalence in the population is at least 0.1 %, with a confidence level of 95 %.

# Sampling scheme for live animals

#### Swine

In Norway there are approximately 160 elite and multiplier breeding herds for swine. More than 95 % of marketed breeding animals are purchased from these herds. All elite and multiplier breeding herds are surveyed annually at herd level. Pooled faecal samples are collected from all pens (up to a maximum of 20) con-taining piglets aged two to six months. If there are less than three pens of piglets at this age, additional indi-vidual faecal samples are collected from all sows (up to a maximum of 59) (5).

The pig population is surveyed by sampling a representative proportion of all pigs slaughtered in Norway. A total of 3,000 lymph node samples from swine (both sows and slaughter pigs) are collected at the slaughterhouses. The sample size for each slaughter-house ranges from 20 to 240 and is based upon the number of onsite slaughtered animals in relation to the national total. The sampling is distributed evenly throughout the year (6).

# Cattle

The surveillance is based on sampling a representative proportion of all cattle slaughtered in Norway. A total of 3,000 lymph node samples from cattle are collected at the slaughterhouses. The sample size for each slaughter-house ranges from 20 to 100 and is based upon the number of onsite slaughtered animals in relation to the national total. The sampling is distributed evenly throughout the year (6).

# Poultry

All breeding flocks and commercial production flocks, except layer flocks with less than 250 birds, are included in the surveillance programme. All breeder flocks are certified and the sampling scheme is in accordance with the old Zoonosis Directive (Council Directive 92/117/ EEC) (Table 1). All broiler flocks and flocks of turkeys, ducks and

Table 1. Sampling of poultry breeders (simplified) in the Salmonella surveillance and control programme in 2006

| Category of poul | ltry   | Time of sampling                               | Sample material                      |
|------------------|--|--|--------------------------------------|
| Grandparents     | Day old  | At arrival                                     | Organs or meconium                   |
|                  | Rearing  | 1-2 weeks, 4 weeks, 9-11 weeks and 13-14 weeks | Faecal samples                       |
|                  | Egg production*<br>- from the house<br>- in the hatchery | Monthly<br>Every 2nd week of production        | Faecal samples<br>Organs or meconium |
| Parents          | Day old  | Day 1  | Organs or meconium                   |
|                  | Rearing  | 4 weeks and 2 weeks before start of production | Faecal samples                       |
|                  | Egg production*<br>- in the hatchery                     | Every 2nd week of production                   | Organs or meconium                   |

\* Hatcheries with a production <1,000 eggs per year are sampled at the poultry house every two weeks.

geese other than breeders are sampled one to three weeks before slaughter (faecal samples), whilst layer flocks are sampled twice during the rearing period and once or twice during the egg laying period (2).

## Clinical cases - all animal species

Animals with clinical symptoms consistent with salmonellosis should be sampled for bacteriological diagnosis. In addition, all sanitary slaughtered animals are tested for the presence of *Salmonella*. Any *Salmonella* isolated from animals, irrespectively of serovar, is notifiable in Norway.

# Sampling scheme for fresh meat and poultry meat

#### Swab samples from carcasses

The testing of slaughtered pigs, cattle and sheep for *Salmonella* is done by swabbing carcass surfaces. For each animal species, a total of 3,000 swab samples should be collected at slaughter. For each slaughterhouse, the sample size ranges from 20 to 100 and from 20 to 240 for cattle and swine, respectively. The number of swab samples of cattle and swine from each slaughterhouse equals the number of lymph node samples. The number of swab samples from sheep ranges from 20 to 160 per slaughterhouse. The sampling is distributed evenly throughout the year. The sampling is done near the end of the slaughter line before the carcasses are refrigerated. Approximately 1,400 cm<sup>2</sup> of each carcass is swabbed (somewhat less for sheep) (6).

#### Neck skin samples

Neck skins from broilers and layers, turkeys, ducks and geese are tested for *Salmonella*. At each slaughterhouse, a minimum of five neck skins samples are collected per day and at least one sample must be taken from each flock slaughtered on a single day.

# Food products

The surveillance and control programme for cutting plants and cold stores are based upon samples of crushed meat taken from the equipment or from trimmings. Each sample consists of 25 grams. Each production line is sampled separately. The sampling is done randomly during operation. The number of samples taken in cutting plants and cold stores is given by the production capacity of the plant, and ranges from one sample per week to two per year (6).

Pre-packed fresh meat intended for cold stores does not have to be examined if they come from cutting plants that are included in the programme. However, freshly packed or repacked meat should be sampled.

## Laboratory methods

All lymph nodes from one animal are divided into two equal parts. One half is used for testing and the other half is stored at 4 °C until the results of the bacteriological examination is ready. The lymph node from at most five animals are pooled and homogenized before bacteriological examination. Swab samples are pooled in groups of five before testing. Each neck-skin sample is divided into two equal parts. One half is pooled with four to eleven other samples. The other half of neck skin samples are stored separately at 4 °C until the results of the bacteriological examination are ready. If the pooled sample is confirmed positive for *Salmonella*, the individual samples are examined separately.

Microbiological examination of the samples is carried out according to the Nordic Committee on Food Analysis Method No. 71, but slightly amended to make the method applicable to the various kinds of materials. This is a qualitative bacteriological method based on selective enrichment and cultivation. All positive samples are confirmed and serotyped by a reference laboratory.

# Results

## Live animals

#### Swine

A total of 2,438 faecal samples from 143 elite and multiplier breeding herds (including AI centres and testing stations) were examined in 2006 (Table 2). *Salmonella* was not detected in any of the samples. A total of 3,484 lymph node samples from slaughtered pigs were examined. Approximately 31 % of the samples were taken from sows and 69 % from slaughter pigs. None of the samples was positive for *Salmonella* (Table 3) giving an estimated *Salmonella* prevalence of 0 % (95 % confidence interval: 0 % - 0.1 %) at the individual carcass level.

#### Cattle

In 2006, a total of 2,317 lymph node samples from cattle were examined (Table 3). None of the samples were positive for *Salmonella* (Table 3) giving an estimated *Salmonella* prevalence of 0 % (95 % confidence interval: 0 % - 0.1 %) at the individual carcass level.

#### Poultry

A total of 6,964 faecal samples from 1,310 different holdings were examined (Table 4). None of the samples were positive for *Salmonella*.

#### Fresh meat and fresh poultry meat

# Swab samples from cattle, sheep and swine carcasses

A total of 7,695 swab samples from 36 slaughterhouses were examined in 2006 (Table 5). *Salmonella enterica* subsp. *diarizonae* was detected in one sample taken from sheep.

#### Neck skin samples from poultry

A total of 5,420 neck skin samples from poultry were examined in 2006. The samples came from all the six poultry slaughterhouses in Norway. Nearly 87 % of the samples came from broilers, 6 % from layers and 6 % from other species (turkeys and ducks). *Salmonella* Anatum was detected in one sample from broiler.

# Cutting plants and cold-stores for fresh meat and poultry meat

A total of 1,405 samples of crushed meat from 75 different plants were examined. *Salmonella* Dublin was detected on one of the samples.

# Discussion

The results from the *Salmonella* surveillance programme in 2006 document that the Norwegian cattle, swine, sheep and poultry populations are only sporadically infected with *Salmonella*. This is in accordance with previous findings (7-11). The estimated prevalence is below 0.2 % in the examined populations for any of the years the surveillance programme for live animals has run. The number of positive samples has never exceeded ten in total per year. *S.* Typhimurium has been isolated most frequently from swine, cattle and poultry, while *S. enterica* subsp. *diarizonae* is found most frequently from sheep. *S.* Enteritidis has never been found in the surveillance programme.

| Herd category                     | No. of herds<br>sampled (total*) | No. of samples<br>examined | No. of positive<br>samples | Salmonella serovar |
|-----------------------------------|----------------------------------|----------------------------|----------------------------|--------------------|
| Elite breeding herds              | 56 (56)                          | 978                        | 0                          |                    |
| Multiplier herds                  | 84 (108)                         | 1,340                      | 0                          |                    |
| A.I. centres and testing stations | 3 (1)                            | 120                        | 0                          |                    |

 Table 2. Sampling in elite and multiplier breeding swine herds in the Salmonella surveillance and control programme in 2006

 No. of berds
 No. of samples

\* Total number of herds is estimated as elite and multiplier breeding herds per 1 January 2006 excluding herds which ended breeding activity during 2006 before being tested.

 Table 3. Number of individual lymph node samples from cattle and swine examined in the Salmonella surveillance and control programme in 2006

| Species        | No. of slaughterhouses<br>sampled (total*) | No. of samples<br>examined | No. of positive<br>samples | Salmonella serovar |
|----------------|--|----------------------------|----------------------------|--------------------|
| Cattle         | 31 (38)                                    | 2,317                      | 0                          |                    |
| Slaughter pigs | 26 (30)                                    | 2,411                      | 0                          |                    |
| Sows           | 15 (30)                                    | 1,073                      | 0                          |                    |

\* Slaughterhouses where the number of slaughtered animals of a species is less than 100 according to the Slaughter Statistics for 2006 are not included in the sampling scheme.

Table 4. Samples from poultry in the Salmonella surveillance and control programme in 2006

| Poultry breeding flocks      | No. of samples tested | No. of holdings<br>tested | No. of positive<br>holdings | Salmonella serovar |
|------------------------------|-----------------------|---------------------------|-----------------------------|--------------------|
| Grandparents                 |                       |                           |                             |                    |
| Layers and broilers          | 12                    | 2                         | 0                           |                    |
| Parents                      |                       |                           |                             |                    |
| Layers and broilers          | 819                   | 70                        | 0                           |                    |
| Turkeys                      | 42                    | 3                         | 0                           |                    |
| Ducks                        | 25                    | 2                         | 0                           |                    |
| Total – Breeders             | 898                   | 77                        | 0                           |                    |
| Other commercial poultry     |                       |                           |                             |                    |
| Pullets                      | 268                   | 25                        | 0                           |                    |
| Layers                       | 1,231                 | 641                       | 0                           |                    |
| Meat production - Broilers   | 4,051                 | 558                       | 0                           |                    |
| - Turkeys                    | 345                   | 63                        | 0                           |                    |
| - Ducks                      | 50                    | 10                        | 0                           |                    |
| Unknown                      | 121                   | 32                        | 0                           |                    |
| Total – Non breeder holdings | 6,066                 | 1,259                     | 0                           |                    |
| Total                        | 6,964                 | 1,310                     | 0                           |                    |

 Table 5. Number of swab samples from carcasses of cattle, swine and sheep and neck skin samples from poultry examined in the Salmonella surveillance and control programme in 2006

| Species |    | hterhouses<br>I (total*) | No. of samples<br>examined | No. of positive<br>samples | Salmonella serovar                         |
|---------|----|--------------------------|----------------------------|----------------------------|--|
| Cattle  | 31 | (38)                     | 2,035                      | 0                          |  |
| Swine   | 25 | (30)                     | 3,122                      | 0                          |  |
| Sheep   | 25 | (36)                     | 2,538                      | 1                          | S. enterica subsp. diarizonae (61:k:1,5,7) |
| Poultry | 6  | (6)                      | 5,420                      | 1                          | S. Anatum                                  |

\* Slaughterhouses where the number of slaughtered animals of a species is less than 100 according to the Slaughter Statistics for 2006 are not included.

Between 15 % and 25 % of the recorded human cases of salmonellosis are domestic in origin showing that domestic food products of animal origin represent a minor risk with regard to *Salmonella* infection in humans. In 2002 it was shown that two clones of *S*. Typhimurium in the wild fauna (wild birds and hedgehogs) represented a risk for human infection (12). Such wild animal reservoirs may also be considered a risk for farm animals. As no increase in prevalence of *Salmonella* has been demonstrated in the programme, it may be assumed that farm animal populations have been and still are well protected from these reservoirs.

The number of swab and lymph node samples examined per species should have been 3,000 per year. The required sample size was reached for the swine population, but not for the cattle and sheep populations. A follow up of the personnel taking and reporting the samples is needed. Never the less, the programme was able to document a very low *Salmonella* prevalence in the examined populations.



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# Residues in live animals and animal products in Norway

Dag Grønningen





# Introduction

Totally 3,579 samples (95.2 % of plan) from animals and primary animal products were collected in 2006. 69 samples (1.9 %) were classified as non-compliant, containing substances from groups: A2 Thyrostats, A3 Steroids, B2b Coccidiostats and B3c Heavy metals.

Since 1993, the surveillance programme has included bovine, porcine, sheep, poultry and reindeer products in accordance with EU Directive 86/469. In 1999 it was expanded to include live animals, milk, eggs, honey, and fish. The number of samples and substances tested in the programme was at the same time substantially increased in accordance with EU Directive 96/23 (1) and EU Decision 97/747/EC (2).

The programme for surveillance of residues in live animals, fish, and animal products were taken over by the Norwegian Food Safety Authority from 1 January 2004. Each year's programme produces a report on land animals and fish seperately. National Veterinary Institute publishes the land animals' part (3). The results of fish and products thereof are reported by National Institute of Nutrition and Seafood Research (in Norwegian 4).

# Aims

The aim of the present programme is to ensure food safety by monitoring the occurrence of residues of veterinary medicines, prohibited substances and environmental contaminants in live animals and animal products. The programme also provides data to satisfy export documentation requirements from the EU, USA and Switzerland.

# Regulations

To prevent consumption of animal products that contain potentially harmful residues, the Residue Control Regulation (RCR) was introduced in 2000 (5). This aims to prevent production, import and sale of products containing residues of prohibited substances, contaminants and veterinary drugs above Maximum Residue Limits (MRL). The legislation implements EU Directive 96/23 and requires control measures for any activity in agricultural and animal production.

The RCR determines MRLs for veterinary drugs. The use of veterinary drugs without MRLs in production animals is prohibited. In 2002 the EU introduced the phrase Minimum Required Performance Limit (MRPL) through Commission Decision 2002/657/EC (6). It is intended to harmonise the analytical performance of methods for substances for which no MRLs have been established or are prohibited.

Since the entry into force of Decision 2002/657/EC (1 September 2002), the correct term for those analytical results exceeding the permitted limits is "non-compliant". A non-compliant result means that the result has a sufficient statistical certainty (99 % for substance for which no MRLs has been established, and 95 % for all other substance) and can be used for legal purposes.

# Materials and methods

# Group of substances

EU regulations define the species (Table 1) and groups of substances (Appendix) to be included in the programme.

Samples of live animals (e.g. bovines, pigs, and poultry) are monitored for the presence of prohibited substances (Group A) only.

Each country may select the specific substances to be monitored. In Norway this is based on data from the Norwegian Medical Agency, as well as advice from the Norwegian School of Veterinary Science, Aker University Hospital and the National Veterinary Institute.

## Sampling plan

The sampling plan for the various animal species and products is determined on the basis of earlier produc-tion (Table 1). The plan is designed to ensure an even sampling throughout the year and throughout the coun-try. Information on each sample is registered in a proto-col at the time of sampling and sent to the central registration unit.

| Categories | Production       |
|------------|------------------|
| Bovine     | 333,653 *        |
| Porcine    | 1,458,430 *      |
| Sheep      | 1,279,552 *      |
| Equine     | 1,984 *          |
| Reindeer   | 2,290 tons       |
| Wild game  | 91,930 animals   |
| Poultry    | 54,011 tons      |
| Milk       | 1,540 mill litre |
| Eggs       | 52,274 tons      |
| Honey      | 886 tons         |

Table 1. The number of animals slaughtered and

production figures for animal products in Norway in 2004

\* Total number of approved carcasses.

# Surveillance and control programmes in Norway · Residues in live animals and animal products · Annual report 2006

#### Laboratory analysis

Samples are analysed within three months of sampling. Values exceeding MRLs and any prohibited substances detected are reported immediately.

All analyses are carried out by national reference laboratories. The Norwegian laboratories are accredited by the Norwegian Accreditation and thereby meet the requirements of the standard ISO/IEC 17025. Substances A1, A3, A4, A5 and B2d are analysed at the Hormone Laboratory, Aker University Hospital. Substances A2 are analysed at Ghent University, Belgium. Substances A6, B1, B2b, B2e, and B2f are analysed at the Laboratory for Veterinary Drug Residue Analysis in Food, the Norwegian School of Veterinary Science (NVH). Substances B2a and B2c are analysed at the Laboratory for Analysis of Veterinary Drugs, NVH. Substances B3a and B3b are analysed at the Laboratory of Environmental Toxicology, NVH, and the Bioforsk Lab, Ås. Substances B3c and B3d are analysed at the Section of Chemistry, National Veterinary Institute.

# **Results and comments**

#### General

It was planned to collect 3,758 samples in 2006. Totally 3,579 samples from animals and primary animal products were collected. 69 samples (1.9 %) were classified as non-compliant.

The report (in Norwegian) delivered to the Norwegian Food Safety Authority contains a more detailed description of the substance being analysed, the laboratory methods, and the results (7).

#### Live animals

Table 2 presents an overview of the number of samples tested in 2006 grouped according to substances, and number of non-compliant samples.

#### Thyrostatics

2-thiouracil was detected in tree samples of bovines, and 5 samples of pigs. See a possible explanation under Animal products; Thyrostatics. Norway considers this as non-compliant laboratory samples.

#### Steroids

17-alfa-nondrelon was detected in two samples of pregnant bovines. The literature tells that bovines produce this substance during the state of pregnancy (8). Norway considers this as non-compliant laboratory samples.

#### Animal products

Table 3 presents an overview of the number of animal products sampled in 2005.

#### Thyrostatics

There was detected 2-thiouracil in 8 samples of bovines, 16 samples of pigs, and two samples of sheep. 2-mercaptobenzimidazol was detected in one duck. This substance has not been detected in Norway before. A possible explanation maybe that the laboratory has developed a method that detects a possible background of natural occurrence of thyrostats in animals fed with cruciferous (9). Norway considers this as non-compliant laboratory samples.

#### Coccidiostats

Trace amount of narasin was detected in two samples of eggs. Administration of this substance to egg producing hens is prohibited. Narasin does not have an established MRL nor MRPL. Norway considers this as non-compliant laboratory samples.

#### Heavy metals

Residues of cadmium exceeding MRLs were detected in 13 samples of bovine, 16 samples of sheep, and in one sample of horse.

|                              | Bov | ines | Pi | igs  | Pou | ltry |
|------------------------------|-----|------|----|------|-----|------|
| Substances                   | No  | NCom | No | NCom | No  | NCom |
| A1 Stilbenes                 | 70  |      | 9  |      | 5   |      |
| A2 Thyrostatics              | 39  | 3    | 9  | 5    | 1   |      |
| A3 Steroids                  | 75  | 2    | 9  |      | 2   |      |
| A4 Resorcyclic acid lactones | 76  |      | 11 |      | 1   |      |
| A5 Beta-agonists             | 76  |      | 7  |      | 3   |      |
| A6 Annex IV substances*      | 0   |      | 16 |      | 28  |      |
| Total A                      | 336 | 5    | 61 | 5    | 40  | 0    |

Table 2. The number of live animals tested in 2006

\*A6: Annex IV: chloramphenicol; nitrofuranes; dimetridazole, metronidazole and ronidazol. No: Number of animals tested in 2006.

NCom: Number of non-compliant animals.

|  | Bov | Bovines | Pi  | Pigs | She | Sheep | Hor | Horses | Poultry | ltry | Reindeer | deer | Milk | ¥    | Eg  | Eggs | Honey | ley  | Wildgame | ame  |
|--|-----|---------|-----|------|-----|-------|-----|--------|---------|------|----------|------|------|------|-----|------|-------|------|----------|------|
| Substances                               | No. | NCom    | No. | NCom | No. | NCom  | No. | NCom   | No.     | NCom | No.      | NCom | No.  | NCom | No. | NCom | No.   | NCom | No.      | NCom |
| A1 Stilbenes                             | 70  |         | 49  |      | 27  |       |     |        | 11      |      | 4        |      |      |      |     |      |       |      |          |      |
| A2 Thyrostatics                          | 44  | ω       | 19  | 16   | 7   | 2     |     |        | 10      |      | 2        |      |      |      |     |      |       |      |          |      |
| A3 Steroids                              | 95  |         | 45  |      | 19  |       |     |        | 11      |      | 2        |      |      |      |     |      |       |      |          |      |
| A4 Resorcyclic acid<br>lactones          | 76  |         | 39  |      | 24  |       |     |        | 6       |      | 2        |      |      |      |     |      |       |      |          |      |
| A5 Beta-agonists                         | 73  |         | 28  |      | 21  |       |     |        | 16      |      | 9        |      |      |      |     |      |       |      |          |      |
| A6 Annex IV<br>substances*               | 58  |         | 33  |      | 18  |       |     |        | 118     |      | 9        |      | 49   |      | 10  |      | 5     |      |          |      |
| Total A                                  | 416 | ∞       | 213 | 16   | 116 | 2     |     |        | 175     | -    | 22       | 0    | 49   | 0    | 10  | 0    | 2     | 0    |          |      |
| B1 Tiamulin (pigs);<br>penicillin (milk) |     |         | 27  |      |     |       |     |        |         |      |          |      | 31   |      |     |      |       |      |          |      |
| B1 Enrofloxaim                           | 25  |         | 20  |      | 22  |       |     |        |         |      |          |      |      |      |     |      |       |      |          |      |
| B1 Sulfonamides                          | 25  |         | 30  |      | 30  |       |     |        | 57      |      | 12       |      | 50   |      | 39  |      |       |      |          |      |
| B1 Tetracyclines                         | 10  |         |     |      |     |       |     |        | 61      |      | 10       |      | 39   |      | 39  |      | Ð     |      |          |      |
| Total B1                                 | 09  | 0       | LT  | 0    | 52  | 0     |     |        | 118     | 0    | 22       | 0    | 120  | 0    | 78  | 0    | വ     | 0    |          |      |
| B2a Anthelmintics                        | 119 |         | 75  |      | 119 |       |     |        | 13      |      | 22       |      | 47   |      |     |      |       |      |          |      |
| B2b Anticoccidials                       | 10  |         | 10  |      | 20  |       |     |        | 92      |      | 2        |      |      |      | 49  | 2    |       |      |          |      |
| B2c Carbamates<br>and pyrethroides       | 34  |         | 10  |      | 32  |       |     |        | œ       |      | 6        |      |      |      | 20  |      |       |      |          |      |
| B2d Sedatives                            | 28  |         | 29  |      | 29  |       |     |        |         |      |          |      |      |      |     |      |       |      |          |      |
| B2e NSAIDs                               | 40  |         | 20  |      | 6   |       | 30  |        | 6       |      | 2        |      | 6    |      |     |      |       |      |          |      |
| B2f Glucorticoids                        | 21  |         | 20  |      | -   |       | 15  |        |         |      |          |      | 17   |      |     |      | 10*   |      |          |      |
| Total B2                                 | 252 | 0       | 164 | 0    | 210 | 0     | 45  | 0      | 122     | 0    | 35       | 0    | 73   | 0    | 69  | 2    | 10    | 0    |          |      |
| B3a Organochlorine<br>compounds          | 16  |         | 19  |      | 15  |       |     |        | 6       |      | 6        |      | 20   |      | 20  |      |       |      |          |      |
| B3b Organophos-<br>phorous compounds     | 20  |         | 20  |      | 15  |       |     |        | ę       |      |          |      | 20   |      |     |      |       |      |          |      |
| B3c Chemical elements                    | 47  | 13      | 25  |      | 39  | 16    | 25  | -      | 4       |      | 46       | 31   | 24   |      | 20  |      | 2     |      | 66       | 45   |
| B3d Mycotoxins                           | 13  |         | 20  |      | 53  |       |     |        | 2       |      |          |      | 46   |      |     |      |       |      |          |      |
| Total B3                                 | 96  | 13      | 84  | 16   | 122 | 16    | 25  | -      | 21      | 0    | 55       | 31   | 110  | 0    | 40  | 0    | ß     |      | 66       | 45   |
| Total B                                  | 408 | 13      | 325 | 16   | 384 | 16    | 70  | -      | 261     | 0    | 112      | 31   | 303  | 0    | 187 | 2    | 20    | 0    | 66       | 45   |
| Total ∆⊥R                                | 824 | 21      | 538 | 16   | 500 | 18    | 70  | -      | 436     | -    | 134      | 31   | 352  | 0    | 197 | 2    | 25    | 0    | 99       | 45   |

Table 3. The total number of animal products/foods in the surveillance and control programme in 2006

No MRLs have been established for wild game. If we use MRLs set for bovine: Residues of cadmium in 31 reindeer, 29 elk, 12 roe deer, and 4 red deer were detected. Norway considers all residues of cadmium exceeding MRLs as noncompliant laboratory samples.

Chemical elements accumulate in organs throughout life as a result of environmental pollution, particularly in free ranging animals (farmed and wild game, sheep).

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# Appendix

# Group A – Substances having anabolic effect and unauthorized substances

- 1. Stilbenes, stilbene derivatives, salts and esters
- 2. Thyrostatics
- 3. Steroids
- 4. Resorcyclic acid lactones
- 5. Beta-agonists
- 6. Annex IV substances. (incl. chloramphenicol, nitrofuranes, dimetridazole and metronidazol)

# Group B – Veterinary drugs and contaminants

- 1. Antibacterial substances, (incl. sulphonamides, fluoroquinolones)
- 2. Other veterinary drugs
  - a. Anthelmintics
  - b. Anticoccidials
  - c. Carbamates and pyrethroids
  - d. Sedatives
  - e. NSAIDs
  - f. Other pharmacologically active substances
- 3. Environmental contaminants and other substances
  - a. Organochlorine compounds, incl PCBs
  - b. Organophosphorus compounds
  - c. Chemical elements
  - d. Mycotoxins



The surveillance and control programmes for paratuberculosis in Norway



Annette Hegermann Kampen Berit Djønne Ola Nyberg

Paratuberculosis was first diagnosed in cattle and goats in Norway in 1907 and 1934, respectively (1, 2). *Mycobacterium avium* subsp. *paratuberculosis* infection is a notifiable disease (List B) in ruminants in Norway. The control of this disease in cattle is enforced by government legislation. Confirmation of infection most often results in the culling of the herd. Affected herd owners are compensated by the government, which also covers the expenses involved in testing. In goat flocks, government restrictions combined with vaccination are used to control paratuberculosis. Vaccination is performed using an inactivated vaccine (3).

A national surveillance and control programme for paratuberculosis was established in 1996 (4, 5, 6).

Descriptions of occurrence of the disease in Norway, control measures taken up to 1995, and results from the surveillance and control programmes from 1996 to 2001, can be found in the annual report for 2001 (5).

### Aim

The aim of the surveillance programme for paratu-berculosis is to estimate the prevalence of the infection in the Norwegian population of vaccinated goats. In addition, cattle, goats from unvaccinated flocks, sheep and llamas in limited numbers are screened for infection with *M. a. paratuberculosis*.

# Materials and methods

Cattle, goats, sheep and llamas were included in the programme in 2006. Faecal samples were collected on the farms by the Norwegian Food Safety Authority, while organ samples were collected at slaughterhouses.

#### Active surveillance

#### Cattle

The target population consisted of all cattle herds delivering milk to dairies in the sampling period and all beef cattle herds receiving state support according to records of July 2005. One hundred and one herds were randomly selected for sampling. Faecal samples were collected from the five oldest cows in each herd.

#### Goats

One hundred and ten vaccinated and twenty unvaccinated flocks were selected for sampling. Faecal samples were taken from the ten oldest goats, or from sick goats.

#### Sheep

Twenty flocks from the areas where goat kids are vaccinated and where paratuberculosis is registered in goat herds were randomly selected. Faecal samples from the ten oldest sheep, or from sick sheep were collected.

#### Llamas

Llama was introduced as a new species to Norway in 1997-98. A few animals have been imported from Sweden and from South America over the last years. Faecal samples from all animals over four years of age are collected each year. In addition, organ samples are collected from Ilamas at slaughter and from animals that die when over four years of age.

#### Herds with restrictions

Infected herds and contact herds are included in the programme as well.

#### Passive clinical surveillance

Clinical surveillance has been a part of the programme since 2000. For cattle, special emphasis is placed on the collection of samples from animals with reduced milk production, loss of weight, diarrhoea lasting more than 14 days, and animals that are over four years of age.

#### Sampled herds and animals

A total of 518 faecal samples and six organ samples were collected from cattle, while 1,130 faecal samples and two organ samples were collected from goats. A total of 212 faecal samples and two organ samples were collected from sheep, and 81 faecal samples were collected from llamas (Table 1).

#### Histopathological examination

Samples from jejunum, ileum, ileocecal valve, and mesenteric lymph nodes were examined histopathologically. The tissue was fixed in 10 % neutral-buffered formalin, processed by routine methods and stained with haematoxylin and eosin (HE) and the Ziehl-Neelsen (ZN) method for acid-fast bacteria.

#### Bacteriological examination

The samples were decontaminated with 4 % sodium hydroxide and 5 % oxalic acid with 0.1 % malachite green (7), and inoculated onto selective and non-selective Dubos medium with mycobactin (2  $\mu$ g/ml) and pyruvate (4 mg/ml) (8). Incubation time was 16 weeks.

Mycobactin dependency, acid-fastness by Ziehl-Neelsen staining, and presence of the insertion segment IS*900* by a PCR technique (9) were used to identify the isolates.

### Results

#### Histopathological examination

Formalin-fixed tissue samples from six cattle from two different herds were examined with no positive results (Table 2).

Two goats from an infected flock were examined. Granulomatous lesions were found in lymph nodes of one goat, but acid fast bacteria were not detected (Table 2).

Two sheep from two contact flocks were examined. Granulomatous lesions and acid fast bacteria were found in the intestines and lymph nodes of one sheep (Table 2).

Samples from one Ilama were submitted in 2006, but the material was unsuitable for histopathological examination (Table 2).

#### Bacteriological examination

A total of 518 cattle in 96 herds were examined for paratuberculosis by bacteriological methods (Table 2). *M. a. paratuberculosis* was not found.

A total of 1,130 dairy goats from 113 flocks were ex-amined for paratuberculosis by bacteriological methods (Table 2). *M. a. paratuberculosis* was isolated from two goats in two new flocks. The kids in these flocks were vaccinated against paratuberculosis since 1992-1993.

An overview of the Norwegian goat population, the density of sampled goats in different areas, and herds with restrictions, are shown in Figure 1.

A total of 212 sheep from 21 flocks were examined for paratuberculosis by bacteriological methods. *M. a. paratuberculosis* was isolated from two sheep from two different herds (Table 2).

A total of 81 Ilamas from 20 herds were examined for paratuberculosis by bacteriological methods (Table 2). *M. a. paratuberculosis* was not isolated.

### Discussion

Since the surveillance programme for paratuberculosis started in 1996, infection with *M. a. paratuberculosis* has been detected in nine cattle herds, four sheep flocks and in 26 goat flocks.

The infection is endemic among goats in six out of 19 counties in Norway. All the cases among cattle and sheep can be attributed to one of two reasons; either brought into the country with imported cattle (seven cattle herds, one sheep flock) or contact with infected goats (two cattle herds, three sheep flocks). Importation of live cattle nearly stopped after 1996 and has been replaced by importation of semen and embryos. But importation of sheep and goats may together with the presence of infected goat flocks represent a risk for spread of the infection to other ruminants.

The total number of milking goats in Norway is 42,500 in 510 flocks. In the six counties with endemic paratuberculosis, there are 237 flocks. Thirty-nine flocks (16 %) were recorded as infected with M. a. paratuberculosis in this area by the end of 2006, and have been given restrictions by the veterinary authorities. The infection was recorded in two new goat herds and two new sheep flocks this year. The infected sheep flocks had close contact had close contact with previously infected goat herds. It is probable that even more flocks are infected because vaccination hides the symptoms. The surveillance programme for 2006 therefore gave priority to samples from vaccinated goat flocks while cattle and sheep were sampled less. By following this priority over a few years, our prevalence estimate could possibly come closer to the true pre-valence in the endemic area. This could be very useful in the near future, because the dairy organisation (TINE) and the Norwegian Goat Health Services have started an eradication programme for three widespread infectious diseases in goats.

The programme started in 2001 and concentrated on caprine arthritis encephalitis and caseous lymphadenitis the first years. From 2005 they included herds with goats infected with paratuberculosis as well.

|        |   | Faecal samples no. of animals | Intestinal samples<br>no. of animals | Total no. of animals | Total no.<br>of herds |
|--------|---|-------------------------------|--------------------------------------|----------------------|-----------------------|
| Cattle | Random sample                                 | 468                           | 0                                    | 468                  | 92                    |
|        | Suspected or imported cases                   | 1                             | 1                                    | 2                    | 2                     |
|        | Control of infected herds and contact herds   | 49                            | 5                                    | 54                   | 2                     |
| Goat   | Vaccinated                                    | 840                           | 0                                    | 840                  | 84                    |
|        | Unvaccinated                                  | 290                           | 0                                    | 290                  | 29                    |
|        | Suspected cases                               | 0                             | 0                                    | 0                    | 0                     |
|        | Control of infected flocks and contact flocks | 0                             | 2                                    | 2                    | 1                     |
| Sheep  | Random sample                                 | 196                           | 0                                    | 196                  | 20                    |
|        | Control of infected flocks and contact flocks | 16                            | 2                                    | 18                   | 2                     |
| Llama  |   | 81                            | 1*                                   | 82                   | 20                    |

Table 1. Number of samples collected for examination for Mycobacterium avium subsp. paratuberculosis in 2006

\*Unsuitable for examination

| Table 2. R | esults of | histopathological | and bacteriological | examination of cattle in 2006 |
|------------|-----------|-------------------|---------------------|-------------------------------|
|            |           |                   |                     |                               |

|         |                    |                | Bacteriology |                        |                | Histopathology |                        |  |  |
|---------|--------------------|----------------|--------------|------------------------|----------------|----------------|------------------------|--|--|
| Species | Type of samples    | No. of samples | No. of herds | No. of pos.<br>samples | No. of samples | No. of herds   | No. of pos.<br>samples |  |  |
| Cattle  | Faeces             | 518            | 96           | 0                      |                |                |                        |  |  |
|         | Intestinal samples | 6              | 2            | 0                      | 6              | 2              | 0                      |  |  |
| Goat    | Faeces             | 1,130          | 113          | 2                      |                |                |                        |  |  |
|         | Intestinal samples | 2              | 1            | 0                      | 2              | 1              | 0                      |  |  |
| Sheep   | Faeces             | 212            | 21           | 1                      |                |                |                        |  |  |
|         | Intestinal samples | 2              | 2            | 1                      | 2              | 2              | 1                      |  |  |
| Llama   | Faeces             | 81             | 20           | 0                      |                |                |                        |  |  |
|         | Intestinal samples | 0              | 0            | 0                      | 0              | 0              | 0                      |  |  |

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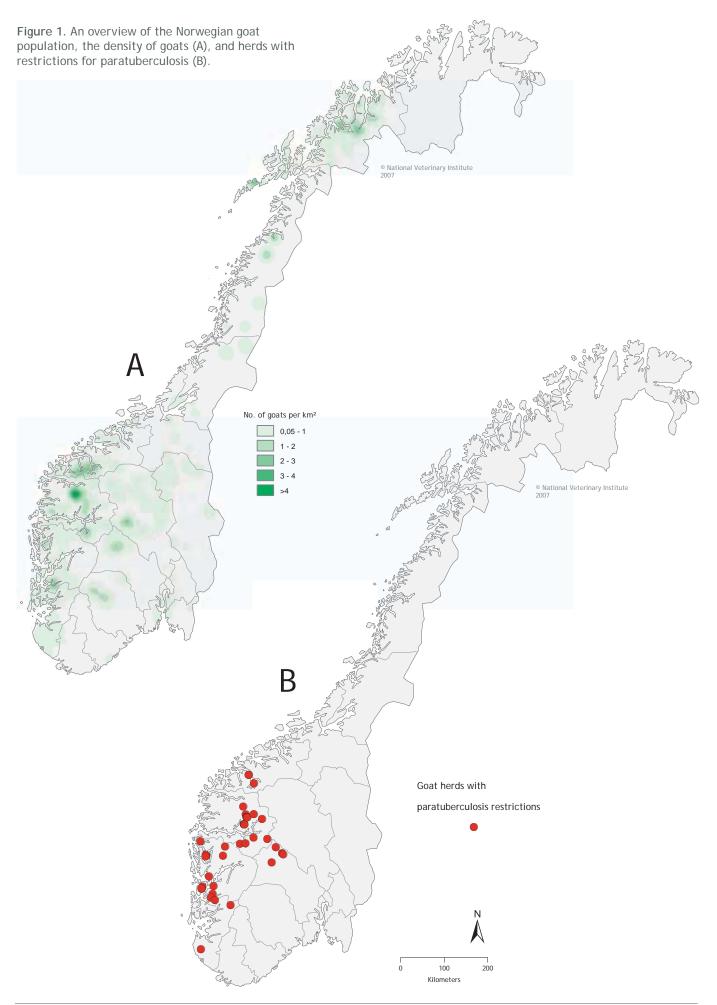
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The surveillance and control programme for bovine spongiform encephalopathy (BSE) in Norway

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# All 20,556 samples originated from 10,825 dairy cattle and beef herds were tested negative for BSE in 2006.

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 enzymed-linked immunosorbent assay (ELISA) method for detection of resistant prion protein (PrP<sup>sc</sup>) (Platelia® BSE ELISA Bio-Rad was replaced by TeSE® ELISA Bio-Rad in June 2003). In addition, clinically suspected animals were investigated by histopathological examination according to the Office International des Epizooties (OIE) protocol (1, 2). The number of samples examined in each category in the period 1998-2005 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 2006 the surveillance programme was in accordance with the Commission Regulations (EC) No 999/2001, No 1188/2003 and No 1915/2003. The programme included examination of the following categories:

- clinically suspected animals irrespective of age
- all animals older than 24 months of age, which have died or been culled, but not slaughtered for human consumption (fallen stock)
- all emergency slaughtered animals older than 24 months
- all animals 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 animals with unknown age or origin irrespective of age
- all imported cattle from any country irrespective of age and the over 24 month old progeny of imported female cattle
- 10,000 randomly selected healthy routinely slaughtered animals older than 30 months

#### Implementation

The farmers were responsible for reporting to the Norwegian Food Safety Authority all cases of clinically suspected animals irrespective of age, fallen stock older than 24 months and when delivering an imported animal or progeny of an imported female animal to slaughter. Brain or head from clinically suspected cattle and fresh material from the *medulla oblongata* sampled from fallen stock were analysed to the National Veterinary Institute, Oslo. Inspectors from the Norwegian Food Safety Authority collected the 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 Sandnes, Trondheim or Harstad.

#### Laboratory methods

#### Clinically suspected animals

The whole brain was divided mid-sagittally into equal halves. One half was 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 PrP<sup>sc</sup> was 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 was prepared for ELISA to detect  $PrP^{Sc}$  (TeSeE<sup>®</sup>, Bio-Rad) as described by the manufacturer.

Table 1. Examination for BSE in cattle sampled by the Norwegian surveillance programme according to categories from1998-2005

| Reason for submission to the laboratory | 1998-2000 | 2001   | 2002   | 2003   | 2004   | 2005   |
|---|-----------|--------|--------|--------|--------|--------|
| Clinically suspected animals            | 78        | 14     | 2      | 2      | 3      | 1      |
| Fallen stock                            |           | 1,352  | 1,482  | 1,872  | 2,145  | 2,318  |
| Emergency slaughtered                   |           | 7,073  | 7,246  | 7,322  | 9,217  | 8,462  |
| Ante-mortem animals                     |           | 2,612  | 3,562  | 4,102  | 1,355  | 102    |
| Imported slaughtered animals            | 19*       | 88     | 39     | 39     | 24     | 10     |
| Healthy slaughtered animals             |           | 2,400  | 9,907  | 10,726 | 10,443 | 10,486 |
| Total                                   | 97        | 13,539 | 22,238 | 24,063 | 23,187 | 21,379 |

\* All the samples were examined in 2000.

Table 2. Examination for BSE in cattle sampled by the Norwegian surveillance programme according to category in 2006

| Reason for submission to the laboratory | No. of samples | No. of<br>rejected samples | Negative | Positive |
|---|----------------|----------------------------|----------|----------|
| Clinically suspected animals            | 0              | 0                          | 0        | 0        |
| Fallen stock                            | 2,364          | 39                         | 2,325    | 0        |
| Emergency slaughter                     | 8,177          | 14                         | 8,163    | 0        |
| Ante-mortem animals                     | 36             | 0                          | 36       | 0        |
| Imported animals                        | 4              | 0                          | 4        | 0        |
| Healthy slaughtered animals             | 10,455         | 4                          | 10,451   | 0        |
| Total                                   | 21,036         | 57                         | 20,979   | 0        |

\* Abnormal findings at ante-mortem examination, rejected for human consumption, or which died at the abattoir or during transport.

#### Risk population and routine slaughtered animals

Non-fixed brain tissue from the *obex* area was prepared for ELISA to detect  $PrP^{Sc}$  (TeSeE<sup>®</sup>, 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 specimen was processed for immunohistochemical detection of  $PrP^{Sc}$  using the same protocol as for specimens from clinical suspects.

Brain samples were rejected for examination if the specimen was 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 21,036 cattle. Of these, 57 (0.3 %) samples were un-suitable for examination. The categories and number of animals examined are presented in Table 2.

For 2.3 % of all samples, herd of origin was not reported. But in case of a positive test result from such a herd, identity can be traced via the carcass number. The remaining 20,556 samples originated from 9,317 dairy cattle herds and 1,508 beef cattle herds. The mean number of examined animals per herd was 1.9.

# Clinically suspected animals (passive surveillance)

In 2006, 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 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 0.97 % of the adult population (National Cattle Registry (Husdyrregisteret), per 31.12.2005), i.e. approximately 3,700 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 time consuming and cumbersome. In addition, a proportion of the cattle is grazing on mountain and forest pastures where sampling of dead animals is difficult. Further reasons may be insufficient information to the farmers relating to their duty to report all cases of fallen stock older than 24 months to the Norwegian Food Safety Authority.

Norwegian cows are slaughtered at a low age, mean age is approximately 50 months for dairy cows and 67 months for suckling cows (suckling cows constitute only 15 % of the cattle population older than 24 months) (National Production Recording Scheme 2004, Norwegian Beef Herd recording System 2004).

The low age at culling implies that 44 % of the samples originated from cattle younger than 4 years. The age distributions of cattle sampled in different categories are shown in Table 3.

Results from the BSE-monitoring programme in the EU 2005 show that only 8 (1.43 %) of 561 verified cases of BSE were younger than 48 months, and 0.04 positive cases were detected per 10,000 tests in cattle 36-47 months, in contrast to 0.57 in cattle 72-83 months (3). These results indicate that BSE-monitoring of animals younger than 48 months is of low value.

The geographical distribution of the cattle population and the animals of different categories tested are presented in table 4. Figure 1 indicates that there is a correlation between the collection of samples for emergency slaughter and healthy slaughtered animals from different regions and the distribution of the cattle population in the regions, but corresponding figures for the fallen stock population show considerable variation between regions.

|                        |                            | Relative number of tested animals |                               |                               |   |                        |  |
|------------------------|----------------------------|-----------------------------------|-------------------------------|-------------------------------|---|------------------------|--|
| Age groups<br>(months) | Total<br>population<br>(%) | Fallen<br>stock<br>(%)            | Emergency<br>slaughter<br>(%) | Ante mortem<br>animals<br>(%) | Healthy slaugh-<br>tered animals<br>(%) | Total<br>tested<br>(%) |  |
| < 24                   | 58.8                       | 2.2                               | 4.4                           | 2.9                           | 2.0                                     | 3.0                    |  |
| 24-29                  | 7.8                        | 11.7                              | 14.8                          | 5.7                           | 8.2                                     | 11.1                   |  |
| 30-35                  | 6.0                        | 12.0                              | 8.1                           | 11.4                          | 10.6                                    | 9.8                    |  |
| 36-47                  | 9.8                        | 18.9                              | 16.0                          | 31.4                          | 23.5                                    | 20.1                   |  |
| 48-59                  | 6.6                        | 17.6                              | 17.1                          | 17.1                          | 20.1                                    | 18.6                   |  |
| 60-71                  | 4.2                        | 14.5                              | 15.4                          | 8.6                           | 15.6                                    | 15.4                   |  |
| 72-83                  | 2.5                        | 9.8                               | 10.9                          | 14.3                          | 9.7                                     | 10.2                   |  |
| 84-95                  | 1.5                        | 6.4                               | 6.4                           | 2.9                           | 5.2                                     | 5.8                    |  |
| 96-107                 | 0.8                        | 3.3                               | 3.7                           | 2.9                           | 2.5                                     | 3.1                    |  |
| 108-119                | 0.5                        | 1.5                               | 1.5                           | 2.9                           | 1.3                                     | 1.4                    |  |
| 120-131                | 0.3                        | 1.1                               | 0.8                           | 0.0                           | 0.6                                     | 0.7                    |  |
| 132-143                | 0.2                        | 0.5                               | 0.4                           | 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                               | 0.4                           | 0.0                           | 0.2                                     | 0.3                    |  |
| Total no. of animals   | 924,379                    | 2,364                             | 8,177                         | 36                            | 10,455                                  | 21,032                 |  |

#### Table 3. Age distribution of cattle tested for BSE-agent in 2006

There were 2,137 samples (10.2 %) 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.

| Table 4 | Regional | distribution | of Norwegian | cattle and | cattle | tested fo | r BSF-adent  | in 2006  |
|---------|----------|--------------|--------------|------------|--------|-----------|--------------|----------|
|         | Negionar | anstribution |              | cattle and | cattic | icsicu iu | I DJL UYCIIL | 111 2000 |
|         |          |              |              |            |        |           |              |          |

|                              |                            | Relative number of tested animals |                               |                               |  |                        |  |  |
|------------------------------|----------------------------|-----------------------------------|-------------------------------|-------------------------------|--|------------------------|--|--|
| Counties                     | Total<br>population<br>(%) | Fallen<br>stock<br>(%)            | Emergency<br>slaughter<br>(%) | Ante mortem<br>animals<br>(%) | Healthy<br>slaughtered<br>animals<br>(%) | Total<br>tested<br>(%) |  |  |
| Oslo, Akershus, Østfold      | 4.5                        | 8.5                               | 5.5                           | 0.0                           | 4.1                                      | 5.1                    |  |  |
| Hedmark, Oppland             | 17.9                       | 13.1                              | 16.0                          | 27.8                          | 15.2                                     | 15.3                   |  |  |
| Buskerud, Vestfold, Telemark | 6.1                        | 8.9                               | 5.9                           | 2.8                           | 6.9                                      | 6.7                    |  |  |
| Rogaland og Agder            | 20.8                       | 27.8                              | 19.3                          | 0.0                           | 21.3                                     | 21.2                   |  |  |
| Hordaland, Sogn og Fjordane  | 10.5                       | 8.9                               | 10.5                          | 0.0                           | 12.4                                     | 11.2                   |  |  |
| Trøndelag, Møre og Romsdal   | 30.2                       | 24.8                              | 34.7                          | 33.3                          | 29.2                                     | 30.8                   |  |  |
| Nordland                     | 7.2                        | 4.7                               | 5.8                           | 30.6                          | 7.3                                      | 6.4                    |  |  |
| Troms og Finnmark            | 2.8                        | 3.3                               | 2.3                           | 5.6                           | 3.7                                      | 3.1                    |  |  |
| Total no. of animals         | 924,379                    | 2,364                             | 8,177                         | 36                            | 10,455                                   | 21,032                 |  |  |

There were 473 samples (2.3 %) from cattle with unknown region. These regions are assumed to be distributed following the region distribution of the cattle from known region within each target group.



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# Conclusion

As mentioned in the first BSE surveillance report in 2001 (4), the Norwegian cattle population has probably never 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. The compiled results from the surveillance and control programme for BSE in the years 2001 to 2006 (5) with app. 125,000 negative samples clearly support this view.

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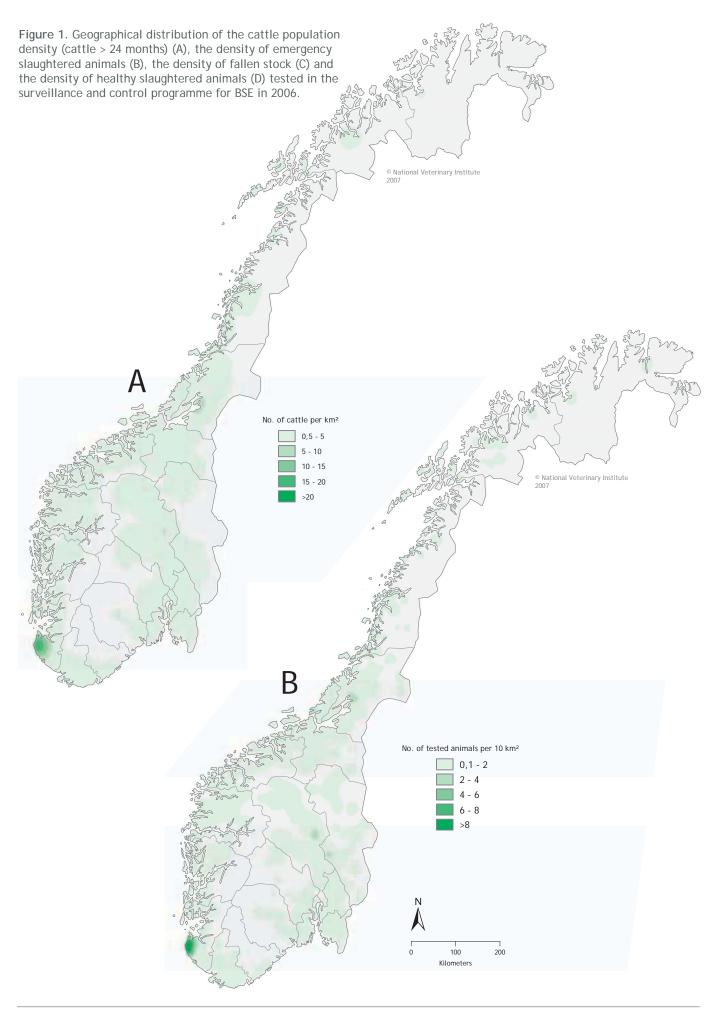
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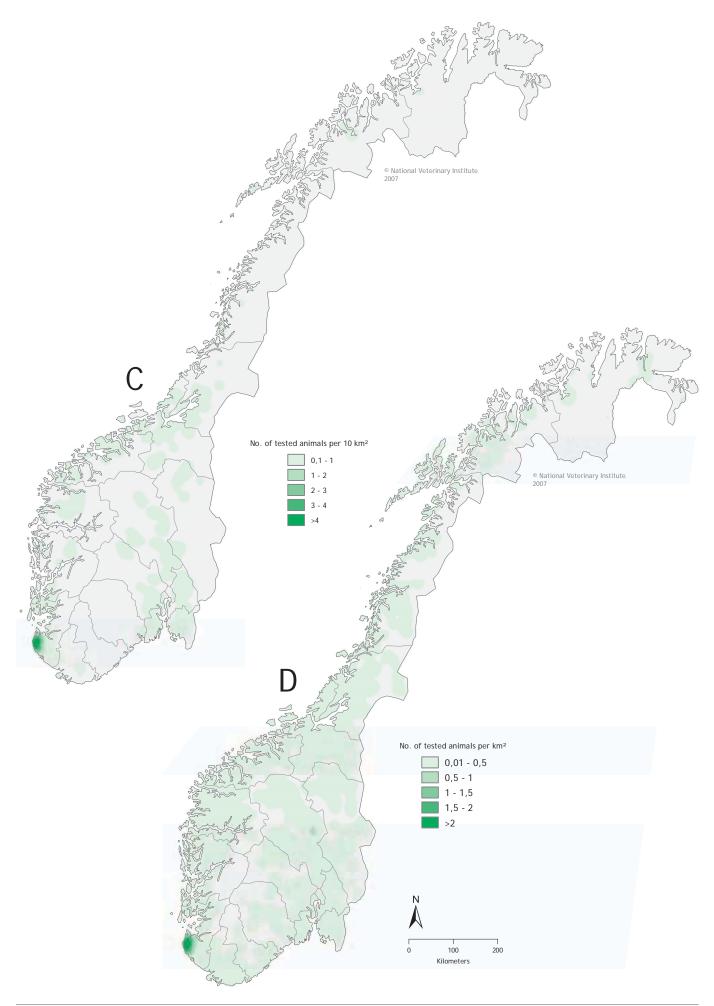
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The surveillance and control programme for infectious bovine rhinotracheitis (IBR) and infectious pustular vulvovaginitis (IPV) in Norway

Annette Hegermann Kampen Jorun Tharaldsen Gry Grøneng

All milk and blood samples tested in 2006 were negative for antibodies against bovine herpes virus (BHV-1).

In the early 1960s, two outbreaks of infectious pustular vulvovaginitis were diagnosed in cattle in Norway. Subsequently, no new cases of BHV-1 (infectious bovine rhinotracheitis/infectious pustular vulvovaginitis - IBR/IPV) were reported until 1993, when several animals in one single herd were found serologically positive after primary testing of bulk milk collected in 1992. Clinical signs of IBR/IPV however were never recorded on the farm. All animals on the farm were slaughtered. Attempts to isolate the virus from organ samples gave negative results. Sixteen contact herds and all dairy herds in the same region were serologically negative (1, 2). Likewise, 40 red deer that were shot in the neighbourhood during the hunting season the same year were serologically negative. After this incident, IBR/IPV irus infection has not been demonstrated in Norway.

EFTA Surveillance Authority (ESA) has recognised Norway as free from IBR since 1994. Decisions concerning the additional guarantees relating to IBR/IPV for bovines destined for Norway are described in ESA Decision 74/94/COL. Maintenance of the ESA Decisions accepting the IBR-free status of Norway requires annual reports of the surveillance of the disease.

The Norwegian Food Safety Authority is responsible for carrying out the surveillance and control programme for IBR/IPV. The National Veterinary Institute is in charge of planning the programme, collecting the bulk milk samples from the dairies and performing the tests. Blood samples from beef herds are collected by inspectors from the Norwegian Food Safety Authority.

### Aims

The aim of the surveillance and control programme for IBR/ IPV is to document freedom from the infection in Norway according to the demands in ESA Decision 74/94/COL with amendments, and to contribute to the maintenance of this favourable situation.

# Material and methods

The surveillance of cattle for IBR/IPV in 2006 included both dairy and beef herds. Bulk milk samples from the dairy herds were provided by the dairies. From the beef herds, individual blood samples were collected on the farms from cattle older than 24 months.

The target population consisted of all cattle herds delivering milk to the dairies in the sampling period. In 2006, bulk milk samples from 1,673 randomly sampled dairy herds were tested. The group of beef herds to be sampled was based on a register of all beef herds receiving governmental support according to recordings of July 2005. A total of 4,624 individual blood samples from 479 beef herds were analysed in pools with a maximum of 20 samples in each. The sampled herds represented approximately 11.4 % of the Norwegian cattle herds.

The number of herds in the surveillance and control programme for IBR/IPV in 2006 is given in Table 1. The geographic distribution of the total number and the number of tested dairy and beef herds are shown in Figures 1 and 2.

All samples were tested for antibodies against bovine herpes virus 1 (BHV-1) using a blocking ELISA (3) at the National Veterinary Institute in Oslo.

### **Results**

All 1,673 bulk milk samples and 4,626 blood samples tested in 2006 were negative for antibodies against BHV-1. Table 2 shows the results of the testing during the period from 1993 to 2006.

# Discussion

The surveillance and control programme for IBR/IPV has been evaluated and shown to have a sensitivity of more 82.9 % when used for bulk milk testing. The test sensitivity is better when testing serum samples, - the specificity estimated to 100 % (4).

The results of the programme since 1992/93 strongly indicate that the Norwegian cattle population is free from IBR/IPV-infection (2, 4, 5).

In addition to the surveillance programme all breeding bull candidates are serologically tested before entering the breeding centres, and all breeding bulls are subject to a compulsory test each year.

Table 1. Total number of dairy herds and beef herds withinthe frame of the Norwegian surveillance and controlprogramme for IBR/IPV in 2006

| Herd category | Total no. of cattle herds* | No. of herds<br>tested | % tested of<br>the total no.<br>of herds |
|---------------|----------------------------|------------------------|--|
| Dairy herds   | 14,800                     | 1,673                  | 11.3                                     |
| Beef herds    | 4,100                      | 479                    | 11.7                                     |
| Total         | 18,900                     | 2,152                  | 11.4                                     |

\* Based on data from the Register of production subsidies as of July 31 2006.

Table 2. Samples in the surveillance and control programmefor IBR/IPV in the Norwegian bovine population during theperiod 1993-2006

|      | Dairy herds                           | Beef                            | herds                           |                               |
|------|---------------------------------------|---------------------------------|---------------------------------|-------------------------------|
| Year | No. of bulk<br>milk samples<br>tested | No. of<br>beef herds<br>sampled | No. of<br>individuals<br>tested | No. of<br>positive<br>samples |
| 1993 | 26,642                                | 0                               | 0                               | 1                             |
| 1994 | 24,832                                | 1,430                           | 5,954                           | 0                             |
| 1995 | 25,131                                | 1,532                           | 9,354                           | 0                             |
| 1996 | 2,863                                 | 303                             | 1,523                           | 0                             |
| 1997 | 2,654                                 | 2,214                           | 16,741                          | 0                             |
| 1998 | 2,816                                 | 2,191                           | 17,095                          | 0                             |
| 1999 | 2,930                                 | 2,382                           | 18,274                          | 0                             |
| 2000 | 1,590                                 | 340                             | 2,892                           | 0                             |
| 2001 | 2,564                                 | 434                             | 3,453                           | 0                             |
| 2002 | 2,308                                 | 462                             | 3,693                           | 0                             |
| 2003 | 1,845                                 | 449                             | 3,901                           | 0                             |
| 2004 | 1,573                                 | 402                             | 3,364                           | 0                             |
| 2005 | 1,919                                 | 484                             | 4,766                           | 0                             |
| 2006 | 1,673                                 | 479                             | 4,624                           | 0                             |

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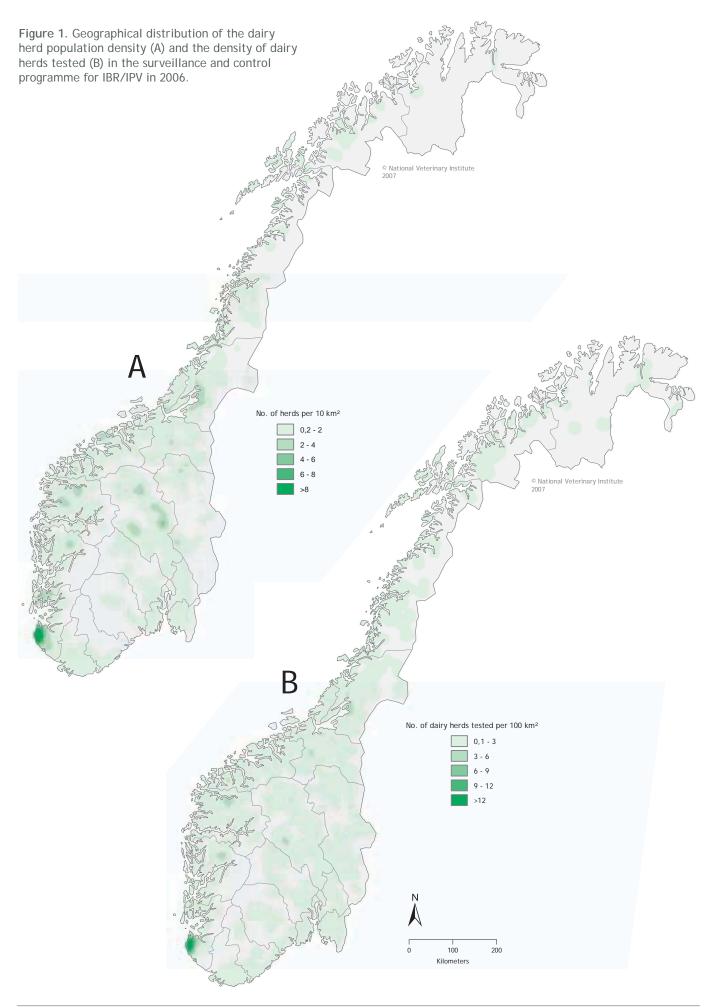
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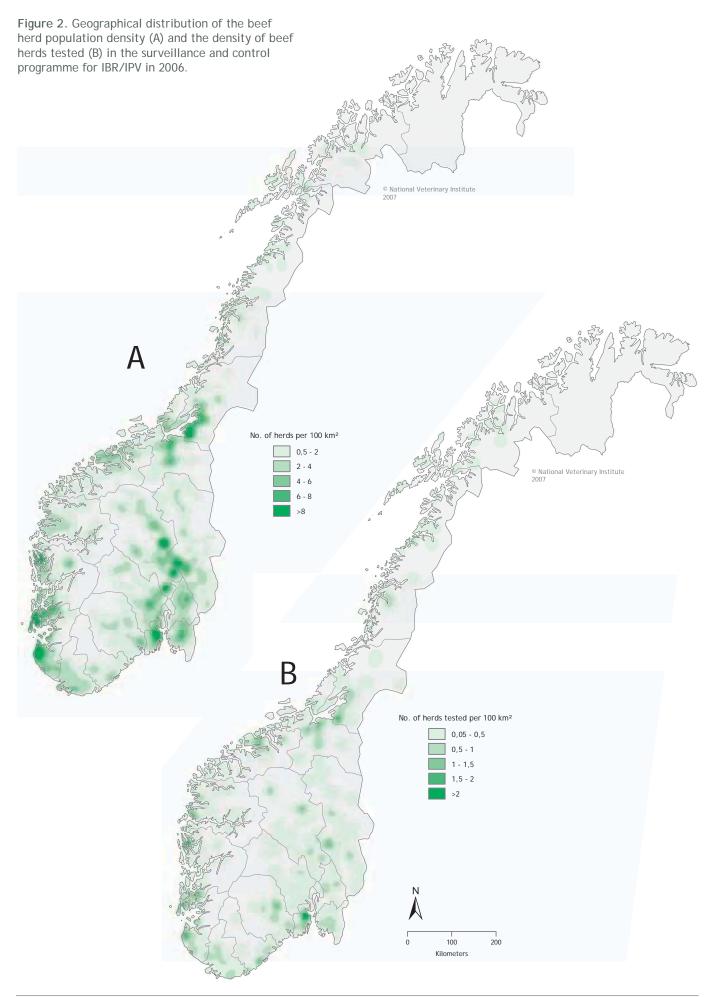
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Surveillance and control programmes in Norway · IBR/IPV · Annual report 2006



Surveillance and control programmes in Norway · IBR/IPV · Annual report 2006



Annual report 2006



The surveillance and control programme for enzootic bovine leukosis (EBL) in Norway

Annette Hegermann Kampen Jorun Tharaldsen Gry Grøneng

All milk and blood samples tested in 2006 were negative for antibodies against bovine leukaemia virus (BLV).

Enzootic bovine leukosis (EBL) had never been reported in Norway until antibodies against BLV were detected in eight dairy herds in samples collected through the surveillance and control programme in 1995 (1) (Figure 1A). No new herds tested positive during the period 1997-2001 (2).

In 2002, a bulk milk sample from one dairy herd tested positive for BLV (Figure 1A). Further investigations showed that only one of the cows in the herd was antibody positive. The cow, which was healthy and had no clinical signs, was slaughtered and pathological investigations gave no indication of leukosis. Further testing of individual blood samples of all cattle older than 24 months in the affected herd and six contact herds was negative. The conclusion was that the positive antibody test was a false positive. The follow-up study was terminated in 2003 with no further positive findings (3, 4). An application for EBL-free status according to Council Directive 64/432/EEC as amended was submitted to the EFTA Surveillance Authority in 2004.

The Norwegian Food Safety Authority is responsible for carrying out the surveillance and control programme for EBL. The National Veterinary Institute is in charge of planning the programme, collecting the bulk milk samples from the dairies, and performing the tests. Official inspectors from the Norwegian Food Safety Authority collected the blood samples from the beef herds.

#### Aims

The aim of the surveillance and control programme for EBL is to document freedom from the infection in Norway and to contribute to the maintenance of this favourable situation.

# Materials and methods

The surveillance and control programme included both dairy and beef herds. Bulk milk samples from the dairy herds were collected from the dairies. From the beef herds, individual blood samples were collected on the farms from cattle older than 24 months.

The target population of dairy herds are all cattle herds delivering milk to dairies during the sampling period. In 2006, bulk milk samples from 1,673 randomly sampled dairy herds were tested for antibodies against BLV. The target population of beef herds is all beef herds receiving governmental support according to recordings of July 2005. A total of 4,624 individual blood samples from 479 beef herds were analysed in pools, with a maximum of 20 samples in each. The sampled herds represented approximately 11.4 % of the Norwegian cattle herds (Table 1).

The geographic distribution of the total number of herds and the tested number of dairy and beef herds are given in Figures 1B, 2A and 2B.

Bulk milk samples and blood samples were examined by an indirect ELISA (SVANOVA®) (5). For verification and for follow-up of suspect cases, LACTELISA BLV Ab and SERELISA BLV Ab from SYNBIOTICS were used.

### Results

All bulk milk samples and blood samples tested in 2006 were negative for antibodies against BLV. Table 2 shows the results of the testing during the period from 1993 to 2006.

# Discussion

The requirement from the EU for granting an EBL freestatus is that the herd prevalence must be lower than 0.2 %, which represents 38 herds out of the total number of 18,900 herds.

No new cases have been reported since 1995 and the continuous surveillance since 1995 shows that the Norwegian cattle population is free from EBL according to the requirements (2, 3, 4, 6). From 1995 to 1999, all cattle herds were tested annually. Since 2000, a minimum of 10 % of dairy and beef cattle herds have been tested each year.

Together with the possible isolation period of six months and the testing protocol for imported animals, the surveillance and control programme for EBL should be an effective means to detect introduction of new infection.

Table 1. Total number of dairy herds and beef herds withinthe frame of the Norwegian surveillance and controlprogramme for EBL in 2006

| Herd category | Total no.<br>of cattle<br>herds* | No. of<br>herds<br>tested | % tested<br>of the<br>total no. of<br>herds |
|---------------|----------------------------------|---------------------------|---|
| Dairy herds   | 14,800                           | 1,673                     | 11.3  |
| Beef herds    | 4,100                            | 479                       | 11.7  |
| Total         | 18,900                           | 2,152                     | 11.4  |

\* Based on data from the Register of production subsidies as of July 31 2006.

Table 2. Antibodies against BLV in the Norwegian bovinepopulation during the period 1995-2006

|      | Dairy herds                             | Beef                            | herds                             |                         |
|------|---|---------------------------------|-----------------------------------|-------------------------|
| Year | No. of bulk<br>milk samples<br>analysed | No. of<br>beef herds<br>sampled | No. of<br>individuals<br>analysed | No. of positive samples |
| 1995 | 25,131                                  | 1,532                           | 9,354                             | 8 (bulk milk)           |
| 1996 | 25,278                                  | 303                             | 1,523                             | 1 (bulk milk)           |
| 1997 | 26,903                                  | 2,214                           | 16,741                            | 0                       |
| 1998 | 23,581                                  | 2,191                           | 17,095                            | 0                       |
| 1999 | 19,933                                  | 2,382                           | 18,274                            | 0                       |
| 2000 | 1,590                                   | 340                             | 2,892                             | 0                       |
| 2001 | 2,564                                   | 434                             | 3,453                             | 0                       |
| 2002 | 2,308                                   | 462                             | 3,693                             | 1 (bulk milk)           |
| 2003 | 1,845                                   | 449                             | 3,901                             | 0                       |
| 2004 | 1,573                                   | 402                             | 3,364                             | 0                       |
| 2005 | 1,919                                   | 484                             | 4,766                             | 0                       |
| 2006 | 1,673                                   | 479                             | 4,624                             | 0                       |

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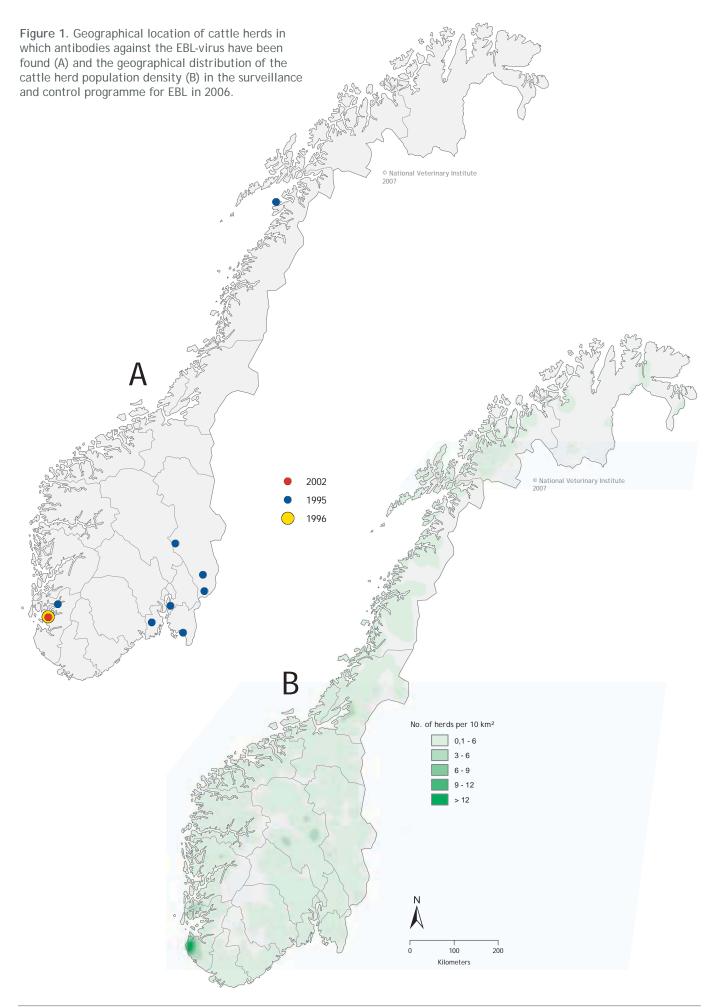
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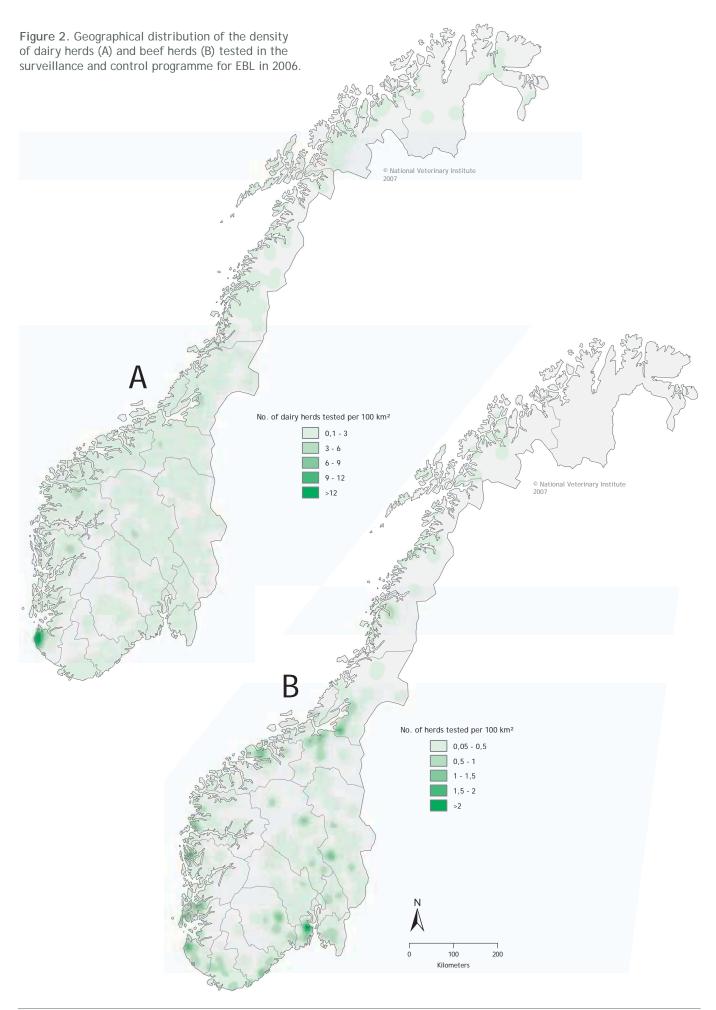
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Annual report 2006



The surveillance and control programme for *Brucella abortus* in cattle in Norway

Annette Hegermann Kampen Berit Djønne Bjørn Bratberg Jorun Tharaldsen Petter Hopp

Brucella abortus *in cattle was not detected in Norway in 2006.* 

Eradication of bovine brucellosis in Norway was achieved in 1950 (1, 2).

Since 1994, the EFTA Surveillance Authority (ESA) has recognised Norway as a state officially free from brucellosis as described in ESA Decision 66/94/COL, later replaced by ESA Decision 227/96/COL.

A surveillance and control programme for *Brucella abortus* was launched in 2000. All samples were negative in 2000, 2001, 2003 and 2004 (2, 3, 4). In 2002 however, two bulk milk samples were antibody positive. Further investigation did not confirm these positive results and it was concluded that the positive serological results most likely were false positive reactions. (5).

Since 2005 the programme has consisted of passive clinical surveillance.

The Norwegian Food Safety Authority is responsible for carrying out the programme. The National Veterinary Institute is in charge of planning the programme, performing the analyses and reporting the results. The samples are collected by inspectors of the Norwegian Food Safety Authority.

# Aim

The aim of the programme is to document freedom from *Brucella abortus* in cattle according to demands in Directive 64/432/EEC with amendments and to contribute to the maintenance of the present favourable situation.

# Material and methods

Herd criteria for submission of clinical material are:

- abortions occurring between the fifth month of pregnancy and 14 days before expected birth
- at least two abortions within this pregnancy period the last twelve months

Material for submission:

- foetus and the foetal membranes
- blood sample from the cow at the time of abortion and a second blood sample collected 14-21 days later

#### Post-mortem investigations

Foetuses are subjected to a full autopsy. Specimens from lungs, myocardium, liver, kidneys, (whole) brain, and foetal membranes are fixed in 10 % neutral phosphate-buffered formalin. The specimens are processed according to a standard routine protocol, sectioned at 5  $\mu$ m and stained with haematoxylin and eosin.

#### **Bacteriological investigations**

Foetal membranes and organs from the aborted foetus (liver, spleen and stomach contents) are sampled. Direct smears from these materials are examined following Gram and Modified Ziehl-Neelsen staining. Samples are cultured on selective Brucella agar containing 5 % horse serum, Amphotericin B, Bacitracin, Polymyxin B and Vancomycin at 37 °C in a 10 %  $CO_2$  atmosphere. The media are examined regularly and incubated for up to 14 days. Suspicious bacterial colonies are tested for motility, nitrate reduction, and for the production of catalase, indol, cytochrome oxidase, and urease. Non-motile, nitrate-reducing, indol-negative, and catalase-, cytochrome oxidase- and urease-producing isolates are sent to a reference laboratory for further identification.

|      |          | Dairy cattle |       | Beef cattle |       | Total   |       |
|------|----------|--------------|-------|-------------|-------|---------|-------|
| Year | Material | Animals      | Herds | Animals     | Herds | Animals | Herds |
| 2000 | Foetuses |              |       |             |       | 17      | 14    |
| 2001 | Foetuses | 21           | 18    | 0           | 0     | 21      | 18    |
| 2002 | Foetuses | 18           | 17    | 10          | 6     | 28      | 23    |
| 2003 | Foetuses | 30           | 25    | 4           | 3     | 34      | 28    |
| 2004 | Foetuses | 25           | 21    | 2           | 2     | 27      | 23    |
|      | Cows     | 28           | 19    | 2           | 2     | 30      | 21    |
| 2005 | Foetuses | 16           | 14    | 8           | 7     | 24      | 21    |
|      | Cows     | 48           | 26    | 8           | 4     | 56      | 30    |
| 2006 | Foetuses | 11           | 11    | 0           | 0     | 11      | 11    |
|      | Cows     | 19           | 13    | 1           | 1     | 20      | 14    |

Table 1. Number of foetuses and cows examined for brucellosis in the Norwegian cattle population during the years 2000-2006

#### Serology

Individual, paired blood samples are tested for antibodies against Brucella abortus in an indirect ELISA (Svanova®). The initial screening is performed using a single well per sample, and doubtful or positive reactions are retested in duplicates. If the result is negative when retested, the sample is concluded to be negative for antibodies against Brucella abortus. If the result still is doubtful or positive, the sample is tested with a competitive ELISA (C-ELISA, Svanova®). Positive samples in this test are subjected to a complement fixation test (CF). If the CF test is also positive, the result is reported with recommendation of a new blood sample from the suspected animal four to six weeks after the initial sampling. If this is positive, or if there should be a need for immediate follow-up, the animal is tested with an intracutane test using Brucellergene OCB from Brucella melitensis (Synbiotics®).

# **Results and discussion**

A total of 11 foetuses from 11 different herds and 36 blood samples from 20 cows (paired samples from 16 cows and 4 single samples) were analysed in 2006 (Table 1).

Post-mortem investigations of foetuses in 2006 did not reveal pathological changes indicative of brucellosis, and all bacteriological and antibody investigations were negative for *Brucella abortus*.

In conclusion, there was no detection of *Brucella abortus* in cattle in Norway in 2006. With the exception of a single relapse in 1953, bovine brucellosis has not been detected in Norway since 1950 (1, 2, 3, 4, 5).

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Annual report 2006



The surveillance and control programme for bovine virus diarrhoea (BVD) in Norway

Annette Hegermann Kampen Johan Åkerstedt Sveinn Gudmundsson Petter Hopp Gry Grøneng Ola Nyberg

The aim of the programme has been achieved as no new herds where put under restrictions due to Bovine virus diarrhoea in 2006.

Bovine virus diarrhoea (BVD) is caused by bovine virus diarrhoea virus (BVDV) in the genus pestivirus. The virus is the cause of mucosal disease and hemorrhagic syndrome, but the economically most important manifestations of disease are related to infection in pregnant animals, resulting in embryonic death, abortion and congenital defects. Persistently infected calves may be born and serve as the main reservoir of infection to other animals (1). Bovine virus diarrhoea is a notifiable disease in Norway. From 1984 to 1986, preliminary investigations indicated that nearly 30 % of the dairy herds had animals with antibodies to BVDV (2). The high prevalence and cost of the disease made control and eradication of the infection in cattle important.

A surveillance and control programme was started in December 1992 (3), financed by the authorities and the industry.

#### Aim

The purpose of the programme until 2006 has been to eradicate BVD from the cattle population in Norway.

# Material and methods

The sample frame in 2006 included all dairy herds in Norway and twenty percent of the Norwegian beef herds.

#### Testing scheme

During the years 1993 to 2006, diagnostic tests were performed on four categories of sampled material (Table 1). Testing for antibodies in bulk milk and pooled milk samples from primiparous cows was performed once a year as a minimum. In some years, intensive testing regimes were applied in certain geographical areas (special zones), while in the rest of the country only a selection of herds were tested. Herds with moderate or high levels of antibodies against BVDV in bulk milk or pooled milk samples were further tested by pooled blood samples from young stock.

Identification of persistently infected animals was done by i) testing blood samples from every individual in the herd for antibodies, and ii) testing for the presence of virus in antibody negative individuals and in animals with weak positive serological results.

#### Laboratory techniques

For detection of antibodies against BVDV in milk and in blood samples, an indirect enzyme-linked immunosorbent assay (ELISA; Svanova Biotech AB, Uppsala, Sweden) was used (4).

An antigen-capture ELISA (Moredun Animal Health, Edinburgh, Scotland, UK) was used for the detection of BVDV in blood samples up to 2003 (5, 6). After the need of virus tests declined to less than 1,000 a year, another antigencapture ELISA (IDEXX Laboratories, Inc., Westbrook, Maine, USA), has been in use. From 2005, positive reactions in new infected herds were verified with the polymerase chain reaction (PCR) and sequence analysis.

Depending on the level of antibodies in bulk milk, dairy herds were divided in four groups (Table 2). The results are expressed as sample to positive ratio (S/P-ratio) (7).

#### Herd restrictions

Detection of antibodies in a pooled serum sample from young animals (seven to twelve months) indicated that BVDV had been present in the herd in the course of the last year before testing. This implied a great risk that one or more animals could be persistently infected, and restrictions were imposed on the herd.

Restrictions were lifted when all persistently infected animals had been identified and culled and new blood samples from young stock were antibody negative in two consecutive testings.

#### Table 1. BVD testing scheme

| Category                            | Specimen                             | Selection  |  |  |  |
|-------------------------------------|--------------------------------------|--|--|--|--|
| 1                                   | Bulk milk                            | All, or randomly selected dairy herds  |  |  |  |
| 2                                   | Pooled milk from primiparous cows    | Dairy herds with elevated bulk milk antibody levels <sup>1</sup>   |  |  |  |
| 3                                   | Pooled blood sample from young stock | Dairy herds with elevated antibody levels in bulk milk or pooled milk from primiparous cows<br>All, or randomly selected beef cattle herds |  |  |  |
| 4                                   | Individual blood samples             | All animals in herds with seropositive pooled blood samples<br>Animals clinically suspected for BVD  |  |  |  |
| <sup>1</sup> Not sampled after 2001 |                                      |  |  |  |  |

Surveillance and control programmes in Norway · BVD · Annual report 2006

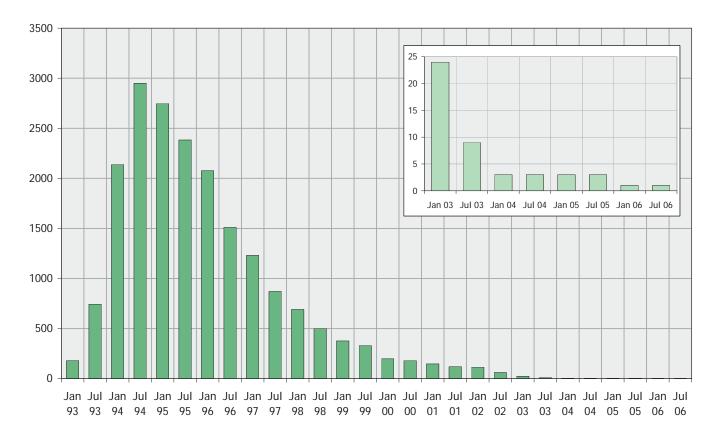


Figure 1. Number of herds with imposed restrictions because of BVDV infection during the period 1993-2006.

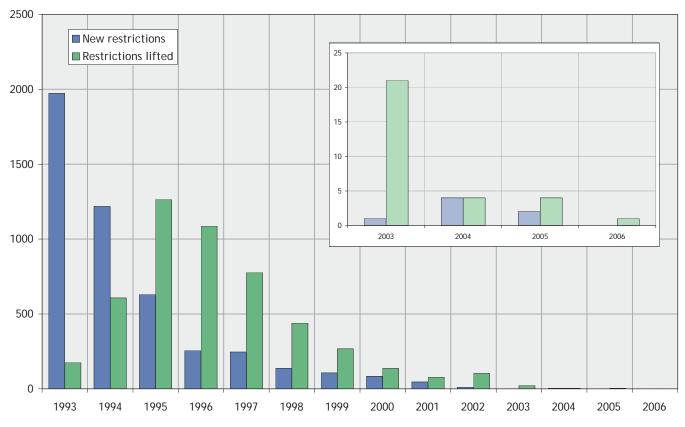


Figure 2. Number of new herds with restrictions imposed/restrictions lifted per year because of BVDV infection during the period 1993-2006.

Table 2. Classification of dairy herds after testing of bulk milk samples for antibodies against BVDV according to the sample to positive ratio (S/P ratio)

| Classification | S/P ratio      | Interpretation               |
|----------------|----------------|------------------------------|
| 0              | < 0.050        | Antibodies not detected      |
| 1              | 0.050 - 0.149  | Low level of antibodies      |
| 2              | 0.150* - 0.549 | Moderate level of antibodies |
| 3              | ≥ 0.550        | High level of antibodies     |
|                |                |                              |

\* Border value changed from 0.250 to be able to discover new infected herds at an early stage. Effective from 1998.

Table 4. Results of Norwegian cattle herds tested forantibodies against BVDV in pooled milk from primiparouscows (not sampled after 2001)

| Year | No. of<br>herds examined | % antibody positive<br>herds |  |  |
|------|--------------------------|------------------------------|--|--|
| 1993 | 5,031                    | 70.7                         |  |  |
| 1994 | 3,228                    | 54.5                         |  |  |
| 1995 | 3,191                    | 44.3                         |  |  |
| 1996 | 1,849                    | 44.1                         |  |  |
| 1997 |                          |                              |  |  |
| 1998 | 1,415                    | 21.5                         |  |  |
| 1999 | 924                      | 24.2                         |  |  |
| 2000 | 100                      | 13.0                         |  |  |
| 2001 | 53                       | 9.4                          |  |  |

Table 3. Classification of Norwegian dairy herds according to BVDV antibody test values of bulk milk

|      |                    | Classification (% of sampled herds) <sup>1</sup> |      |                  |       |  |
|------|--------------------|--|------|------------------|-------|--|
| Year | No. of dairy herds | 0  | 1    | 2                | 3     |  |
| 1993 | 26,424             | 63.0   | 14.1 | 15.9             | 7.1   |  |
| 1994 | 26,148             | 63.4   | 12.2 | 14.5             | 9.9   |  |
| 1995 | 25,577             | 63.7   | 10.6 | 12.5             | 13.2  |  |
| 1996 | 25,167             | 70.5   | 15.4 | 10.7             | 3.5   |  |
| 1997 | 24,862             | 74.3   | 15.7 | 8.7              | 1.2   |  |
| 1998 | 24,038             | 81.3   | 9.1  | 9.2 <sup>2</sup> | 0.4   |  |
| 1999 | 23,584             | 85.6   | 8.8  | 5.6              | < 0.1 |  |
| 2000 | 21,796             | 88.3   | 6.3  | 5.3              | 0.1   |  |
| 2001 | 19,910             | 91.9   | 4.7  | 3.2              | 0.2   |  |
| 2002 | 18,771             | 94.4   | 3.1  | 2.2              | 0.3   |  |
| 2003 | 17,549             | 96.7   | 2.1  | 1.1              | 0.02  |  |
| 2004 | 7,365 <sup>3</sup> | 95.8   | 2.8  | 1.3              | 0.1   |  |
| 2005 | 7,481 <sup>3</sup> | 98.0   | 1.2  | 0.8              | 0.03  |  |
| 2006 | 14,620             | 96.8   | 2.7  | 0.5              | 0.01  |  |

<sup>1</sup> Based on S/P ratios, see Table 2. <sup>2</sup> Cut-off value changed from 1998, see Table 2. <sup>3</sup> Selected herds.

# Results

#### Bulk milk samples

Bulk milk samples from a total of 14,620 dairy herds were tested for antibodies against BVDV in 2006. In 2006, 3.2 % of the herds were classified in group 1 or above (Table 3).

#### Pooled milk samples

For completeness of historical data, results of pooled milk testing from primiparous cows are also included (Table 4).

#### Pooled blood samples

Blood samples for serological testing of pooled samples were submitted from 998 different dairy herds (10 %) and beef cattle herds (90 %). One of these samples was sero-positive (Table 5). Samples from one herd were excluded from examination.

#### Individual blood samples

A total of 113 animals from 28 herds were investigated by pooled blood samples from young stock in 2006. BVDV was not detected in any of the animals (Table 6).

#### Herds with restrictions

Restrictions were not imposed on any new herds because of suspected BVD in 2006 (Figure 1). One herd had restrictions at the beginning of the year, but these were lifted in November 2006 (Figure 2). Table 5. Serological results for BVDV in pooled blood samplesfrom young stock of cattle in Norway

| Year | Tested samples<br>(No.) | Positive samples<br>(%) |  |  |
|------|-------------------------|-------------------------|--|--|
| 1993 | 5,000                   | 46.5                    |  |  |
| 1994 | 4,107                   | 38.2                    |  |  |
| 1995 | 5,347                   | 23.5                    |  |  |
| 1996 | 3,163                   | 21.9                    |  |  |
| 1997 | 3,292                   | 16.0                    |  |  |
| 1998 | 3,407                   | 10.8                    |  |  |
| 1999 | 3,060                   | 8.6                     |  |  |
| 2000 | 1,610                   | 8.6                     |  |  |
| 2001 | 4,198                   | 2.5                     |  |  |
| 2002 | 2,854                   | 1.8                     |  |  |
| 2003 | 2,100                   | 1.0                     |  |  |
| 2004 | 1,351                   | 1.4                     |  |  |
| 2005 | 1,230                   | 0.3                     |  |  |
| 2006 | 997                     | 0.1                     |  |  |

# Discussion

No herds had restrictions because of BVD at the end of 2006. Testing of bulk milk from all dairy herds and a 20 % representative sample of all beef cattle herds during 2006 with no findings of new infected herds, indicates that the ultimate goal of eradicating BVD in Norway may now be considered achieved.

During the programme period, the number of herds with restrictions decreased from 2,950 in 1994 to none at the end of 2006 (Figure 1).

A cost-benefit analysis has estimated that the Norwegian BVD control programme saves the Norwegian dairy industry between 50 and 200 million Norwegian kroner every year (8).

Several factors have been important for the success of the Norwegian BVD eradication programme:

Since the beginning in 1992, the surveillance and control programme for BVD has been a joint effort of farmers, industry, authorities and diagnostic laboratories (9, 10). All parties involved have been focusing on the same task in a long-term perspective. Also, stable funding and the will to continue the programme after its original end-point in 1997 have been important to achieve the goal.

Another important point for making the eradication programme effective, was that BVD was classified as a notifiable list B disease in Norway. This enabled the authorities to impose restrictions on sale and movement of animals from herds with suspected or diagnosed persistently infected animals. This prevented spread and also served as an incentive for farmers to get rid of such animals.

The project has highlighted the importance of continuous information and education of farmers and practitioners as important factors for motivation.

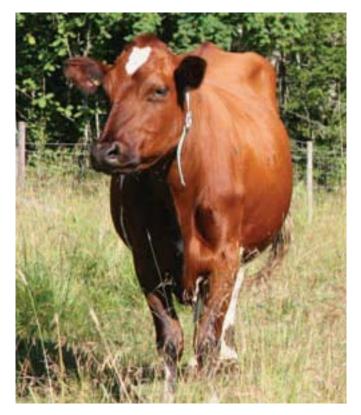
The surveillance and control programme for BVD in Norway was designed without the use of vaccines, allowing cheap and effective serological surveillance in the initial screening of the herds.

The progress in reducing the number of infected herds was excellent during the first years, but less so in the later period of the programme (10). The main reason for the reduced reduction rate of herds with restrictions was that the numbers of new infected herds each year were relatively high (Figure 2).

Table 6. Test results for BVD in individual blood samples from Norwegian cattle during the period 1998-2006

|                | No. examined for antibodies |                     | No. of antibody positive | No. examined for virus |                     | Virus positive herds |      | Virus positive samples |     |
|----------------|-----------------------------|---------------------|--------------------------|------------------------|---------------------|----------------------|------|------------------------|-----|
| Year           | Herds                       | Samples             | Samples                  | Herds                  | Samples             | No.                  | %    | No.                    | %   |
| 1993 +<br>1994 |                             | 46,000 <sup>1</sup> |                          |                        | 21,500 <sup>1</sup> |                      |      | 1,300 <sup>1</sup>     | 6.0 |
| 1995           |                             | 36,065              |                          |                        | 18,302              |                      |      | 1,180                  | 6.5 |
| 1996           |                             | 21,437              |                          |                        | 11,665              |                      |      | 685                    | 5.9 |
| 1997           | 1,515                       | 16,023              | 7,898                    | 1,348                  | 8,125               | 265                  | 19.7 | 525                    | 6.5 |
| 1998           | 780                         | 7,091               | 3,668                    | 694                    | 4,119               | 98                   | 12.6 | 198                    | 2.8 |
| 1999           | 648                         | 7,619               | 3,915                    | 581                    | 4,292               | 92                   | 14.2 | 224                    | 2.9 |
| 2000           | 423                         | 6,947               | 3,524                    | 370                    | 3,553               | 72                   | 17.0 | 129                    | 1.9 |
| 2001           | 386                         | 6,287               | 2,946                    | 314                    | 2,895               | 56                   | 14.5 | 174                    | 2.8 |
| 2002           | 284                         | 3,962               | 1,334                    | 232                    | 2,390               | 28                   | 9.9  | 43                     | 1.1 |
| 2003           | 149                         | 1,135               | 295                      | 110                    | 677                 | 9                    | 6.0  | 22                     | 1.9 |
| 2004           | 84                          | 1,017               | 311                      | 82                     | 635                 | 2                    | 2.4  | 6                      | 0.6 |
| 2005           | 48                          | 356                 | 84                       | 47                     | 287                 | 1                    | 2.1  | 4                      | 1.1 |
| 2006           | 28                          | 113                 | 1                        | 28                     | 112                 | 0                    | 0    | 0                      | 0   |

<sup>1</sup> Approximate numbers.



To be able to find new infected herds at an early stage, the classification of dairy herds was changed in 1998 when the lower border value for herds classified in group 2 was reduced (Table 2). In the following years, a total of 1,781 additional herds were classified in this group. Pooled blood samples from 1,286 of these herds have been tested, and of them, 269 were seropositive. Persistently infected animals were found in 132 (7.4 %) of the additional herds classified in group 2. Earlier identification of these infected herds may have speeded up the eradication rate.

The majority of new infected herds were located in the same areas as remaining herds with restrictions. In the years 2001 to 2003, geographical areas with many infected herds were selected for enforced eradication of BVD (special zones). Specific testing schemes prevented that persistently infected animals could be sold or were allowed access to common pastures. Intensive surveillance of infected herds included direct information to farmers, veterinarians and other advisors on introduction, clinical signs and eradication of BVD, as well as how to avoid reinfection (11). Figures 1 and 2 indicate that these measures were effective in order to reduce the number of infected herds.

Although Norway is currently free from BVD, there are still challenges for the future. Import of animals and unknown wildlife reservoirs may pose a threat to the present status. This reminds us that farmers, practitioners and authorities should be alert, and efficient surveillance should be continued to detect a possible reintroduction of BVD in Norway and control its spread.

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The surveillance and control programme for bovine tuberculosis in Norway

Annette Hegermann Kampen

## *In 2006,* Mycobacterium *sp. was not detected in samples submitted from slaughterhouses.*

Apart from two single-herd outbreaks in Sogn og Fjordane county in 1984 and 1986 Norway has been considered free from bovine tuberculosis since 1963 (1, 2, 3, 4). And since 1994, the EFTA Surveillance Authority (ESA) has recognised Norway as officially free from bovine tuberculosis, as described in ESA Decision 225/96/COL replacing ESA Decision 67/94/COL. In 2000, a surveillance and control programme for bovine tuberculosis was launched. The programme includes compulsory veterinary inspection of all bovine carcasses at slaughter, with submission of suspicious materials to the National Veterinary Institute for further examination.

#### Aims

The aims of the programme are to document absence of bovine tuberculosis, according to Directive 64/432/EEC with amendments, and to contribute to the maintenance of this favourable situation.

## Material and methods

Submission of material from slaughterhouses Lung tissue, lymph nodes and other organs with pathological lesions where bovine tuberculosis can not be excluded, are submitted for examination.

The Food Safety Authority collects the samples during routine meat inspection.

#### Histopathological examination

Tissues are fixed in 10% neutral phosphate-buffered formalin for more than 24 hours, processed according to a standard routine protocol, embedded in paraffin, sectioned at 5  $\mu$ m and stained with haematoxylin and eosin and Ziehl-Neelsen (5).

#### Bacteriological examination

Samples are examined as described in the OIE manual (5). Samples are homogenised, decontaminated with 5 % oxalic acid and centrifuged. The top layer of the sediment is used for culturing and microscopic examination. The sediment is inoculated onto slopes of Petragnani medium, Stonebrink's medium and Middelbrook 7H10 medium. The slopes are incubated aerobically at 37 °C for two months and checked every week for growth of acid-fast bacilli, determined by the Ziehl-Neelsen method.

#### **Results and discussion**

Table 1 shows the number of samples collected and the results since the programme started in 2000. In 2006, three samples were submitted. All were negative for Mycobacterium sp.

The low number of submitted samples indicates a low prevalence of suspicious pathological lesions. Continuous surveillance by veterinary meat inspection, early and effective eradication campaigns, combined with restricted import of live cattle, have contributed significantly to this situation.

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Table 1. Number of samples tested for bovine tuberculosisduring the period 2000-2006

|      | No. of  | No. of | No. of p | oositive |
|------|---------|--------|----------|----------|
| Year | samples | herds  | Samples  | Herds    |
| 2000 | 0       | 0      | 0        | 0        |
| 2001 | 3       | 3      | 0        | 0        |
| 2002 | 0       | 0      | 0        | 0        |
| 2003 | 1       | 1      | 0        | 0        |
| 2004 | 4       | 4      | 0        | 0        |
| 2005 | 1       | 1      | 0        | 0        |
| 2006 | 3       | 3      | 0        | 0        |

# Survey of specific serogroups of *E. coli* in sheep in Norway



Anne Margrete Urdahl Petter Hopp

2006 sees the start of a surveillance of strains of Escherichia coli in sheep that are pathogenic to humans to investigate possible geographical variation and risk factors.

*Escherichia coli* are bacteria normally present in the intestinal flora of both humans and animals. Some *E. coli* may be pathogenic for humans. Of these, only Shiga toxin-producing *E. coli* (STEC), also known as Verotoxin-¬producing *E. coli* (VTEC), has a defined zoonotic origin with domestic ruminants regarded as the major reservoir.

The Shiga toxins are encoded by the genes  $stx^1$  and  $stx^2$ . The toxins are the major virulence factors of STEC and the cause of haemorrhagic uremic syndrome (HUS) seen in humans.

The attachment in the human gastrointestinal tract by *E. coli* through a complicated mechanism encoded among others by *eae* is the actual cause of (hemorrhagic) diarrhoea seen in these patients. This virulence characteristic is also seen among other *E. coli*.

The most well known human pathogenic serotypes of STEC are 026:H11, 0111:H8, 0103:H2, 0145:H21 and 0157:H7. However, other serotypes may also cause human infections as observed in the Norwegian outbreak in 2006 with 17 human cases caused by STEC 0103:H25. The source of the infection was dry-cured sausages with the bacteria originating from contaminated sheep meat.

Until 2006, the annual reported incidence of human STEC infections in Norway has been low (0-17 cases per year) with approximately half of the cases domestically acquired (1).

#### The animal reservoir

There is limited knowledge of the prevalence of STEC in the Norwegian ruminant populations. Studies performed in Norway from 1995 to 1999 reported cattle herd prevalences of STEC 0157 of 0.5 % to 1 % (2, 3). Only one study has focused on detecting herd prevalence of STEC 0157 in sheep. The study did not detect any STEC 0157 (2).

In a surveillance programme for STEC 0157 in cattle, sheep, and goat carcasses running in the period 1998-2004, the total carcass prevalence was 0.06 % for cattle and 0.03 % for sheep. None of the 510 goat carcasses tested were positive (4).

There are less data on the other serogroups. Two studies in cattle have focused on detecting serogroups O26, O103, O111, and O145. The detection of *eae*-negative STEC O103 was reported from 3.2 % of the herds in one of the studies. In both studies *stx*-negative *E. coli* of the serogroups O26, O103, O145 and O111 were detected (4). In a study of one sheep flock conducted in 2000, 2 lambs (1.6 %) were positive for STEC O103 (5). The isolates were not H-typed, but carried *stx*<sup>1</sup> and *eae*. In addition, *stx*-negative isolates were detected from 62 of the total 96 samples tested. International studies also report *stx*-negative and *eae*-positive *E. coli*, and *stx*- and *eae*-negative *E. coli* isolates of these serogroups (O26, O103, O111, O145 and O157), indicating that these are relatively common in the microbial flora of animals. During the 2006 outbreak, *stx*-negative and *eae*-positive *E. coli* O103:H25 was detected from several sources of sheep origin with no clear epidemiological link, indicating that this serotype is common among sheep in Norway. However, the relationship and ratio between true *stx*- and *eae*-negative *E. coli*, true *stx*-negative *and eae*-negative *E. coli*, stx-negative and *eae*-negative *E. coli*, star-negative *and eae*-negative *E. coli*, true *stx*-negative *and eae*-negative *E. coli*, star-negative *and eae*-negative *E. coli* (STEC), and *stx*- and *eae*-positive *E. coli* (STEC) of a serotype, is unknown and more data is needed to assess this problem.

The outbreak emphasised the need for more knowledge regarding Shiga toxin-producing *E. coli* in the sheep population. The Norwegian Food Safety Authority therefore decided to initiate a national surveillance programme. The National Veterinary Institute was asked to design the programme, perform the analyses, and the reporting the results. The samples would be collected by inspectors from the Norwegian Food Safety Authority.

## Aims

The aims of the survey are to gather knowledge on the occurrence of some specific serogroups of *E. coli* and their virulence factors in sheep, and to investigate possible geographical variation and risk factors.

## Material and methods

In November 2006 faecal samples were planned and collected from 100 randomly selected sheep flocks (farms). Only sheep flocks with at least 50 sheep  $\geq$ 1 year old were eligible. From each of these flocks, 50 single faecal samples should be taken from the youngest animals (lamb first, then one-year olds etc.). Another 500 flocks are planned for sampling during the autumn of 2007.

Autumn was chosen as sampling period to give representative data from the time of year when most sheep is slaughtered and thereby indications of possible contamination risks to sheep products. Lambs are chosen as young ruminants shed more of these bacteria and are also proportionally slaughtered most.

From each farm, a questionnaire addressing potential risk factors for the occurrence of STEC is to be filled in.

From each farm, pools of 10 individual samples will be analyzed for the various *E. coli* serogroups. A modified method of NMKL 164 where the IMS method has been further modified by inclusion of an ELISA step is used for detection of *E. coli* 0157 and 0103. IMS-ELISA for other serogroups is currently under development. ELISA positive samples will be plated onto selective agar for colony isolation. Thereafter, *E. coli* isolates will be 0:H serotyped and further characterized for virulence factors by PCR.

## **Results and discussion**

In 2006, 4,621 samples were collected from 94 sheep flocks. All the faecal samples were frozen at -80 °C on arrival at the laboratories. The samples will be analyzed in 2007.

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# The surveillance and control programme for maedi in Norway



Annette Hegermann Kampen Jorun Tharaldsen Snorre Stuen Gry Grøneng Ola Nyberg

## *In 2006, none of the investigated flocks were diagnosed with maedi.*

Maedi is a progressive viral pneumonia in sheep first described in Iceland in 1939 (1). The disease occurs in several European countries as well as in other continents. The disease visna is caused by the same virus as maedi, but is a neuropathogenic manifestation of the infection (1, 2). Maedi-visna is classified as a list B disease in Norway and is notifiable to the Office International des Epizooties.

In Norway, maedi was officially reported for the first time in 1972 (3) and a nationwide disease control programme was launched in 1975.

In November 2002 and January 2003, post mortem examinations of lungs from two diseased sheep from two different farms in Nord-Trøndelag county showed histopathological changes consistent with maedi. The diagnoses were confirmed by serological tests of blood samples. During the following investigations more than 15,000 sheep in 300 flocks were serologically examined for maedi-visna infection, and 50 flocks were found to be seropositive (4, 5). The outbreak demonstrated that maedi-visna infection was more widespread in Norway than previously anticipated, and this necessitated a new nationwide surveillance and control programme, which was started in November 2003 (4, 6).

An overview of the number of new infected flocks registered each year up to 2006 is given in Figure 1.

## Aim

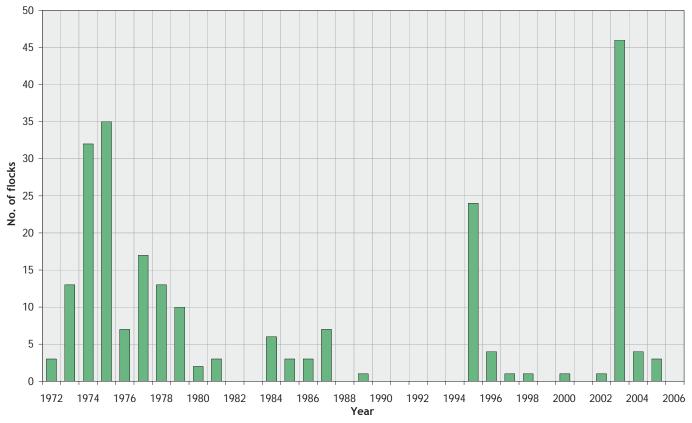
The aims of the surveillance and control programme for maedi are to document the status for maedi-visna virus infection in sheep in Norway, and to identify infected flocks for disease control.

## Materials and methods

Ram circles and their member flocks registered by The Norwegian Sheep and Goat Breeders Association constitute the target population for the programme. Approximately 2,200 flocks were part of this breeding system in 2006, of a total of 15,800 sheep flocks. Of these 661 flocks were selected for testing. In addition, sheep from 300 randomly selected flocks not belonging to any ram circle were included.

Thirty animals per flock were sampled in flocks with less than 100 sheep, 35 animals were sampled in flocks with 100 to 200 sheep, and 40 animals per flock were tested in flocks with more than 200 animals. All rams and the oldest sheep among those more than one-and-a-half years old, were sampled in each flock.

The programme in 2006 was based on serological examination of blood samples from the selected sheep for antibodies against maedi-visna virus with the ELISA from Pourquier (ELISA CAEV/MAEDI-VISNA serum verifi-cation kit, Institut Pourquier, Montpellier, France). Sero-positive ELISA-results were retested in duplicate with the same ELISA and veri-



**Figure 1**. The number of new flocks infected with maedi registered during the period 1972 to 2006. The bars for 2003 – 2006 show both seropositive flocks detected through the investigations after the outbreak in Nord-Trøndelag county and seropositive flocks discovered in the programme.

fied by an agar gel immunodiffusion test (AGIDT, Meditect, Veterinary Laboratories Agency, Weybridge, UK). In the case of inconclusive results (including single reactors), new blood samples from the animals were taken one to two months after the first sampling. These samples were tested in duplicate in both tests (7).

Due to the known cross-reactions in the serological tests between maedi-visna virus and caprine arthritis encephalitis virus (CAEV) infection, blood samples from seropositive flocks with both sheep and goats are tested with a PCR-method developed at the National Veterinary Institute. The PCR-method is designed to amplify sequences from both CAEV and maedi-visna virus, followed by sequencing to differentiate the two virus types.

The meat inspectors at the abattoirs still play an important role in the programme by monitoring sheep and especially sheep lungs for detection of suspicious cases consistent with maedi-visna virus infection.

#### **Results**

Samples from a total of 911 flocks were analysed in 2006, this is approximately 6 % of the total Norwegian sheep flocks. Of these flocks, 598 are members of ram circles, corresponding to approximately 27 % of the total number of flocks in ram circles (Table 1). The geographical distribution of the Norwegian sheep population and the density tested flocks are shown in Figure 2.

In 2006, none of the investigated flocks were diagnosed with maedi. Six sheep from a flock with close contact with goats were positive in the serological tests, while seven gave inconclusive results. Sheep from this flock had been confirmed to be infected with CAEV in 2004.

#### Discussion

The programme, which started in 2003, was designed to increase the sensitivity of detecting infected flocks without increasing the costs per flock. This was done by increasing the number of sampled animals per flock and applying a more sensitive, but less labour-intensive test.



The sample size per flock was adjusted so that if none of the tested animals were seropostive, the prevalence of maedi-visna infected animals in a flock would be less than 6 %, given a confidence level of 95 % and 100 % test sensitivity.

The ELISA employed in this programme is considered to be more sensitive than the traditionally used agar gel immunodiffusion test. The ELISA is also more objective and less dependent of the operator's skill than the AGIDT. To gain experience with the different tests, and to ascertain the sensitivity and the specificity for the ELISA from Pourquier, another ELISA and the AGIDT were previously used when the first test was positive. The disadvantage with this test regimen was that in some cases the results were difficult to interpret, which lead to more inconclusive results and required testing of new blood samples. From 2006, the second ELISA test was omitted, as a study showed that this would increase the overall sensitivity of the test regimen without lowering the specificity (7).

Results from the surveillance and control programme for maedi, including data from November 2003 through 2005, show a preliminary prevalence of less than 0.2 % positive flocks (4, 8). Knowledge about the distribution of the disease so far indicates that it is regionally clustered, and that a more extensive spread of maedi-visna virus has probably been prevented by the restrictions on transfer of sheep across county borders.

| Table 1. The number of flocks | and sheep tested in the | e Norwegian surveillance an | d control programme for maedi |
|-------------------------------|-------------------------|-----------------------------|-------------------------------|
|                               |                         |                             |                               |

| Year | Total no. of<br>sheep flocks* | No. of flocks included in the programme | No. of flocks sampled | No. of animals tested | No. of positive<br>flocks |
|------|-------------------------------|---|-----------------------|-----------------------|---------------------------|
| 2003 | 18,400                        | 2,227                                   | 456**                 | 13,951                | 1                         |
| 2004 | 17,439                        | 2,600                                   | 1,230                 | 36,911                | 1                         |
| 2005 | 16,500                        | 2,519                                   | 940                   | 29,248                | 2                         |
| 2006 | 15,800                        | 2,198                                   | 911                   | 27,846                | 0                         |

\* Based on data from the register of production subsidies as of 31 July 2006, \*\* Sampling period: November 20 to December 31.

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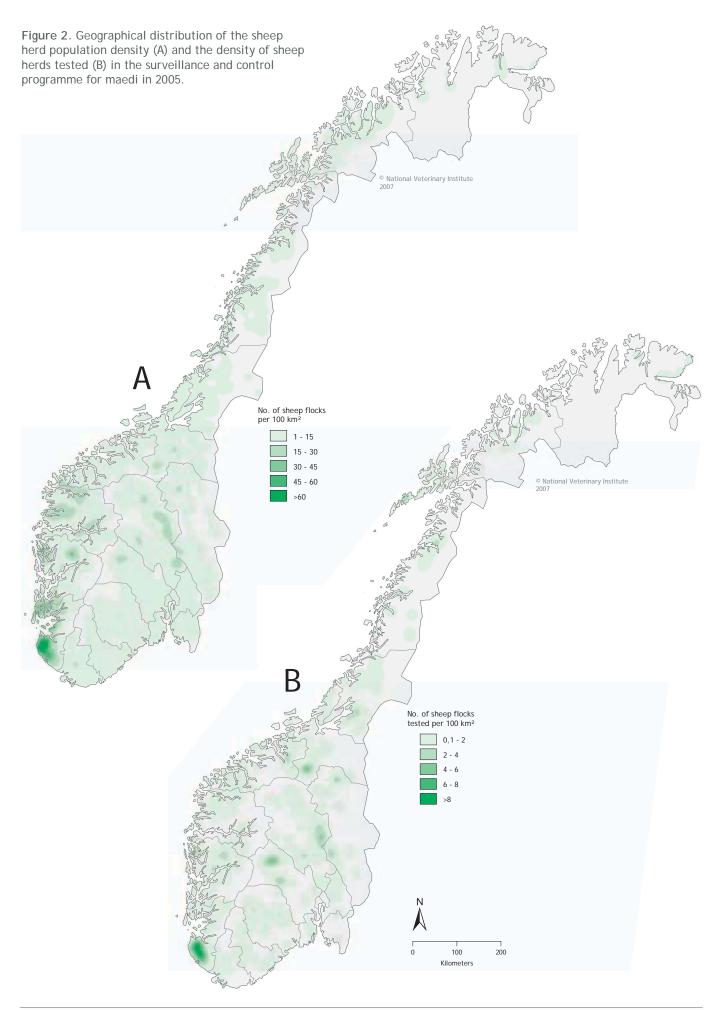
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The surveillance and control programme for *Brucella melitensis* in sheep in Norway



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Brucella melitensis was not detected in any sheep flock sampled for surveillance in 2006.

Brucellosis in sheep and goats is mainly caused by *Brucella melitensis*, although infection with *Brucella abortus* and *Brucella ovis* can also occur. The infection usually results in abortion in pregnant ewes and can cause orchitis and epididymitis in affected rams (1). *Brucella melitensis* infection is a zoonosis, and the bacterium causes a serious infection in humans known as Malta fever characterised by undulant fever, chills, sweat and debilitation.(2).

*Brucella melitensis* is prevalent in sheep and goats in several Mediterranean countries (1), but has never been diagnosed in animals in Norway or any of the other Nordic countries (3, 4). Brucellosis is classified as a list A disease in Norway and is notifiable to the Office International des Epizooties.

After the agreement on the European Economic Area in 1994, Norway achieved status as free from *Brucella melitensis* in small ruminants on a historical basis. However, documentation is required to maintain the status. Hence, a surveillance and control programme for *Brucella melitensis* in sheep was established in 2004.

The Norwegian Food Safety Authority is responsible for carrying out the programme. While the National Veterinary Institute is in charge of planning the programme, performing the analyses and reporting the results, the samples are collected by inspectors from the Norwegian Food Safety Authority. In addition, sheep from 300 randomly selected flocks not belonging to any ram circle were included in the programme.

All individuals were sampled in flocks of less than 30 animals. In flocks of 30 to 100, 100 to 200, and more than 200 sheep, samples from 30, 35, and 40 animals were analysed, respectively. The number of herds in the surveillance and control programme for *Brucella melitensis* in sheep in 2006 is given in Table 1.

Blood samples are examined for antibodies against *Brucella melitensis* using the rose bengal plate agglutination test (RBT) for the initial screening. A competitive ELISA (C-ELISA, Svanova Biotech AB, Uppsala, Sweden) was used to follow up unclear or positive reactions due to cross reactions.

#### Results

A total of 27,812 samples from 911 sheep flocks were analysed in 2006. This is approximately 6 % of the total Norwegian sheep flocks. Of these flocks, 598 flocks were members of ram circles, corresponding to approximately 27 % of the total number of flocks in ram circles in Norway.

All samples tested for antibodies against *Brucella melitensis* in 2006 were negative. The results from the surveillance and control programme for *Brucella melitensis* in sheep in 2004-06 are shown in Table 1.

The geographic distribution of the total number and the number of tested sheep flocks in 2006 are shown in Figure 1.

## Aims

The aims of the programme are to document freedom from *Brucella melitensis* in sheep according to the demands in EU Directive 91/68/EEC with amendments and to contribute to the maintenance of this favourable situation.

## Material and methods

Herds belonging to ram circles registered by the Norwegian Sheep and Goat Breeders Association and their associated flocks constituted the main test population. Approximately 2,200 flocks were part of this breeding system in 2006, of a total of 15,800 sheep flocks. Six hundred and sixty one flocks in the breeding system were selected for sampling.

## Discussion

Approximately 90 % of the Norwegian flocks in ram circles were screened for antibodies against *Brucella melitensis* during 2004 and 2005 (4). In 2006, a new round of testing started, aiming at testing all flocks in ram circles within a three-year period.

The surveillance programme was evaluated in 2006. When taking into account results accumulated from 2004 to 2006, it was estimated that there is a 99 % probability that the prevalence of sheep flocks being positive for *Brucella melitensis* is lower than 0.2 % (5).

Table 1. Results and total number of sheep flocks within the frame of the Norwegian surveillance and control programme for *Brucella melitensis* in sheep in 2004-2006

| Year | Total no. of<br>sheep flocks* | Total no. of sheep >1 year of age | No. of flocks tested | No. of animals tested | No. of positive samples |
|------|-------------------------------|-----------------------------------|----------------------|-----------------------|-------------------------|
| 2004 | 17,439                        | 918,500                           | 1,655                | 50,501                | 0                       |
| 2005 | 16,500                        | 927,400                           | 935                  | 28,406                | 1**                     |
| 2006 | 15,800                        | 894,100                           | 911                  | 27,812                | 0                       |

\* Based on data from the register of production subsidies as of July 31 2006. \*\* Probably unspecific reaction.

## References

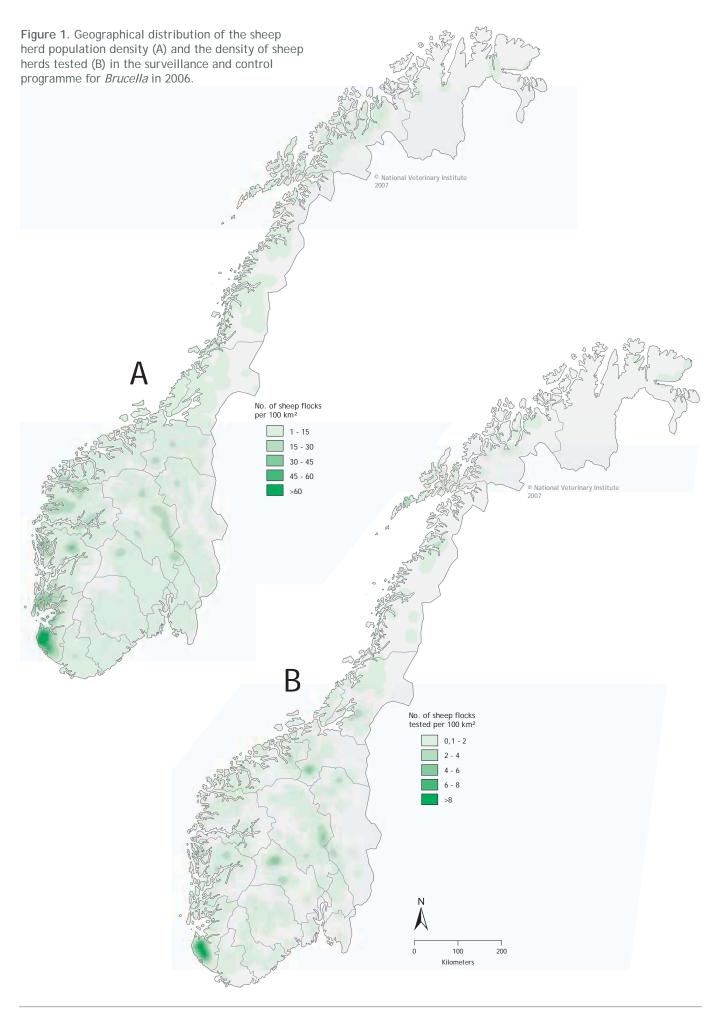
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# The surveillance and control programme for scrapie in Norway



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In 2006, classical scrapie was diagnosed in seven sheep coming from one flock. Scrapie Nor98 was diagnosed in eight sheep and one goat coming from nine different flocks.

Scrapie was first diagnosed in indigenous Norwegian sheep in 1981. Increasing numbers of scrapie-infected flocks were identified in the 1990s, culminating with 31 detected flocks in 1996 (Figure 1). By the end of 2005, scrapie had been diagnosed in a total of 110 sheep flocks. Before 2006, scrapie has never been diagnosed in goats in Norway (1). Scrapie has been a notifiable disease in Norway since 1965, and control measures have involved destruction of all sheep in affected flocks and in close contact flocks until 2004. A national scrapie surveillance and control programme was launched by the National Animal Health Authority in 1997 (from 2004: the Norwegian Food Safety Authority) (2).

In 1998 a new type of scrapie, scrapie Nor98, was detected in Norway. The diagnosis of scrapie Nor98 is verified by Western blot. Scrapie Nor98 differs from classical scrapie in several aspects, including the Western blot profile, the distribution of protease resistant prion protein (PrP<sup>sc</sup>) in the brain, and absence of detectable PrP<sup>sc</sup> in lymphoid tissue (3). The main clinical sign observed in scrapie Nor98 cases has been ataxia. The PrP genotype distribution among scrapie Nor98 cases differs markedly from that of the previous cases with classical scrapie (4).

#### Aims

The aims of the surveillance and control programme are to identify scrapie infected sheep and goat flocks to support disease control, and to estimate its prevalence in sheep and goats in fallen stock and in the sheep population slaughtered for human consumption.

#### Materials and methods

In 2006, the surveillance programme was performed according to the European Union Regulations, Regulation (EC) No. 999/2001 Annex III, with amendments and included examination of the following categories of small ruminants:

- all small ruminants with clinical signs consistent with scrapie, irrespective of age
- 10,000 sheep older than 18 months, which had died or been killed on the farm, but not slaughtered for human consumption (fallen stock)
- 10,000 randomly sampled healthy sheep older than 18 months slaughtered for human consumption
- 1,000 goats older than 18 months which had died or been killed on the farm, but not slaughtered for human consumption (fallen stock)
- 5,000 randomly sampled healthy goats older than 18 months slaughtered for human consumption

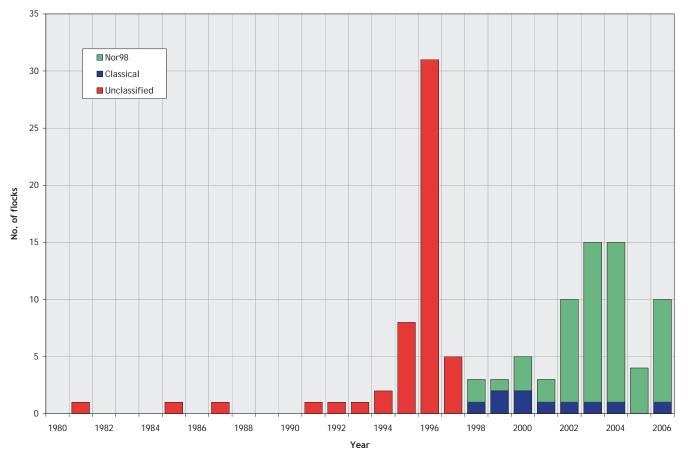


Figure 1. Annual number of sheep flocks diagnosed with classical scrapie and scrapie Nor98 during the time period 1980-2006. Before 1998 the cases were not classified according to type of scrapie, but the majority of the scrapie cases are supposed to be the classical type.

The sheep and goat farmers were responsible for reporting to the local Norwegian Food Safety Authority; when they encounter sheep and goats with clinical signs consistent with scrapie, and small ruminants older than 18 months that died or were killed on the farm due to disease. The local Norwegian Food Safety Authority evaluated the reported cases and if indicated, would follow up with either a post mortem examination at a laboratory, or a collection of a brain sample at the farm for laboratory examination. The Norwegian Food Safety Authority carried out inspections of goat herds and sheep flocks, all of which should be inspected every second or third year. The Norwegian Food Safety Authority also sampled slaughtered sheep and goats at the abattoirs, while the National Veterinary Institute was responsible for laboratory examinations and reporting of the results.

#### Animals with clinical signs consistent with scrapie

A total of 25 sheep and 3 goats with clinical signs consistent with scrapie were subject to clinical evaluation. The animals were subject to either post mortem examination at a laboratory, or formalin-fixed and unfixed brain halves and medial retropharyngeal lymph nodes were submitted for laboratory examination. All the animals were examined at the National Veterinary Institute.

#### Surveillance of fallen stock

Samples from approximately 4,600 sheep and 340 goats found dead, or which were killed on the farm, but not slaughtered for human consumption, were submitted for examination. The majority of the samples consisted of retropharyngeal lymph nodes, and unfixed *medulla oblongata* obtained through the *foramen magnum* using a metal spoon specially designed at the National Veterinary Institute. Alternatively the samples consisted of formalinfixed and unfixed brain halves and unfixed retropharyngeal lymph nodes. The samples were examined at the National Veterinary Institute in Oslo.

#### Abattoir surveillance

Approximately 10,300 randomly collected brain samples from apparently healthy sheep older than 18 months and 5,300 randomly collected brain samples from apparently healthy goats older than 18 months were collected. The sheep samples were collected at 30 abattoirs, which process all the commercially slaughtered sheep in Norway.

The samples were obtained throughout the year, with approximately 40 % of the samples collected in September and October, which is the main slaughtering season for sheep in Norway. To ensure an appropriate distribution of the samples, the Veterinary Officers at the local Norwegian Food Safety Authority were responsible for the sampling to be representative for each region and season, and the sample selection should be designed to avoid overrepresentation of any group as regards to the origin, species, age, breed, production type or to any other characteristic.

The brain samples consisted of *medulla oblongata*, and often also a small part of the *cerebellum* and midbrain, obtained through the *foramen magnum* using the specially

designed metal spoon. The samples were examined at the National Veterinary Institute in Sandnes, Trondheim and Harstad.

#### Laboratory examination procedures

Clinically suspect animals were subject to histopathological examination of brain tissue and immunohistochemical examination of brain and lymphoid tissue for PrPsc. In addition a rapid test (TeSeE sheep & goat ® Bio-Rad) was performed on brain and lymphoid tissues. From fallen stock a pooled brain tissue sample (obex and cerebellum when available) was initially examined by the rapid test. The abattoir samples (*obex*) were also initially examined by the rapid test. The TeSeE sheep & goat ® Bio-Rad test was performed according to the protocol given by the manufacturer. Immunohistochemistry and Western blot were used as confirmative tests on the samples from fallen stock and the abattoirs. Immunohistochemistry was performed using a monoclonal anti-PrP-antibody (F89/160.1.5) (5). A commercially available kit (Envision+® System HRP [AEC] DakoCytomation) was used to enhance the sensitivity of the method. The confirmative tests, immunohistochemistry and Western blot analyses for PrP<sup>sc</sup> (TeSeE<sup>™</sup> Western Blot Bio-Rad) were carried out at the National Veterinary Institute in Oslo, which is the national reference laboratory for TSEs.

#### PrP genotyping

PrP genotyping was performed on all scrapie positive sheep. To obtain an indication of PrP genotype distribution in the Norwegian sheep population every 16<sup>th</sup> sheep slaughtered and examined for PrP<sup>sc</sup> was PrP genotyped (Regulation (EC) No. 999/2001 Annex III, as amended by Regulation (EC) No 2245/2003).

Genotyping of scrapie positive sheep was performed on unfixed brain samples at the Department of Production Animal Clinical Sciences, Norwegian School of Veterinary Science. Genomic DNA was isolated using the DNeasy Tissue kit (QIAGEN). Polymorphisms in the PrP gene were detected through automated sequencing of a PCR-generated product covering codons 99 to 209 of the PrP open reading frame (forward primer 5' AGGCTGGGGTCAAGGTGGTAGC; reverse primer 5' TGGTACTGGGTGATGCACATTTGC). Genotyping of unfixed brain samples from the abattoir was performed at the Department of Basic Sciences and Aquatic Medicine, Norwegian School of Veterinary Science. DNA was extracted using the DNeasy 96 Tissue Kit (QIAGEN). The samples were amplified with the described forward and reverse primers modified by 5' attachment of M13-21 and M13 rev tails allowing the use of commercially available fluorescence labelled primers, and sequenced using Big Dye Primer chemistry (Applied Biosystems). Polymorphisms were identified by manual inspection of the sequence electropherograms.

#### Prevalence

The classical scrapie prevalences and scrapie Nor98 prevalences in the fallen stock and abattoir populations were estimated assuming a binominal distribution.

## Results

#### Sheep

Scrapie was diagnosed in 15 sheep from nine flocks. Five scrapie cases were identified in fallen stock, and four cases were apparently healthy animals slaughtered for human consumption (Table 1). One sheep from fallen stock had classical scrapie. This sheep, affected with classical scrapie, was analysed by scrapie/BSE discriminatory Western Blot (bio-Rad) and BSE was excluded. Additionally six sheep with classical scrapie were diagnosed in the same flock in connection with scrapie eradication.

Scrapie Nor98 were diagnosed in eight flocks. Both the diagnoses classical scrapie and scrapie Nor98 were based on the unique Western blot profile.

The individual age and breed were registered and the prion protein genotype examined for all nine scrapie cases (Table 2).

The identity of the flock was reported for 14,471 (94.1 %) of the total of 15,373 samples from sheep. In the event of a positive sample from slaughtered animals, the flock identity of the remaining samples (5.9 %) could be traced via the carcass number. The 14,471 samples were collected from 5,783 different sheep flocks. The mean number of animals tested per flock was 2.4 (range 1-71, flocks eradicated due to scrapie are excluded). From 1,763 flocks more than two samples were tested.

#### Goat

Scrapie Nor98 was demonstrated in one goat. The goat which was four years old came from Troms County, and it was submitted as part of the surveillance of fallen stock. None of the 48 flock mates' goats were found positive for  $PrP^{Sc}$ .

The identity of the herd was reported for 5,404 (94.7 %) of the total of 5,705 samples from goats. In the event of a positive sample from slaughtered animals, the herd

|   |                | · · · · · · · · · · · · · · · · · · · |          |          |
|---|----------------|---------------------------------------|----------|----------|
| Reason for submission to the laboratory             | No. of samples | No. of rejected<br>samples            | Negative | Positive |
| Sheep   |                |                                       |          |          |
| Animals with clinical signs consistent with scrapie | 25             | 0                                     | 25       | 0        |
| Fallen stock  | 4,607          | 11                                    | 4,591    | 5        |
| Healthy slaughtered animals                         | 10,316*        | 6                                     | 10,306   | 4        |
| Animals killed under scrapie eradication            | 425            | 0                                     | 419      | 6        |
| Total sheep   | 15,373         | 17                                    | 15,341   | 15       |
| Goats   |                |                                       |          |          |
| Animals with clinical signs consistent with scrapie | 3              | 0                                     | 3        | 0        |
| Fallen stock  | 347            | 2                                     | 344      | 1        |
| Healthy slaughtered animals                         | 5,305          | 2                                     | 5,303    | 0        |
| Animals killed under scrapie eradication            | 50             | 2                                     | 48       | 0        |
| Total goats   | 5,705          | 6                                     | 5,698    | 1        |

 Table 1. Brain samples from sheep and goats submitted for examination for scrapie in 2006

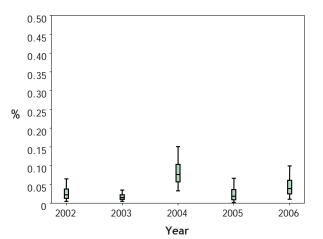
\* 104 samples from unspecified small ruminants tested negative. These samples are included in the figures given for sheep.

Table 2. Year of birth, reason for submission to laboratory examination, breed, prion protein genotype and type of scrapie of the scrapie cases detected in 2006

| Case no. | Year of birth | Reason for submission to<br>laboratory examination <sup>1)</sup> | Breed <sup>2)</sup>   | Prion Protein<br>Genotype | Scrapie type |
|----------|---------------|--|-----------------------|---------------------------|--------------|
| 1        | 1998          | Fallen stock   | Norwegian white breed | AF <sub>141</sub> RQ/ARR  | Nor98        |
| 2        | 2003          | Fallen stock   | Goat                  | ARQ/ARQ                   | Nor98        |
| 3        | 2004          | Fallen stock   | Sheep                 | VRQ/VRQ                   | Classic      |
| 4        | 1998          | Healthy slaughtered animals                                      | Norwegian white breed | AHQ/ARR                   | Nor98        |
| 5        | 2001          | Healthy slaughtered animals                                      | Norwegian white breed | AF <sub>141</sub> RQ/ARQ  | Nor98        |
| 6        | 1999          | Healthy slaughtered animals                                      | Norwegian white breed | AF <sub>141</sub> RQ/ARQ  | Nor98        |
| 7        | 2000          | Fallen stock   | Sjeviot               | AHQ/AF <sub>141</sub> RQ  | Nor98        |
| 8        | 2001          | Healthy slaughtered animals                                      | Norwegian white breed | AHQ/AF <sub>141</sub> RQ  | Nor98        |
| 9        | 1996          | Fallen stock   | Norwegian white breed | AHQ/ARH                   | Nor98        |
| 10       | 2002          | Fallen stock   | Norwegian white breed | AHQ/AF <sub>141</sub> RQ  | Nor98        |

<sup>1)</sup> The categories are: Healthy slaughtered animals, Animals killed under scrapie eradication measures, Suspect (clinical signs consistent with scrapie including animals showing clinical signs at ante-mortem inspection), Fallen stock (monitoring of fallen stock including animals examined because of other diseases than scrapie).

<sup>2)</sup> Crossbred long-tailed breeds: Rygja Sheep, Steigar Sheep, Dala Sheep, Norwegian White Sheep; indigenous short-tailed breed: Spæl Sheep.



**Figure 2.** Box and whiskers plot of the prevalence of scrapie Nor98 in slaughtered animals during 2002- 2006. The boxes represent the 25 % to 75 % quartiles and the whiskers represent the 2.5 % and 97.5 % exact binomial confidence intervals.

identity of the remaining samples (5.3 %) could be traced via the carcass number. The 5,404 samples were collected from 557 different goat herds. The mean number of animals tested per herd was 9.6 (range 1-169). From 395 flocks more than two samples were tested.

The geographical distribution of the sheep and goat populations is shown in Figures 4A and 4B. The origin of the sheep and goat samples and the origin of the scrapie cases are shown in Figures 5A and 5B.

The prevalence of scrapie in the fallen stock of sheep was estimated to 0.09 % (0.02-0.2 %), (95 % confidence interval [CI]), and the prevalence of scrapie in sheep slaughtered for human consumption was estimated to 0.04 % (0.01-0.1 %), (95 % CI).

PrP genotyping was performed on 578 sheep randomly sampled from the healthy slaughtered population. The PrP genotypes are grouped in accordance with the British National Scrapie Plan (NSP) (Table 3).

Table 3. PrP genotypes in the healthy slaughtered population in 2005 grouped in accordance with the British National Scrapie Plan (NSP)

| Genotype category  | Number | %     |
|--|--------|-------|
| NSP1, genetically most resistant, ARR/ARR  | 106    | 18.3  |
| NSP2, genetically resistant, ARR/ARQ,<br>ARR/ARH, ARR/AHQ, VRR/ARQ                       | 197    | 34.1  |
| NSP3, genetically low level resistant,<br>ARQ/ARQ  | 117    | 20.2  |
| NSP3, genetically low level resistant,<br>AHQ/AHQ, ARH/ARH, ARH/ARQ, AHQ/ARH,<br>AHQ/ARQ | 75     | 13.0  |
| NSP4, genetically susceptible, ARR/VRQ   | 21     | 3.6   |
| NSP5, genetically highly susceptible,<br>ARQ/VRQ, ARH/VRQ, AHQ/VRQ, VRQ/VRQ              | 62     | 10.7  |
| Total  | 578    | 100.0 |

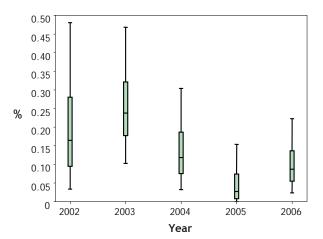


Figure 3. Box and whiskers plot of the prevalence of scrapie Nor98 in fallen stock during 2002- 2006. The boxes represent the 25 % to 75 % quartiles and the whiskers represent the 2.5 % and 97.5 % exact binomial confidence intervals.

#### Discussion

Scrapie Nor98 was diagnosed in eight sheep, each case originating in different flocks. The ages and genotypes of these sheep, and the results of the immunohistochemical examinations, were in accordance with the previous experience of scrapie Nor98 (6, 7, 8). There were two scrapie Nor98 cases, which had genotypes considered relatively resistant (NSP2) towards classical scrapie, and 6 cases had genotypes less resistant (NSP3) towards classical scrapie. Examination of 46 scrapie Nor98 cases has shown that the PrP genotype distribution differs markedly from that of the previous cases with classical scrapie and that polymorphisms at codon 141 and 154 in the ovine prion protein gene are associated with scrapie Nor98 (4).

In contrast the case with classical scrapie had the most susceptible genotype for this disease.

Following the EU Regulation (EC) No. 999/2001 Annex VII, as amended by Regulation (EC) No 1915/2003 all sheep in the eight scrapie Nor98 flocks were genotyped. Animals with a VRQ allele and animals without at least one ARR allele were killed and animals older than 18 months were examined for PrP<sup>sc</sup>, but no additional animals with scrapie Nor98 were detected in these flocks. In the flock with the classical scrapie case, it was additionally diagnosed six sheep with classical scrapie. In flocks with classical scrapie all small ruminants are killed.

The absence of additional scrapie Nor98 cases in the eradicated flocks this year as well as previous years, suggests that scrapie Nor98 is, if contagious at all, less contagious than classical scrapie. This is supported by a case-control study on scrapie Nor98 in Norwegian sheep flocks, where animal-to-animal contact or movement of sheep between sheep flocks were not found as risk factors for scrapie Nor98 (6).

Scrapie Nor98 was diagnosed in several different breeds. The sheep were between four and ten years old, which is in agreement with the result from previous years with the mean age being six years (Table 2). In contrast, the age of the classical case was two years and the mean age of cases with classical scrapie has been 3.5 years. The scrapie Nor98 cases detected in 2006 were located in counties where the disease has previously been diagnosed. Scrapie Nor98 is diagnosed in most parts of Norway, in 14 of 19 counties. In contrast, the classical form of scrapie, included the diagnosed case this year, has been detected only in the western part of Norway (3 counties) and in Nordland County.

The prevalence estimates of scrapie Nor98 in fallen stock and in sheep slaughtered for human consumption had varied during 2002-2006; however most estimates have been within the confidence intervals (Figure 2 and Figure 3) (7, 8, 9). The results from the surveillance programmes indicate that the prevalence of scrapie Nor98 in the sheep population has not changed since the start of the programme.

Classical scrapie was not diagnosed in 2005 and was last detected in one flock in 2004. When the classical form of scrapie was detected, the whole flock was killed. By the detection of classical scrapie in fallen stock, classical scrapie was for the first time detected through the active surveillance programmes. In the previous years, classical scrapie was detected only by examination of clinical cases or by follow up of contact flocks. By virtue that more than 95,000 sheep were examined since 2002, the prevalence of this type of scrapie is considered to be very low.

The difference between the number of examined sheep from fallen stock (4,607) and the calculated number according to EU regulation No 2245/2003 (10,000), may partly be due the fact that about 60 % of the fallen stock population die while on remote mountain and forest pastures. An additional explanation is that sheep and goat farmers are not informed of their duty to report to The Norwegian Food Safety Authority that all small ruminants that die, or are killed due to disease, on their farms. Inspite of that, the numbers of animals examined in the sheep fallen stock and slaughtered populations are sufficient to estimate the prevalences of scrapie Nor98 in these populations.

For monitoring of sheep, between one and 71 animals have been tested for PrP<sup>sc</sup> in the same flock. This indicates that in some flocks more than expected number of animals have been examined after random sampling from the slaughtered population. The mean Norwegian flock size counts 57 breeding sheep older than 12 months. Sheep from 5,783 of the approximate total of 15,800 flocks have been examined.

The first scrapie case in goats in Norway was diagnosed in 2006. This was a scrapie Nor98 type and the goat came from a county with a large goat population. Both classical and atypical scrapie in goats, are diagnosed in several countries in Europe (10). From 2005 Norway increased the scrapie testing of goats considerably, thus this diagnose in goat was not unexpected.

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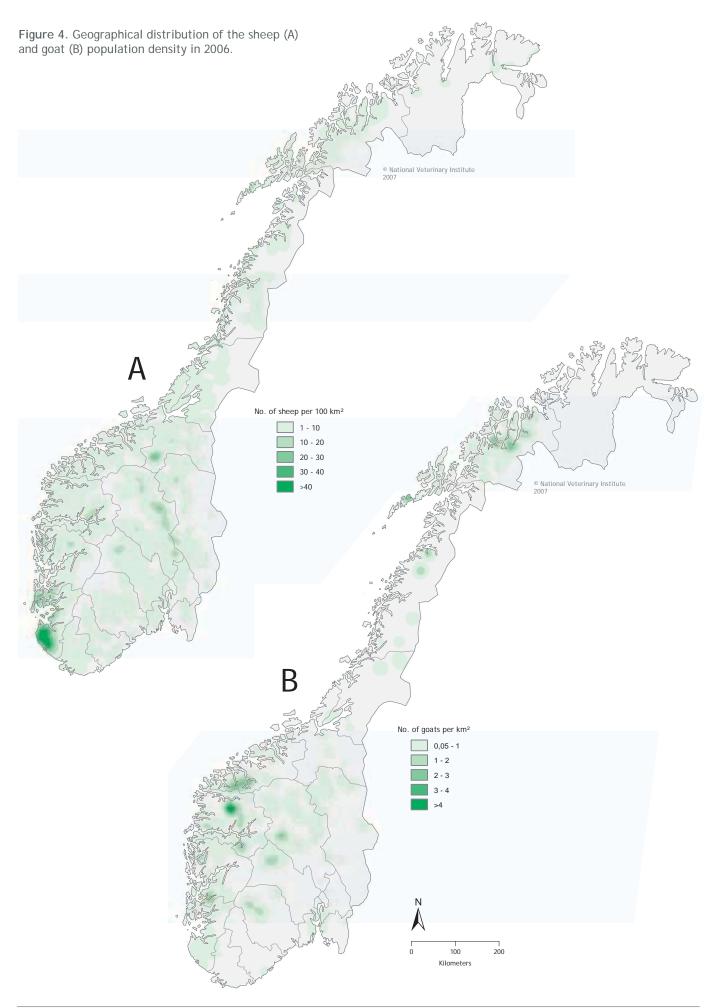
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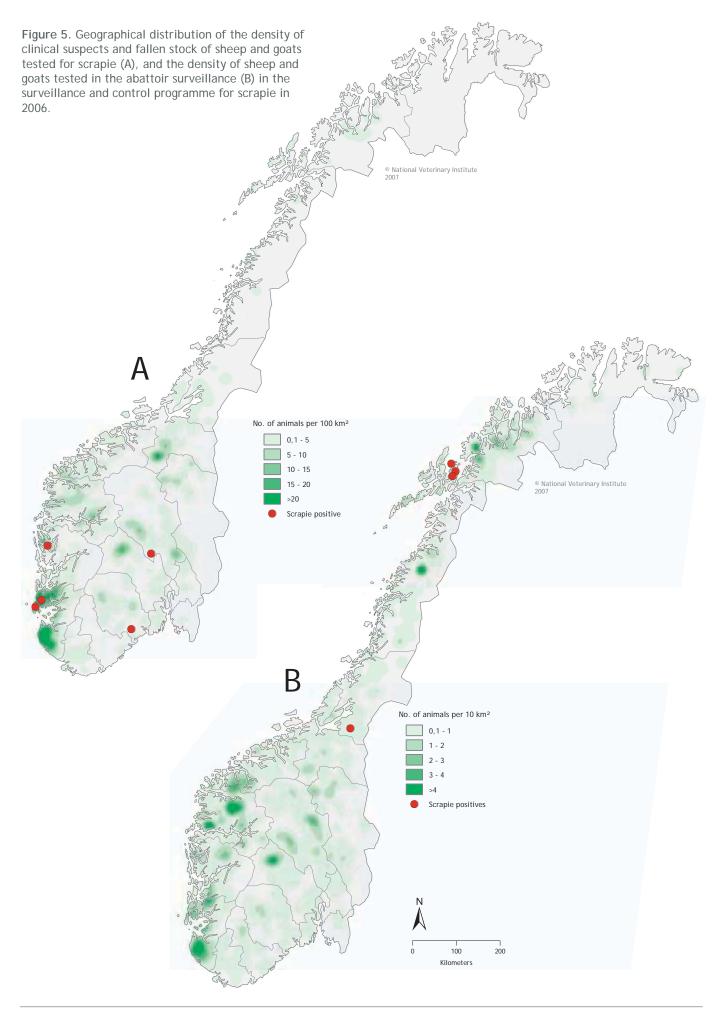
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The surveillance and control programme for specific virus infections in swine herds in Norway



Annette Hegermann Kampen Jorun Tharaldsen Peder Andreas Jansen

Surveillance in 2006 did not detect any cases of Aujeszky's disease, transmissible gastroenteritis, porcine respiratory corona virus, porcine respiratory and reproductive syndrome or swine influenza.

The national surveillance and control programme for specific virus infections in swine was launched in 1994 in order to document the status of Aujeszky's disease (AD), transmissible gastroenteritis (TGE), and porcine respiratory corona virus (PRCV) in the Norwegian swine population. Porcine respiratory and reproductive syndrome (PRRS) and swine influenza (SI) were included in the programme in 1995 and 1997, respectively. From 1997 to 1999 porcine epidemic diarrhoea (PED) was also included (1, 2), (Table 1).

The EFTA Surveillance Authority (ESA) has recognised the swine population in Norway as free from AD since July 1 1994, and has defined additional guarantees to protect the swine health status in Norway. The additional guarantees relating to AD for pigs destined for Norway are described in ESA Decision 75/94/COL, amending ESA Decision 31/94/COL, later replaced by ESA Decision 226/96/COL.

An overview of the material from previous years is presented in Figure 1. The Norwegian Food Safety Authority is responsible for running the programme, while the National Veterinary Institute is responsible for planning, laboratory analyses and reporting.

#### Aims

The aims of the programme are, through serological surveillance, to document absence of specific infectious diseases in the Norwegian swine population and to maintain this favourable situation.

Table 1. Monitoring of the Norwegian swine population for antibodies against Aujesky's disease (AD), transmissible gastroenteritis (TGE), porcine respiratory corona virus (PRCV), porcine epidemic diarrhoea (PED), porcine respiratory and reproductive syndrome (PRRS) and swine influenza (SI) during the years 1994 to 2005

| Year  | Herds<br>tested | Animals<br>pos/tested | Diseases included            |
|-------|-----------------|-----------------------|------------------------------|
| 1994  | 1,112           | 0/12,010              | AD, TGE, PRCV                |
| 1995  | 956             | 0/11,197              | AD, TGE, PRCV, PRRS          |
| 1996  | 468             | 0/4,968               | AD, TGE, PRCV, PRRS          |
| 1997  | 512             | 0/4,925               | AD, TGE, PRCV, PRRS, PED, SI |
| 1998  | 491             | 2*/4,695              | AD, TGE, PRCV, PRRS, PED, SI |
| 1999  | 470             | 0/4,705               | AD, TGE, PRCV, PRRS, PED, SI |
| 2000  | 458             | 0/4,600               | AD, TGE, PRCV, PRRS, SI      |
| 2001  | 472             | 0/4,972               | AD, TGE, PRCV, PRRS, SI      |
| 2002  | 492             | 0/4,899               | AD, TGE, PRCV, PRRS, SI      |
| 2003  | 483             | 0/4,783               | AD, TGE, PRCV, PRRS, SI      |
| 2004  | 492             | 0/4,935               | AD, TGE, PRCV, PRRS, SI      |
| 2005  | 468             | 1*/4,644              | AD, TGE, PRCV, PRRS, SI      |
| Total |                 | 3*/71,333             |                              |

 $^{\ast}$  2 positive for SI  $\rm H_{3}N_{2}$  in 1998 and 1 positive for PRCV in 2005, probably unspecific reactions.

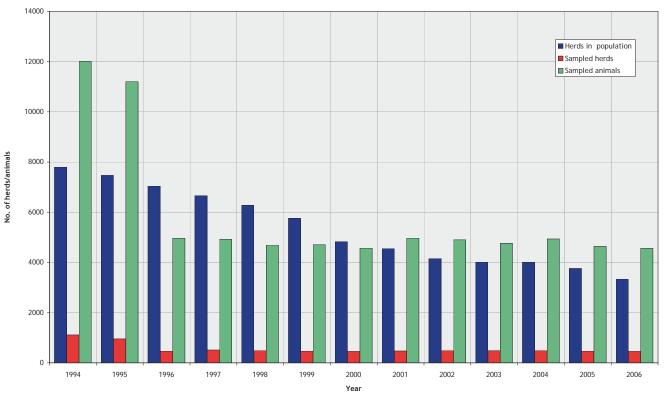


Figure 1. The size of the sampling frame and the number of sampled herds and animals in the Norwegian surveillance and control programme for specific virus infections in swine during the period 1994-2006.

## Material and methods

All the 164 nucleus and multiplying herds were to be tested. In addition, the nucleus units of all the 13 sow pools and a random selection of the remaining swine population were included in the programme. The random selection was conducted from all swine herds receiving governmental production subsidies according to records of 31 July 2005. The register contains 3,339 commercial swine herds of which 280 integrated and piglet-producing herds and 60 fattening herds were selected

The counties Østfold, Akershus, Vestfold and Rogaland were considered to be "high risk areas", and a relatively larger proportion of farms from these counties was selected.

Samples were collected at the farms except for the fattening herds which collected at six different abattoirs. From all herds, samples from ten pigs were to be collected.

#### Aujeszky's disease

All serum samples were tested for antibodies against AD virus using a commercial blocking ELISA (SVANOVIR<sup>™</sup>). The test detects antibodies against glycoprotein B (previously glycoprotein II) on the surface of the virus. For follow up of positive or dubious results, the SVANOVIR<sup>™</sup> PRV-gE was used.

## Transmissible gastroenteritis virus and porcine respiratory coronavirus

A combined blocking ELISA (SVANOVIR<sup>™</sup>) was used to detect antibodies against TGEV/PRCV. Depending on the reaction pattern of two different monoclonal antibodies against TGEV/PRCV and TGEV respectively, the test is able to distinguish between antibodies against TGEV and PRCV.

#### Porcine reproductive and respiratory syndrome

All serum samples were tested for antibodies against PRRS virus using the HerdChek PRRS 2XR Antibody Test Kit (IDEXX) which detects the most predominant European or American type of PRRS viruses. In the case of dubious or positive results, the samples were retested with blocking ELISAs and immune-peroxidase tests (IPT) at the Danish Institute for Food and Veterinary Research.

#### Swine influenza

To test for swine influenza, the samples were analysed for antibodies against the serotypes  $H_1N_1$  and  $H_3N_2$  in the hemagglutination inhibition test (HI), according to the method described in the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (3). The antigens were produced at the National Veterinary Institute in Oslo.

Table 2. Number of samples submitted to the laboratory and the test results for AD, swine influenza, and PRRS, PRCV and TGE in 2006

| Disease | Received | Rejected | Negative | Positive |
|---------|----------|----------|----------|----------|
| AD      | 4,569    | 14       | 4,555    | 0        |
| SI      | 4,569    | 17       | 4,552    | 0        |
| PRRS    | 4,569    | 10       | 4,559    | 0*       |
| TGE     | 4,569    | 27       | 4,542    | 0        |
| PRCV    | 4,569    | 28       | 4,541    | 0        |

 $^{\ast}$  The results from three samples from two flocks were inconclusive

All the serological analyses were performed at the National Veterinary Institute in Oslo. All inconclusive or positive samples in the routine tests were re-tested by specified reference tests.

#### **Results**

Blood samples from 4,569 individual animals were submitted and the results are shown in Table 2.

The distribution of tested herds in relation to type of production is given in Table 3. The mean number of animals tested per farm was 10 (range 2 - 25).

## Discussion

The results from the surveillance and control programme support freedom from specific virus infections in the Norwegian swine population. To date, there have been no clinical recordings indicating the presence of any of the viral infections included in this surveillance and control programme (1, 2, 4, 5).

The Norwegian swine industry has structurally changed during the last ten years with decline in number of herds but and increase in herd size. The produced tonnage of pork meat has been relatively stable.

The EU has not approved the programmes for virus infections other than AD for granting of additional guarantees, so they are continuously based on national decisions.

The fraction of sampled farms has not declined substantially since the start of the programme, the figures being 14.3 % and 13.7 % in 1994 and 2006, respectively. The geographical distribution of investigated farms is in accordance with the spatial distribution of the total swine herd population (Figure 2).

Farmed wild pigs and pigs kept as pets are not included in the programme. No wild boar population is registered in Norway.

Table 3. Distribution of swine herds in the surveillance and control programme 2006 related to the type of production

| Category                              | No. of<br>herds tested | % of<br>herds tested | Total no. of<br>individual samples<br>collected | % of individual samples collected |
|---------------------------------------|------------------------|----------------------|---|-----------------------------------|
| Nucleus herds and multiplying herds   | 142                    | 31                   | 1,457   | 32                                |
| Sow pools                             | 10                     | 2                    | 104   | 2                                 |
| Integrated and piglet-producing herds | 246                    | 54                   | 2,418   | 53                                |
| Fattening herds                       | 59                     | 13                   | 590   | 13                                |
| Total                                 | 457                    |                      | 4,569   |                                   |



The Norwegian swine population is relatively isolated. In 2006, the import consisted of only one live pet pig from Germany and 170 doses of swine semen from Finland and Sweden. In Sweden both PRCV and swine influenza occur.

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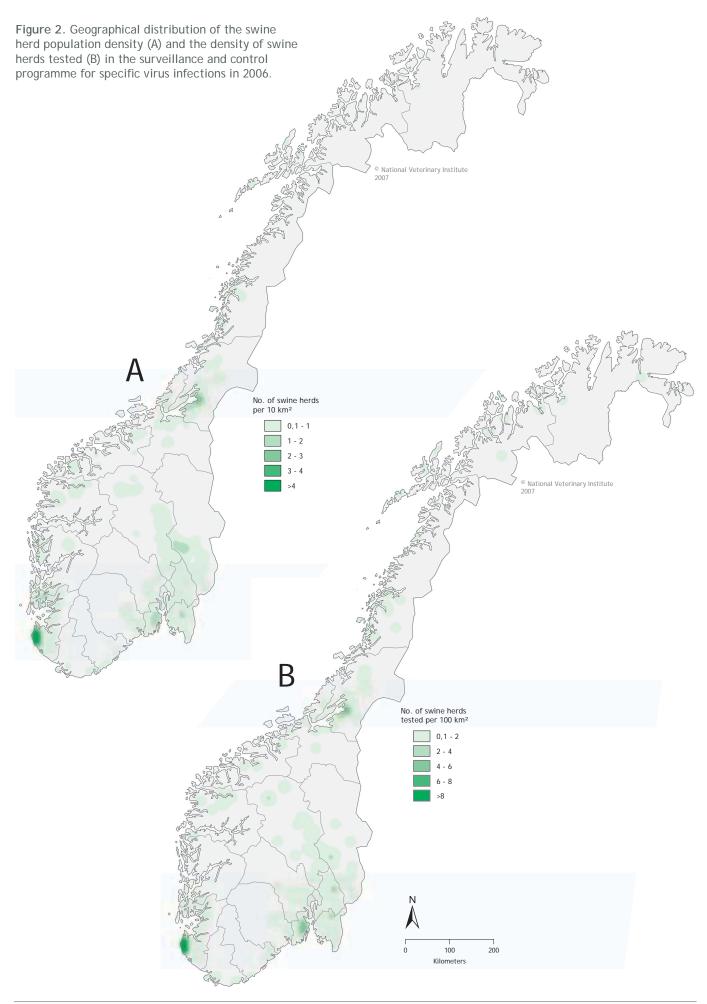
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Surveillance and control programmes in Norway · Specific virus infections in swine herds · Annual report 2006

The surveillance and control programme for chronic wasting disease (CWD) in wild and captive cervids in Norway



Turid Vikøren Ståle Sviland Petter Hopp Torfinn Moldal

Chronic Wasting Disease (CWD) was not detected in any of the animals tested in 2006.

CWD is a transmissible spongiform encephalopathy (TSE) of cervids (1, 2). A few species of the family Cervidae are known to be naturally susceptible to the disease: mule deer (Odocoileus hemionus), white-tailed deer (O. virginianus), Rocky Mountain elk (Cervus elaphus nelsoni), and moose (Alces alces). The disease has also been diagnosed in blacktailed deer (O. hemionus coulumbianus) in captivity. CWD was first described as a clinical syndrome termed "chronic wasting disease" in captive mule deer in Colorado, USA in the late 1960s and subsequently identified as a TSE in 1978 (1). In the mid-1980s, the disease was diagnosed in freeranging elk and deer. At present there is an endemic area for CWD in deer and elk comprised of northern Colorado, southern Wyoming, and western Nebraska. In recent years CWD has also been found in other U.S. states and in the two Canadian provinces Saskatchewan and Alberta.

The main clinical features of CWD-affected animals are progressive weight loss, changes in behaviour, and depression. In the terminal stages excessive drinking, urination and salivation are common. The clinical course of CWD has a span from a few days to approximately a year, however most animals die within a few weeks to some months. Affected animals are generally older than eighteen months (1, 2).

CWD is, like scrapie in small ruminants and bovine spongiform encephalopathy (BSE) in cattle, characterised by the accumulation of an abnormal form of the prion protein ( $PrP^{Res}$  or  $PrP^{CWD}$ ) in the central nervous system. In most of the CWD-affected animals, PrP<sup>cwD</sup> is also detectable in the lymphoid tissues (3). The histopathological changes are, like the other TSEs, characterised by vacuolation of the brain tissues (2). The diagnosis CWD relies on the detection of the PrP<sup>cwD</sup> by immunological methods such as immuno-histochemistry, ELISA or Western Blot.

In Norway, TSEs are restricted to some cases of both the classical and the atypical type (Nor98) scrapie in sheep and a single case of atypical scrapie in goat in 2006 (8). In 1994 a case of Feline Spongiform Encephalopathy (FSE) was detected.

Chronic wasting disease is yet to be diagnosed in cervids in Europe. The number of animals tested is however, low, despite efforts from Germany (4, 5), Belgium (6, 7), Finland and Norway.

Four cervid species are prevalent in natural populations in Norway: moose, red deer (*Cervus elaphus*), roe deer (*Capreolus capreolus*), and reindeer (*Rangifer tarandus*) Red deer predominate along the west coast, whereas moose and roe deer mainly inhabit other areas of the country. The wild reindeer live in dispersed populations in separate high mountain areas in southern Norway. The number officially hunted in 2006 was: 35,000 moose, 29,200 red deer, 29,900 roe deer, and 5,100 wild reindeer. Additionally, Norway has a semi-domestic reindeer population, mainly kept in the northern parts of the country, presently counting about 200,000 animals.

In Norway, red deer farming is not yet a large industry; however the number of herds is rising, with current estimated standing at 50 - 100 farms. Most of the farms keep red deer, and only a few keep fallow deer (*Dama dama*).



Surveillance and control programmes in Norway · CWD in cervids · Annual report 2006

Based on the fact that Norway has large populations of various cervids, a number of them grazing in regions where scrapie is detected, a voluntary survey for CWD in Norwegian wild and captive cervids has been set up. A passive surveillance programme on CWD was initiated in 2003, which includes all the four cervid species naturally occurring in Norway. During 2004-2006 a number of samples from slaughtered semi-domestic reindeer from several regions in the country also have been examined.

A small population (approximately 200) of free-ranging musk ox (*Ovibus moschatus*), inhabits the Dovre high mountain plateau in Mid-Norway. TSE has not been diagnosed in the musk ox, but the species has been included in the programme from 2004.

In 2006, the European Community put a motion that the Member States should carry out a survey for CWD in cervids (SANCO/960/2006), which was passed according to the Commission decision of 19 March 2007 (document number C(2007) 860). As an EEA EFTA state, the document is of relevance for Norway. This survey shall be completed no later than the end of the 2007 hunting season. The target species relevant for Norway is wild red deer and the survey implies sampling of a) clinical/sick, euthanized animals, b) traffic killed animals, c) animals found dead, and d) healthy animals shot during hunting. Additionally, for moose, roe deer, reindeer, and farmed deer the categories a) - c) should be sampled. All sampled animals should be over 18 months of age.

#### Aim

The aim of the programme is to detect the possible occurrence of CWD in the Norwegian cervid population.

# Material and methods

#### Material

As part of the EC survey, samples from adult wild red deer shot during the ordinary hunting season, September-November 2006 were tested. Tested animals also included captive deer and wild cervids older than 18 months that died or were euthanized due to disease or injuries. Additionally, cervids older than one year necropsied at the National Veterinary Institute were subjected to CWD testing. Some musk oxen and semi-domesticated reindeer were also tested.

#### Laboratory examinations procedures

A rapid test (TeSeE<sup>®</sup> Bio-Rad) was used to screen brain samples for detection of the PrP<sup>Res</sup> (PrP<sup>CWD</sup>). All the samples were analysed at the National Veterinary Institute in Oslo, which is the National Reference Laboratory for TSEs in Norway.

The National Veterinary Institute is part of the group "Control for Cervids" within the NeuroPrion Network of Excellence aiming at optimising diagnostics tools in Europe for the detection of CWD.

# Results

None of the 211 samples analysed in 2006 tested positive for CWD in the rapid test (Table 1).

Totally 176 of the tested animals were exclusively examined for CWD (Table 1). The remaining 35 animals represent routine necropsy material examined.

Four of the tested red deer were captive. All the tested reindeer were semi-domestic animals sampled during slaughter.

 Table 1. The number of cervids tested in the Norwegian surveillance and control programme for transmissible spongiform

 encephalopathies distributed in reason for submission

|          | Routine | necropsy |        | Total  |                                    |             |     |
|----------|---------|----------|--------|--|------------------------------------|-------------|-----|
| Species  | Captive | Wild     | Hunted | Traffic killed,<br>found dead or<br>euthanized<br>Wild | Found dead or<br>culled<br>Captive | Unspecified |     |
| Moose    |         | 11       |        |  |                                    | 1           | 12  |
| Red deer | 2       | 4        | 114    | 6  | 2                                  | 1           | 129 |
| Musk ox  |         | 13       |        |  |                                    |             | 13  |
| Reindeer |         |          |        |  |                                    | 48          | 48  |
| Roe deer |         | 5        | 4      |  |                                    |             | 9   |
| Total    | 2       | 33       | 118    | 6  | 2                                  | 50          | 211 |

#### Discussion

No animals were detected positive for CWD in 2006. The total number of samples collected and analysed is low. Chronic wasting disease has so far not been diagnosed in cervids in Europe. A study of brain tissue from 654 roe deer and 189 red deer in Bavaria came out negative for CWD (4). In another survey in Germany, 7,056 samples from deer (roe deer, red deer, fallow deer (*Dama dama*)) collected during ordinary hunting 2002-2005 tested negative for CWD (5). The authors concluded that CWD is unlikely to exist in free-living cervids from Germany. Also in Belgium, samples of spleen and brain from roe deer and red deer have tested negative for CWD (6, 7).

Among the Norwegian cervid species, a higher risk for CWD can be assumed for red deer since it is the same species as Rocky Mountain elk, and for moose. Moose has been found naturally infected with CWD in CWD-endemic areas in Colorado, USA. The first case was diagnosed in a survey of hunted animals in 2005, and in 2006 two new moose from the same population tested positive for CWD. Thus, the cases diagnosed in moose probably represent preclinical CWD. Also, the disease has been transmitted experimentally to moose by oral inoculation of brain tissue from a CWD affected mule deer (9). Roe deer, reindeer and musk ox has so far not been found naturally infected with CWD.



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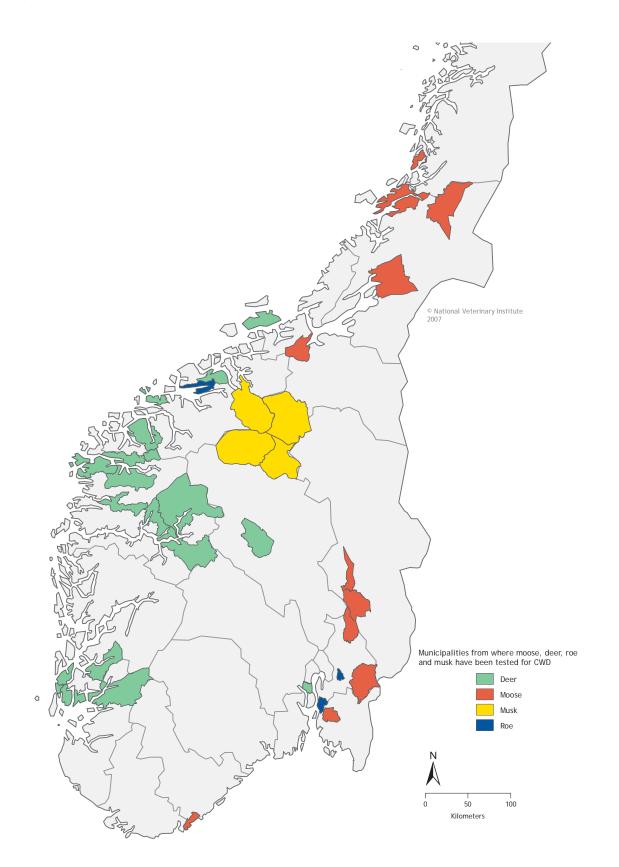
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Surveillance and control programmes in Norway · CWD in cervids · Annual report 2006

Figure 1. Municipalities in Norway from which cervids and musk ox were tested for chronic wasting disease (CWD) in 2006.



The surveillance and control programme for avian influenza (AI) in wild birds in Norway



Bruce David Kjell Handeland Jorun Tharaldsen Christine Monceyron Jonassen Knut Madslien Gry Grøneng

Results were negative for the 2006 surveillance for highly pathogenic avian influenza virus in wild birds.

The surveillance also revealed that Mallards, Wigeons, Gulls, and Teals are the most relevant reservoirs of influenza A virus in Norway.

The Norwegian Food Safety Authority is responsible for the implementation of the active surveillance programme for avian influenza (AI) in wild birds. The programme, which was started in 2005, is based on virological investigations in healthy, live or hunted birds. The National Veterinary Institute is responsible for planning, laboratory investigations and reporting components of the programme.

Al is a serious, highly contagious disease of poultry and other captive birds caused by many different subtypes of influenza type A viruses. The level of risks posed by the different subtypes to animal and public health is very variable and, are sometimes unpredictable. This is due to rapid virus mutation and possible re-assortment of the genetic material between different subtypes.

Wild waterfowls are the natural reservoirs for all influenza A virus subtypes. Infected birds do not usually develop clinical disease, but shed large amounts of virus in their faeces (1). The highly pathogenic avian influenza (HPAI) virus H5N1 is primarily shed via the airways (2).

HPAI has never been reported in wild birds of Norway.



#### Aims

The aim of the national surveillance programme for AI in wild birds is to study and understand the threats posed by wild birds in relation to influenza viruses of avian origin, with special emphasis to H5 and H7 viruses.

# Materials and methods

In 2006 the programme for wild birds consisted of molecular screening of cloacal and tracheal swabs from healthy birds shot mainly during the 2006 hunting season. Sampling equipment consisted of a sample tube containing a virus transport medium. Swabs were sent to hunters in the counties of Rogaland (South-Western Norway), Østfold and Hedmark (Eastern Norway), and Sør- and Nord-Trøndelag (Central Norway). Choice of hunters was based on their proficiency during previous hunting seasons. The hunters were also given written instructions on how to sample the animals. They were requested to fill in registration forms for individual birds. The swabs were taken from shot birds, and then placed in the transport medium. The swabs were sent by overnight post to the National Veterinary Institute in Oslo. The samples were frozen at 70 °C upon arrival.

The sampling comprised the following species (number sampled); Herring Gull (*Larus argentus*, 363) Mallard (*Anas platyrhynchos*, 359), Common Gull (*Larus canus*, 173), Wigeon (*Anas penelope*, 137), Teal (*Anas crecca*, 100), Great Black-Backed Gull (*Larus marinus*, 34), Black-headed Gull (*Larus ridibundus*, 19), Oystercatcher (*Haematopus ostralegus*, 18), Goldeneye (*Bucephala clangula*, 15), Lapwing (*Vanellus vanellus*, 15), Goosander (*Mergus merganser*, 7), Red-breasted Merganser (*Mergus serrator*, 6) Lesser Black-backed Gull (*Larus fuscus*, 5), Lesser White-fronted Goose (*Anser erythropus*, 5), Common Scoter (*Melanitta nigra*, 2), Hooded Crow (*Corvus conix*, 2), Kittiwake (*Rissa tridactyla*, 2) and Bean goose (*Anser fabalis*, 1). Also, samples from 6 unidentified species of Gull, 3 of Duck and 2 unmarked samples gave a total of 1,274 samples tested.

#### H5/H7

The samples were registered upon arrival and screened using a reverse transcriptase polymerase chain reaction (RT-PCR). The screening RT-PCR used was a pan-influenza A virus RT-PCR that reveals the presence of all subtypes of influenza type A virus. The method does not, however, give information as to which hemagglutinin (H) or neuraminidase (N) subtype is present in influenza positive samples. Therefore, the samples found to be positive in the initial pan-influenza A virus RT-PCR were further subtyped, using RT-PCRs specific for H5 and full-length RT-PCRs for the H and N genes. Samples positive for the pan-influenza A virus RT-PCR were also inoculated in embryonated eggs for virus isolation following the procedures described in the OIE Manual (3), with some minor modifications.

# Results

In total, samples from 1,274 birds were analysed. Of these, 85 were positive for influenza A virus. None of the samples were positive for HPAI viruses.

The prevalence for influenza A virus in waterfowls were as follows: Mallard 13.6 % (49/359), Widgeon 2.9 % (4/137), Teal 6.0 % (6/100), Goldeneye 0 % (0/15), Goosander 0 % (0/7), Red-breasted merganser 0 % (0/6), Lesser White-fronted Goose 0 % (0/5), Common Scoter 0 % (0/2) and Bean Goose 0 % (0/1). In addition, 3 negative samples were collected from unidentified birds defined as ducks.

And in other categories of birds; Herring Gull 3.6 % (13/363), Common Gull 3.5 % (6/173), Great Black-backed Gull 5.9 % (2/34), Black-headed Gull 26.3 % (5/19), Oystercatcher 0 % (0/18), Lapwing 0 % (0/15), Lesser Black-backed Gull 0 % (0/5), Carrion Crow 0 % (0/2) and Kittiwake 0 % (0/2). An additional 6 negative samples from unspecified Gulls, and 2 samples from unidentified birds were also negative.

None of the samples were H7 positive. Six Mallards were found to carry H5N2 subtypes. After sequencing of the H gene identified these viruses as low pathogenic avian influenza (LPAI) viruses. In addition, 4 samples were analysed as LPAI H5; 2 from Mallards, one from Herring Gull, and one from Wigeon.

The other subtypes identified included H1N1, H2N6, H3N6, H3N8, H4N2, H4N6, H6N1, H6N2, H9N2,H9N5, H10, H11, H12N2, H12N3, H12N8, H13N2, H13N6, H13N8 and H16N3.

# Discussion

Similar to 2005, there were positive samples from Mallards, Wigeons and Teals this year. But in comparison with the national surveillance programme for AI in wild birds 2005, the general prevalence of AI infection amongst the waterfowl tested in 2006 was lower. The prevalence amongst these species in 2005 was 20.4 %, 12.5 % and 30.9 %, respectively (4). Mallards were found to harbour the highest diversity of H and N subtypes. Subtype H5N2 was found in six Mallards, and H5 was found in a Mallard and a Wigeon.

New as of this year was the sampling of species from the Gull family. Four of the 5 gull species were positive for influenza A infection. The Lesser Black-backed Gull was negative. This was also the Gull species that was least sampled. The high prevalence recorded in samples from the Black-headed Gull (26.3 %) may be biased due to the low sampling number (n=19).

Also sampled were 4 other species, two of which are listed on EU's list of risk species - the Lapwing and the Carrion Crow. These birds - Lapwing, Oystercatcher, Carrion Crow and Kittiwake - all tested negative. The low sampling of these species makes it difficult to offer any conclusions as to their importance in their role in the threat of avian influenza.

The findings of this study indicate that Mallards, Wigeons, Gulls, and Teals are the most relevant reservoirs of influenza A virus in Norway.

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The surveillance and control programme for avian influenza (AI) in poultry in Norway



Bruce David Jorun Tharaldsen Gry Grøneng

The 2006 surveillance of avian influenza in poultry and birds in Norway did not detect any signs of Avian Influenza infection.

The Norwegian Food Safety Authority is responsible for implementing the surveillance programme for avian influenza (AI) in poultry. The programme, which was started in 2005, is based on serological investigations of poultry. The National Veterinary Institute is responsible for planning, laboratory investigations and reporting components of the programmes.

Al is a serious, highly contagious disease of poultry and other captive birds caused by many different subtypes of influenza type A viruses. The level of risks posed by the different subtypes for animal and public health, is very variable and can be unpredictable. This is due to the rapid virus mutation and possible re-assortment of the genetic material between different subtypes.

Current knowledge indicates that the health risks posed by the so-called Low Pathogenic AI (LPAI) viruses are lower than that posed by Highly Pathogenic AI (HPAI) viruses. The HPAI viruses originate from a mutation of LPAI viruses of either H5 or H7 subtype. HPAI can cause disease in poultry resulting in mortality rate exceeding 90 %.

In general, domestic poultry populations are free from Al viruses. However, wild waterfowl are the natural reservoirs for all influenza A virus subtypes. Infected birds do not usually develop clinical disease, but may shed large amounts of virus in their faeces upon infection (1). An avian influenza virus surveillance programme in wild waterfowl in Norway was started in 2005. The national surveillance and control programme for Al in poultry was started in 2006 and is modelled on EU's Council Directive 2005/94/EC, also known as the "Al Directive".

Al has never been reported or diagnosed in poultry in Norway.

#### Aims

The aim of the national surveillance and control programme for AI in poultry is to document that the various poultry populations in Norway are free of influenza A virus of subtypes H5 and H7 and to contribute to the maintenance of this status.

# Materials and methods

The programme in 2006 consisted of serological screening of blood samples from poultry. Poultry deemed at risk for exposure to influenza type A were preferentially sampled as outlined in EU's AI Directive Annex I. The basis for sample selection was based upon a risk assessment published by the National Veterinary Institute in February 2006 (2). The sample selection included chickens, turkeys, ducks, geese, quail and ostrich.

In addition to the samples taken from farms on the basis of the risk assessment (2), samples from breeding flocks were also tested for AI. According to the national regulations for certification of poultry breeding farms (Forskrift om sertifisering av fjørfevirksomheter av 18.11.94), blood samples from 60 birds must be taken at least once a year from every breeding flock. These blood samples are to be tested for Newcastle disease, as Norway has the status of a nonvaccinating country. Such samples from chicken, turkey, and duck flocks were included in the national surveillance and control programmes for AI.

Blood samples were collected from all species of poultry from at least 10 birds per holding, with the exception of ducks and geese. If there were more than one shed on the holding, all sheds were sampled. From ducks and geese, 50 samples were to be taken from each selected holding. In those instances where the flock size was less than the number required, all birds in the flock were sampled.

The samples taken from chickens were tested for the presence of antibodies against Influenza A virus. Due to the limitations on the species spectrum of the ELISA for influenza A virus, samples from the remaining species were tested for influenza A virus subtype H5 and subtype H7.

#### Influenza A

An ELISA kit produced by IDEXX was used for the testing of antibodies against influenza A virus. The test has been demonstrated to detect antibody reactivity to 20 different subtypes of avian influenza including 14 hemagglutinin glycoproteins and the H5N1 subtype.

This test is only validated for use in chickens. If tests were positive, samples were examined further for presence of H5 or H7 with the haemagglutination inhibition test, see below.



#### H5/H7

All serum samples from species other than chicken were tested for specific antibodies against both H5 and H7 with the haemagglutination inhibition test described in the OIE diagnostic manual (3).

### Results

Tables 1 and 2 show the number of flocks and birds tested in the different poultry species in the national surveillance and control programmes for Al in 2006. Twelve chicken flocks - 7 breeder, 2 commercial and 3 hobby - gave inconclusive results when tested for antibodies against Influenza A virus. Subsequent testing of these samples with haemagglutination inhibition tests however showed no sign of antibodies against either H5 or H7. All other samples were negative.

A number of samples (n=1100) taken for the purposes of diagnosing disease, production problems and the control of imported animals were also screened for antibodies against Influenza A virus (n=272) or H5/H7 (n=828). All were negative.

Table 1. Number of commercial flocks, hobby flocks and birds tested in the surveillance and control programme for AI in poultry in 2006

| Species               | Commercial<br>flocks tested | Hobby flocks<br>tested | Total number of birds tested |
|-----------------------|-----------------------------|------------------------|------------------------------|
| Chicken               | 74                          | 25                     | 959                          |
| Turkey                | 47                          | 1                      | 485                          |
| Duck                  | 4                           | 8                      | 257                          |
| Goose                 | 1                           | 2                      | 54                           |
| Quail                 | 2                           | 2                      | 87                           |
| Ostrich               | 3                           | 0                      | 23                           |
| Peacock <sup>1</sup>  | 0                           | 1*                     | 9                            |
| Pheasant <sup>1</sup> | 0                           | 1*                     | 1                            |
| Penguin <sup>1</sup>  | 0                           | 1*                     | 8                            |
| Total                 | 131                         | 41                     | 1,883                        |

<sup>1</sup> Not tested as a part of surveillance program, \* zoo animals

Table 2. Number of certified breeder flocks and birds testedin the surveillance and control programmes for AI in poultryin 2006

| Species | Number of flocks | Number of birds tested |
|---------|------------------|------------------------|
| Chicken | 127              | 1,276                  |
| Turkey  | 4                | 40                     |
| Duck    | 3                | 30*                    |
| Total   | 134              | 1,316                  |

\* Mistake made in number of samples tested from ducks

### Discussion

An adequate number of flocks were sampled with respect to Norwegian population of commercial poultry. A misinterpretation of the AI Directive resulted in the testing of only 10 samples from each of three duck breeder flocks being tested for AI, when the required number should be 50 birds per flock.

With respect to hobby poultry, it is difficult to know the true population numbers. The Norwegian Food Safety Authority is working on a voluntary registry for people who keep poultry hobby on a hobby basis. If they succeed, this will be invaluable for future surveillance coverage.

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http://www.oie.int/eng/normes/mmanual/A\_00037.htm

The surveillance and control programme for infectious laryngotracheitis (ILT) and avian rhinotracheitis (ART) in poultry flocks in Norway



Bruce David Jorun Tharaldsen Gry Grøneng

#### *Surveillance in 2006 did not detect infectious laryngotracheitis (ILT) and avian rhinotracheitis (ART) in chicken and turkey flocks.*

The Norwegian Food Safety Authority is responsible for the implementation of the surveillance and control programmes for infectious laryngotracheitis (ILT) and avian rhinotracheitis (ART) in chicken and turkey flocks, respectively. Started in 1998, these programmes are based on serological investigations. The National Veterinary Institute in Oslo is responsible for the planning, laboratory investigations and the reporting components of the programmes.

ILT is a severe respiratory disease in chickens, and was first described in the USA in the 1920s. Since then, the disease has been seen in most parts of the world, including most European countries (1). However, ILT has not been diagnosed in commercial chicken flocks in Norway since 1971, although clinical outbreaks of ILT have occurred sporadically in Norwegian hobby flocks since 1998 (2). ILT is an OIE listed disease, and in Norway, it is a notifiable list A-disease.

ART is a highly contagious infection which affects the upper respiratory passages of poultry. The disease is called turkey rhinotracheitis (TRT) in turkeys and swollen head syndrome (SHS) or ART in chicken. The disease is caused by avian pneumovirus (APV), and was first described in South Africa in the 1970s. Since then, the disease has been diagnosed in most countries (1) and sporadically in our neighbouring countries. In Norway ART is a notifiable list B-disease although the disease is not notifiable in the OIE-system.

ART had never been diagnosed in Norwegian poultry until the national surveillance and control programmes for ART demonstrated the presence of antibodies against APV in 2003 and 2004. The two affected farms; one broiler breeder farm and one layer breeder farm were located in the same area, approximately four kilometres apart. However, a common infection source was never identified. In spite of numerous failed attempts to isolate and identify the infectious agent that caused the seroconversion, none were found. The diagnosis for ART was thus based on serology only, like in many other countries (1).

Clinical symptoms were not observed in any of the flocks that tested positive in 2004/2005. As the use of stamping out measures was unable to check the spread of the infection, chickens were excluded from the national surveillance and control programme for ART as of May 2005.

#### Aims

The aims of the national surveillance and control programmes for ILT and ART are to document that the commercial poultry populations in Norway are free from these infections, and to contribute to the maintenance of this status.

# Materials and methods

According to the national regulations for certification of poultry breeding farms (3), blood samples from 60 birds must be taken at least once a year from every breeding flock at the farms. These blood samples are to be tested for Newcastle disease, as Norway is a non-vaccinating country. Thirty of the 60 samples from chicken and turkey flocks are included in the national surveillance and control programmes for ILT and ART. Blood samples from chickens are tested for antibodies against ILT; while the samples from turkeys are tested for antibodies against APV. In addition to the turkey breeding flocks, blood samples are collected at the abattoir from 40 randomly selected turkey flocks. These samples are also tested for antibodies against APV.

#### ILT

An indirect ELISA-test produced by Synbiotics, was used for the testing of antibodies against the ILT-virus.

#### ART

All serum samples were tested for specific antibodies against APV with a blocking-ELISA produced by SVANOVA, Uppsala, Sweden.

Flocks with single positive reactions are followed up by repeated sampling, and if false positive results can't be ruled out by this procedure, serum samples with a positive reaction in the ELISA-tests are submitted to the Veterinary Laboratories Agency (VLA), Weybridge, England for testing using virus neutralisation tests.

#### **Results**

All 869 blood samples analysed in the surveillance programme for ART were negative.

All 3811 blood samples analysed in the surveillance programme for ILT were negative.

Tables 1 and 2 show the number of farms, flocks and birds tested in the different poultry production types in the national surveillance and control programmes for ILT and ART, respectively, in 2006.

#### Discussion

According to the plan for sampling from randomly selected turkey flocks at the time of slaughter, too few flocks were sampled (25 of 40). This discrepancy was a result of the change in the sampling programme for ART not enforceable by law until October 2006. This hampered the collection of samples at the abattoirs.

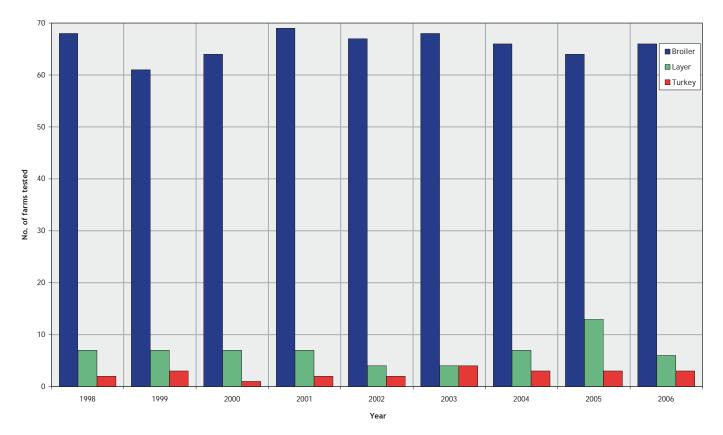


Figure 1. The number of farms tested in the surveillance and control programmes for infectious laryngotracheitis (ILT) and avian rhinotracheitis (ART) in poultry flocks in Norway during the time period 1998-2006.

| Table 1 Number of forme     | flools and hirds tested in the surveillance and control | programmas for IIT in poultry in 2004 |
|-----------------------------|---|---------------------------------------|
| Table 1. Number of Tarms    | flocks and birds tested in the surveillance and control | programmes for it is nooutry in 2006  |
| rabie in italikeer et laine |   |                                       |

| Production | No. of farms tested | No. of flocks tested | Total no. of<br>birds tested | Flocks with seropositive samples |
|------------|---------------------|----------------------|------------------------------|----------------------------------|
| Broiler    | 66                  | 112                  | 3,361                        | 0                                |
| Layer      | 6                   | 15                   | 450                          | 0                                |
| Total      | 72                  | 127                  | 3,811                        | 0                                |

Table 2. Number of farms, flocks and birds tested in the surveillance and control programmes for ART in poultry in 2006

| Production      | No. of farms tested | No. of flocks tested | Total no. of<br>birds tested | Flocks with seropositive samples |
|-----------------|---------------------|----------------------|------------------------------|----------------------------------|
| Turkey breeders | 3                   | 4                    | 120                          | 0                                |
| Turkey          | 25                  | 25                   | 749                          | 0                                |
| Total           | 28                  | 29                   | 869                          | 0                                |

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The surveillance and control programme for *Campylobacter* in broiler flocks in Norway



Merete Hofshagen Margareth Opheim

Campylobacter sp. was detected in 4.9 % of the 3,908 flocks investigated in 2006.

Campylobacteriosis is currently the most commonly reported bacterial infectious disease in the Norwegian human population. The incidence increased by 145 % from 1997 to 2001 but has since then declined slightly. In almost half of the cases, the infection is acquired in Norway. Consumption of poultry meat purchased raw has been identified as a significant risk factor together with drinking undisinfected water, eating at barbecues, occupational exposure to animals, and eating undercooked pork (1).

The action plan regarding *Campylobacter* in Norwegian broilers has been running since spring 2001 (2, 3, 4). The action plan is a joint effort involving several stakeholder groups from "stable-to-table". The Norwegian Zoonosis Centre at the National Veterinary Institute coordinates the programme, and is responsible for the collection and analyses of data and the communication of results.

#### Aim

The objective is to reduce the human exposure to thermophilic *Campylobacter* through Norwegian broiler meat products.

# Materials and methods

The action plan consists of three parts; a surveillance programme including all Norwegian broiler flocks, a followup advisory service to farms with *Campylobacter* positive flocks, and surveys of broiler meat products. The action plan is updated regularly and the details for 2006 together with other information regarding the action plan, including the results from the product survey, can be found at www. zoonose.no.

#### Surveillance

All Norwegian broiler flocks that are slaughtered before 50 days of age are sampled pre-slaughter by the owner maximum four days before slaughter. The sample consists of ten pooled swabs from fresh faecal droppings. The samples are submitted to the National Veterinary Institute's laboratory in Trondheim, where they are analysed by PCR. The carcasses from the positive flocks are either heat treated or frozen for a minimum of three weeks before being marketed. All flocks are tested upon arrival at the slaughter plant by sampling ten caeca per flock at the slaughter line. Contents of the ten caeca are pooled into one sample and analysed by local laboratories. Samples are analysed using

the method described in NMKL no. 119, 1990, with minor modifications. Carcasses from flocks which are only positive at the slaughterhouse sample are not automatically heat treated or frozen.

#### Follow-up of positive flocks

An advisor from the poultry industry or the Municipal Food Safety Authority will pay a follow-up visit to *Campylobacter* positive broiler farms. The visit should result in measures on the farm to reduce the risk of flocks becoming contaminated with *Campylobacter* in the future.

#### Surveys of broiler meat products

According to the plan for 2006, four Municipal Food Safety Authorities should collect 25 samples from retail each month from March to December. Samples were analysed using the method described in NMKL no. 119, 1990, with minor modifications.

#### **Results**

A total of 3,908 flocks from 526 broiler farms were tested. These flocks were slaughtered in 4,053 batches (a batch is defined as all chickens from one flock slaughtered on the same day). A total of 127 flocks were slaughtered in two or more batches. In addition, four flocks were split in two batches as they were slaughtered at two different slaughterhouses on the same day.

Overall, 190 (4.9%) flocks (191 (4.7%) batches) were positive for *Campylobacter* sp. either at pre-slaughter, at slaughter, or at both sampling times. For positive slaughterhouse samples confirmed by the reference laboratory, *C. jejuni* was isolated from 92.6% and *C. coli* from 7.4%. For five flocks, a positive diagnosis was not verified; one of these flocks was negative at the pre-slaughter sample.

Of the 190 positive flocks, 142 (74.7 %) tested positive at pre-slaughter sampling. These carcasses were frozen or heat treated in order to prevent contaminated poultry from reaching the general market as fresh broiler meat. A total of 22 flocks (11.6 %) tested positive only at pre-slaughter.

The positive flocks came from 140 (26.6 %) of the tested farms. Of these positive farms, 113 (80.7 %) had only one positive event during 2006 (a positive event is defined as one positive flock or as several parallel positive flocks from different houses) and produced 119 (62.6 %) of the positive flocks. A total of 19 (13.6 %) of the farms had two positive events (producing 44 (23.2 %) of the positive flocks), and eight (1.5 %) had three positive events. The 27 farms with two or more positive events in 2006 (5.1 % of all farms) accounted for 37.4 % of all positive flocks.

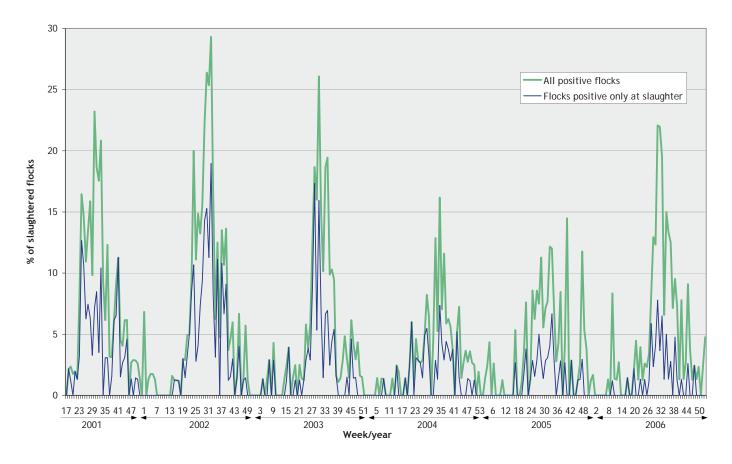


Figure 1. Weekly incidence of Campylobacter sp. in slaughtered Norwegian broiler flocks from week 18 in 2001 throughout 2006.

The proportion of *Campylobacter* positive flocks and the proportion of flocks testing positive only at slaughter has varied substantially since the action plan was launched (Figure 1). Regional differences in the proportions of positive flocks and farms are shown in Table 1.

#### Discussion

In the first years of the action plan, when the pre-slaughter samples were taken approximately eight days before slaughter, approximately 50 % of the positive flocks were detected only at slaughter. From 1 March 2005 onwards, all flocks had to be sampled maximum four days before slaughter. This contributed to the fact that in 2005, 31.8 % of the positive flocks were detected only at slaughter and in 2006 this was further reduced to 25.3 %.

Most farmers follow the guidelines regarding time of preslaughter sampling. A total of 216 (5.3 %) slaughter batches were sampled earlier than four days before slaughter, mostly in connection with holidays. In total, less than 0.7 % of the flocks were not sampled according to the action plan (i.e. sampled only once).

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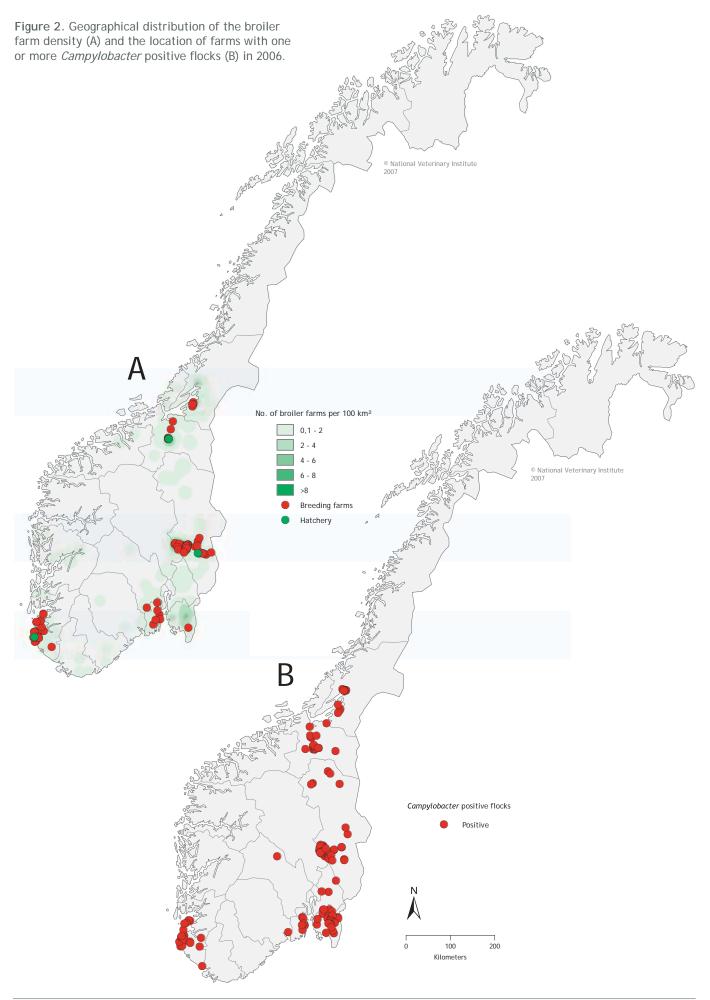
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Table 1. Campylobacter positive farms and flocks by county in Norway 2006

|                 |     | Farms        |        | Flocks |              |       |  |
|-----------------|-----|--------------|--------|--------|--------------|-------|--|
| County          | N   | No. positive | (%)    | N      | No. positive | (%)   |  |
| Østfold         | 81  | 25           | (31.0) | 718    | 32           | (4.0) |  |
| Akershus        | 14  | 3            | (21.0) | 111    | 6            | (5.0) |  |
| Hedmark         | 113 | 45           | (40.0) | 878    | 62           | (7.0) |  |
| Oppland         | 10  | 0            | (0.0)  | 55     | 0            | (0.0) |  |
| Buskerud        | 11  | 1            | (9.0)  | 67     | 1            | (1.0) |  |
| Vestfold        | 34  | 6            | (18.0) | 225    | 6            | (3.0) |  |
| Telemark        | 5   | 1            | (20.0) | 24     | 1            | (4.0) |  |
| Aust-Agder      | 3   | 0            | (0.0)  | 24     | 0            | (0.0) |  |
| Vest-Agder      | 4   | 1            | (25.0) | 24     | 1            | (4.0) |  |
| Rogaland        | 92  | 27           | (29.0) | 735    | 38           | (5.0) |  |
| Hordaland       | 14  | 0            | (0.0)  | 86     | 0            | (0.0) |  |
| Møre og Romsdal | 3   | 0            | (30.0) | 22     | 0            | (0.0) |  |
| Sør-Trøndelag   | 67  | 20           | (15.0) | 416    | 27           | (6.0) |  |
| Nord-Trøndelag  | 75  | 11           | (31.0) | 523    | 16           | (3.0) |  |
| Total           | 526 | 140          | (26.6) | 3,908  | 190          | (4.9) |  |



Surveillance and control programmes in Norway · Campylobacter in broiler flocks · Annual report 2006

The surveillance and control programme for viral haemorrhagic septicaemia (VHS) and infectious haematopoietic necrosis (IHN) in Norway

Hege Hellberg Birgit Dannevig Irene Ørpetveit Attila Tarpai



# VHS virus and IHN virus were not detected on any of the sites tested for surveillance in 2006.

Viral haemorrhagic septicaemia (VHS) and infectious haematopoietic necrosis (IHN) are two important rhabdovirus infections in salmonid fish (1). VHS occurs in continental Europe and is an important disease in rainbow trout (Oncorhynchus mykiss) farming due to its clinical and economic consequences. In 2006, VHS was diagnosed in rainbow trout in Great Britain, a country previously free of the disease. VHS also re-occurred in Romania in 2006, where the disease was diagnosed in rainbow trout imported as embryonated spawn from Denmark. A specific strain of VHS virus has caused disease in several wild Pacific fish species (2). VHS has also been diagnosed in farmed Japanese flounder (Paralichthys olivaceus) and turbot (Schophthalmus maximus) (3, 4). This marine strain is not pathogenic to rainbow trout. Marine VHS virus has been isolated from several marine fish species in North European coastal waters (the English Channel, the Baltic Sea, the North Sea, the Norwegian Sea, Skagerak) (1).

Infectious haematopoietic necrosis has led to serious economic losses in farmed rainbow trout and salmon, and the disease has also had an impact on wild populations of Pacific salmon. The disease was first described in Europe in 1985, in France and Italy. In 2006, IHN was diagnosed in rainbow trout in the Czech Republic and Slovenia. The disease has never been diagnosed in Norway. For more detailed information on VHS and IHN, reference is made to previous reports of the surveillance and control programmes (5, 6).

In 1994, Norway obtained disease free status for VHS and IHN based on health control information and virological examinations carried out in fish farms since 1967 (7). Norway has operated a surveillance programme in accordance with Directive 91/67 EEC since the autumn of 1994 (8). The Norwegian Food Safety Authority is responsible for the programme and for inspection and sampling. The National Veterinary Institute is responsible for laboratory procedures and analyses in accordance with Commission Decision 2001/183/EC (9) and prepares the report.

#### Aim

The aim of the programme is to document the absence of VHS virus and IHN virus in Norwegian fish farms and maintain Norway's approved zone status.

# Materials and methods

#### Sampling

Sampling and inspection is carried out by the District Offices of the Norwegian Food Safety Authority. The yearly sampling schedules covers approximately 50 % of farms (sites) producing susceptible species. According to Directive 91/67/EEC (8) and Decision 2001/183/EC (9), all fish farms producing species susceptible to VHS and IHN should be sampled over a two-year period. Inspection and sampling is carried out when the water temperature is below 14 °C. Thirty fish are sampled from each site. Organ samples for virological examination for VHS virus and IHN virus must contain spleen, anterior kidney and heart or brain. For brood fish, ovarian fluid can be included. Samples from ten fish may be pooled to form a single sample. For fry (<4 cm), samples must include head and viscera, and five individuals may be pooled to form a single sample. In farms containing rainbow trout, all samples must be derived from this species. In farms where rainbow trout is not present, all other susceptible species must be sampled on an equal basis. Samples are collected in transport medium for virological analysis and sent to the National Veterinary Institute for analysis.

#### Analysis

Samples must arrive at the laboratory within 48 hours of sampling. According to the specifications of Decision 2001/183/EC (9), the samples must be kept cool during transport; the temperature shall not exceed 10 °C. At arrival, samples are homogenised and suspended in the original transport medium and centrifuged at 4 °C. Infectious pancreatic necrosis (IPN) virus is ubiquitous in Norwegian fish farms and all samples are neutralised with IPN virus antiserum prior to inoculation on cell cultures to prevent IPN virus from masking possible VHS/IHN virus present in the samples. Neutralized homogenate is then inoculated on BF-2 and EPC cells as specified (9). Inoculated cells are incubated at 15 °C for 7 to 10 days and observed for cytopathogenic effect (CPE). If no CPE is observed, subcultivation is performed on fresh cell cultures. If CPE is observed, virus is identified as specified by Decision 2001/183/EC and recommendations from EU reference laboratory for fish diseases in Århus, Denmark.

#### **Results**

In 2006, a total of 1,239 pooled samples (12,390 individual fish) from 392 sites were examined (Table 1 and 2, Figure 1 and 2). VHS virus and IHN virus were not detected.

In samples from seven submissions, CPE appeared in the BF-2 cell cultures that could not be ascribed to neither IPN virus nor VHS virus as tested by virus neutralization test and reverse transcription polymerase chain reaction (RT-PCR), respectively. Further investigations of cell cultures exhibiting CPE by immunofluorescence tests and RT-PCR revealed the presence of salmonid alpha virus, the causative agent of pancreas disease (PD). Three of these submissions, all from locations in Hordaland, consisted of samples from rainbow trout. The other four submissions (two from Hordaland, one from Sogn and Fjordane and one from Finnmark) consisted of samples from Atlantic salmon. PD was diagnosed in all but two sites either before or after submission of samples for VHS and IHN surveillance. The two non-diagnosed sites both contained rainbow trout.

| Table 1. | Different | categories | of fish | analysed | for VHS | virus | and IHN | virus in 200 | 06 |
|----------|-----------|------------|---------|----------|---------|-------|---------|--------------|----|
|          |           |            |         |          |         |       |         |              |    |

|  | Fry - smolt |                     | On-gr     | rowing              | Broo      | d fish              | Total     |                     |
|--|-------------|---------------------|-----------|---------------------|-----------|---------------------|-----------|---------------------|
|  | No. sites   | No. of fish sampled | No. sites | No. of fish sampled | No. sites | No. of fish sampled | No. sites | No. of fish sampled |
| Atlantic salmon<br>( <i>Salmo salar</i> L.)    | 74          | 2,420               | 235       | 7,010               | 8         | 240                 | 316       | 9,670               |
| Rainbow trout<br>( <i>O. mykiss</i> )          | 9           | 270                 | 35        | 1,040               | 6         | 180                 | 49        | 1,490               |
| Brown trout<br>(Salmo trutta L.)               | 23          | 860                 | 1         | 30                  | 1         | 20                  | 24        | 910                 |
| Arctic char<br>( <i>Salvelinus alpinus</i> L.) | 2           | 70                  | 6         | 180                 |           |                     | 8         | 250                 |
| Turbot<br>(Scophthalmus maximus L.)            |             |                     | 1         | 30                  |           |                     | 1         | 30                  |
| Sea trout<br>( <i>S. trutta</i> L.)            | 2           | 40                  |           |                     |           |                     | 2         | 40                  |
| Total  | 104*        | 3,660               | 276*      | 8,290               | 15        | 440                 | 392*      | 12,390              |

\* The total number of sites may be less than the sum of sites per species as some sites produce more than one species.

| Table 2. Number of farms | and species analysed for VHS | 5 virus and IHN virus du | uring the time period 1995-2006 |
|--------------------------|------------------------------|--------------------------|---------------------------------|

| Farm types                        | 1995 | 1996 | 1997 | 1998 | 1999 | 2000 | 2001 | 2002 | 2003 | 2004 | 2005 | 2006 |
|-----------------------------------|------|------|------|------|------|------|------|------|------|------|------|------|
| Per production type               |      |      |      |      |      |      |      |      |      |      |      |      |
| Hatcheries                        | 71   | 169  | 162  | 30   | 27   | 45   | 30   | 32   | 54   | 51   | 125  | 104  |
| On-growing farms                  | 207  | 340  | 346  | 478  | 527  | 447  | 508  | 414  | 429  | 303  | 280  | 276  |
| Brood stock farms                 |      |      |      | 2    | 3    | 7    | 7    | 14   | 2    | 9    | 14   | 15   |
| Per species                       |      |      |      |      |      |      |      |      |      |      |      |      |
| Farms with Atlantic salmon        | 225  | 425  | 392  | 417  | 462  | 382  | 408  | 372  | 387  | 295  | 345  | 316  |
| Farms with rainbow trout          | 31   | 63   | 69   | 66   | 62   | 83   | 93   | 61   | 74   | 48   | 61   | 49   |
| Farms with brown trout            | 15   | 13   | 38   | 21   | 27   | 28   | 24   | 23   | 24   | 21   | 8    | 24   |
| Farms with char                   | 1    | 7    | 6    | 5    | 4    | 10   | 8    | 9    | 9    | 5    | 7    | 8    |
| Farms with turbot                 | 6    | 1    | 1    |      | 1    | 1    | 4    |      | 1    | 1    |      | 1    |
| Farms with sea trout              |      |      |      | 2    | 3    | 2    | 4    | 1    | 2    | 2    | 2    | 2    |
| Farms with brook trout            |      |      |      | 2    |      | 1    | 1    | 2    | 1    | 2    |      |      |
| Farms with relict Atlantic salmon |      |      |      | 1    |      |      |      |      |      | 1    |      |      |
| Total                             | 278  | 509  | 506  | 510  | 554  | 494  | 534  | 468  | 498  | 375  | 417  | 392  |

# Discussion

In 2006, 180 samples from 6 sites were rejected compared to 90 samples from 3 sites in 2005. This represents an improvement from 2004, when 450 samples from 15 sites were rejected. In 2004, temperatures exceeded 10 °C in the rejected samples, due to the use of unsuitable transport boxes. This was remedied in 2005, and subsequent rejections were mainly due to mail delivery failures.

The isolation of salmonid alpha virus in samples received for surveillance of VHS virus and IHN virus may represent a problem for the detection of the two rhabdoviruses. Presently, neutralising antibodies against salmonid alpha virus are not available and it is not known whether replication of salmonid alpha virus will inhibit replication of VHS virus in the BF-2 cells (10). Therefore, tissue homogenates of salmonid alpha virus-positive samples are always examined for VHS virus by RT-PCR to ensure the absence of this virus. Six of the sites where salmonid alpha virus was detected are located in the region of Hordaland/Sogn and Fjordane where PD is endemic. One site is located in Finnmark, where PD was first diagnosed in 2003. The number of PD outbreaks in Norway has increased yearly, and the disease is spreading along the coast. Thus it is likely that the problem of salmonid alpha virus interfering with the surveillance of VHS and IHN will persist.

#### Conclusion

No suspected or confirmed cases of VHS virus or IHN virus have been registered in Norwegian fish farms in 2006, based on the examinations carried out in the surveillance and control programme for VHS and IHN at the National Veterinary Institute.

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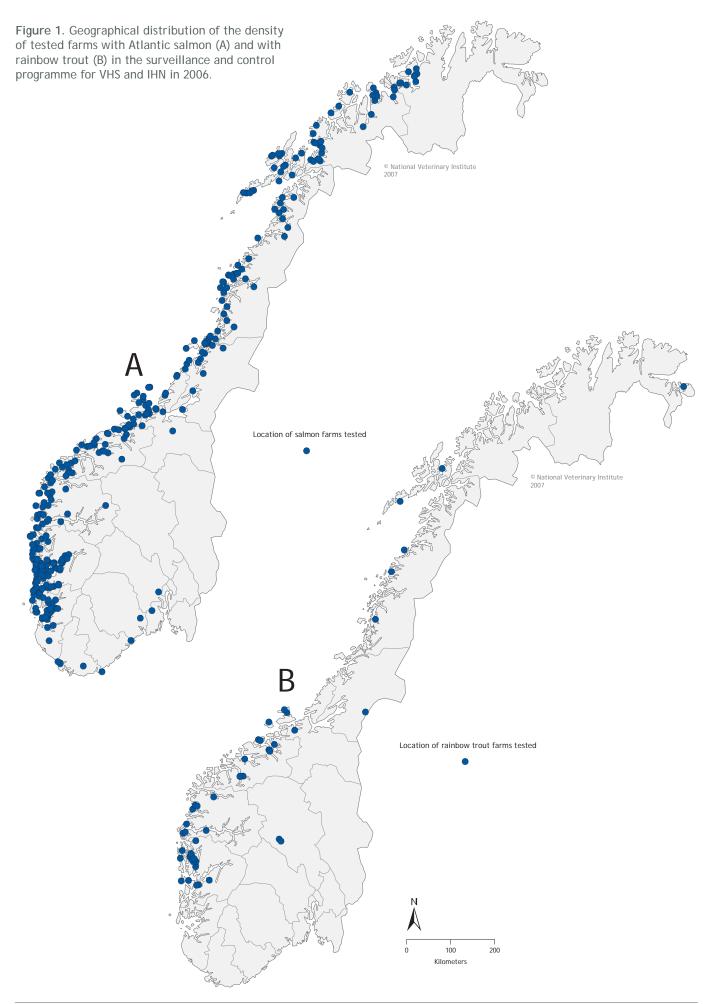
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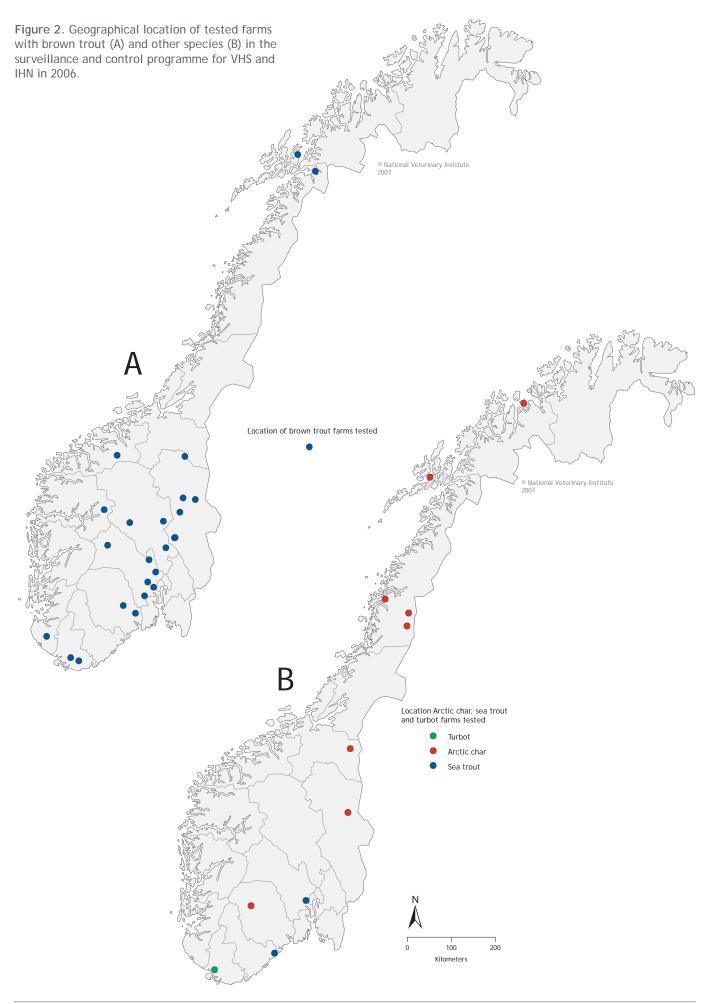
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The surveillance and control programme for *Gyrodactylus salaris* in Atlantic salmon and rainbow trout in Norway



Tor Atle Mo Kari Norheim Peder Andreas Jansen

# *In 2006,* Gyrodactylus salaris *was detected in two rivers. No commercial salmon farms were infected*

During the period of 1975 to 2006, Gyrodactylus salaris has been detected in Atlantic salmon fingerlings/parr from 46 rivers, 13 hatcheries/farms with Atlantic salmon parr/ smolts and 26 hatcheries/farms with rainbow trout (Oncorhynchus mykiss). The policy of the Norwegian Authorities is to eradicate G. salaris from infected rivers and farms. In farms, the procedure is to eliminate the hosts (salmon and rainbow trout). By doing so, the parasite is also eliminated because it does not have specialized free-living stages or intermediate hosts. In rivers, acidified aluminium sulphate is now the main chemical used to kill the parasite but not the fish host. By 31 December 2006, G. salaris was confirmed to be eradicated from 15 rivers and from all hatcheries/fish farms. The eradication has not been confirmed for nine additional rivers. The parasite is known to be present still in 22 additional rivers in Norway.

*G. salaris* is a notifiable (Group B) disease in Norway. It is listed as "Other significant disease" in the Office International des Epizooties (OIE). Surveillance of *G. salaris* has been performed in Norwegian salmon rivers since late 1970s (1, 2, 3, 4, 5). Surveillance is not performed in rivers or farms known to be infected unless measures for eradication of the parasite have just been carried out or other circumstances that justify the need for surveillance.

The Norwegian Food Safety Authority is responsible for sampling rivers and fish farms although County Environmental Departments and other institu-tions/companies are commissioned to do the actual sampling. The National Veterinary Institute in Oslo (the OIE reference laboratory for the disease) is responsible for examination of samples and taxonomical studies if *Gyrodactylus* is detected.

#### Aim

The surveillance programme aims to trace any spread of *Gyrodactylus salaris* to new river systems or fish farms (or to rivers and farms cleared of infection).

### Materials and methods

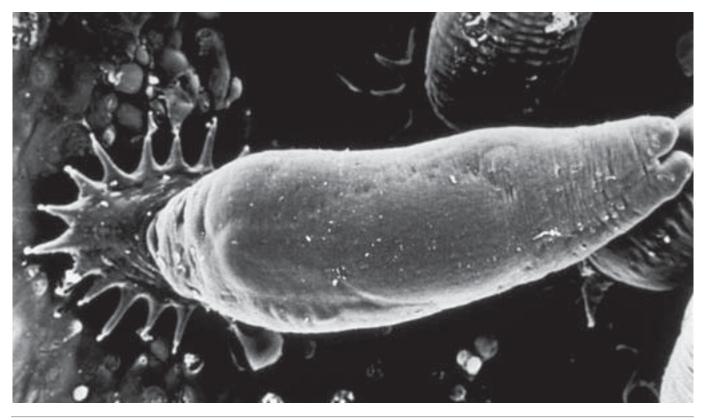
At least 30 Atlantic salmon are sampled from each farm and river. In rivers fingerlings/parr/smolts are caught by means of electrical fishing gear. In some of the large rivers, sampling is done at different dates and at different sampling stations. Farmed fish are caught by net. The fish are killed and preserved in 96 % ethanol. The samples are sent to the National Veterinary Institute in Harstad where body surface and fins are examined by a magnifying microscope (10 - 15 times magnification). However, only fins (except adipose fin) are sampled and preserved for examination from fish >15 cm.

#### **Results**

Altogether, 3,082 specimens from 94 rivers and 1,862 specimens from 57 farms were examined in 2006 (Tables 1 and 2). *G. salaris* was detected in two rivers but no farms were infected.

# Conclusion

*G. salaris* extended its range to river Ranelva while the river Hestdalselva had been rotenone treated in 2003 to eradicate the parasite.



Surveillance and control programmes in Norway · Gyrodactylus salaris · Annual report 2006

#### Table 1. Rivers examined for Gyrodactylus salaris in 2006

| County           | No. of rivers | Species         | No. of fish<br>examined | Detections     |
|------------------|---------------|-----------------|-------------------------|----------------|
| Finnmark         | 7             | Atlantic salmon | 310                     | 0              |
| Troms            | 7             | Atlantic salmon | 236                     | 0              |
| Nordland         | 16            | Atlantic salmon | 496                     | 2 <sup>1</sup> |
| Nord-Trøndelag   | 14            | Atlantic salmon | 423                     | 0              |
| Sør-Trøndelag    | 5             | Atlantic salmon | 170                     | 0              |
| Møre og Romsdal  | 15            | Atlantic salmon | 430                     | 0              |
| Sogn og Fjordane | 10            | Atlantic salmon | 302                     | 0              |
| Hordaland        | 6             | Atlantic salmon | 217                     | 0              |
| Rogaland         | 1             | Atlantic salmon | 32                      | 0              |
| Vest-Agder       | 2             | Atlantic salmon | 61                      | 0              |
| Aust-Agder       | 1             | Atlantic salmon | 31                      | 0              |
| Telemark         | 1             | Atlantic salmon | 32                      | 0              |
| Vestfold         | 2             | Atlantic salmon | 121                     | 0              |
| Buskerud         | 1             | Atlantic salmon | 30                      | 0              |
| Akershus         | 2             | Atlantic salmon | 65                      | 0              |
| Oslo             | 3             | Atlantic salmon | 96                      | 0              |
| Østfold          | 1             | Atlantic salmon | 30                      | 0              |
| Total            | 94            |                 | 3,082                   | 2              |

1 new river and 1 reappearance after rotenone treatment.

| County           | No. of farms | Species                        | No. of fish<br>examined | Detections |
|------------------|--------------|--------------------------------|-------------------------|------------|
| Troms            | 5            | Atlantic salmon                | 150                     | 0          |
| Nordland         | 9            | Atlantic salmon                | 270                     | 0          |
| Nord-Trøndelag   | 2            | Atlantic salmon, rainbow trout | 90                      | 0          |
| Sør-Trøndelag    | 4            | Atlantic salmon, rainbow trout | 150                     | 0          |
| Møre og Romsdal  | 10           | Atlantic salmon, rainbow trout | 330                     | 0          |
| Sogn og Fjordane | 8            | Atlantic salmon, rainbow trout | 272                     | 0          |
| Hordaland        | 11           | Atlantic salmon, rainbow trout | 360                     | 0          |
| Rogaland         | 5            | Atlantic salmon                | 150                     | 0          |
| Telemark         | 2            | Atlantic salmon                | 60                      | 0          |
| Buskerud         | 1            | Atlantic salmon                | 30                      | 0          |
| Total            | 57           |                                | 1,862                   | 0          |

#### Table 2. Fish farms examined for Gyrodactylus salaris in 2006

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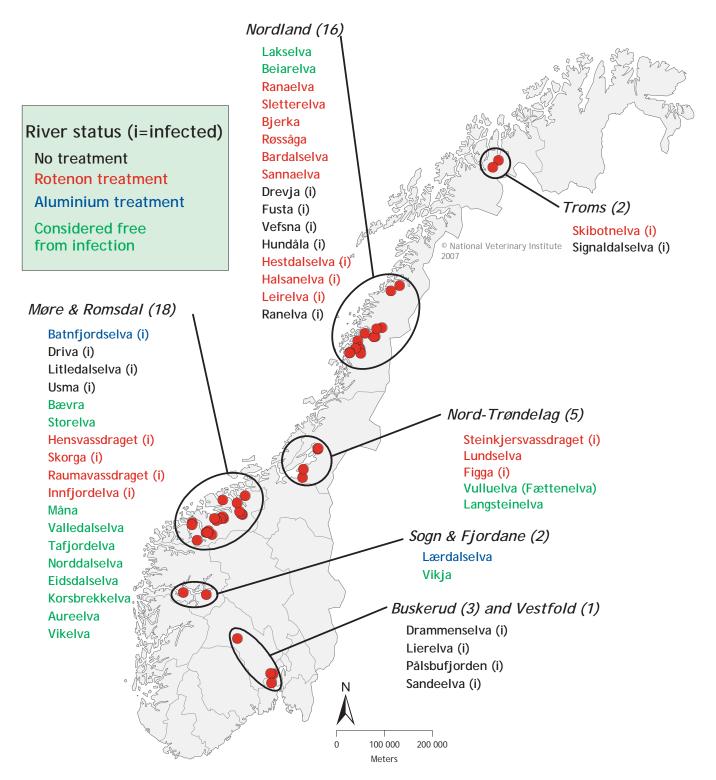
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# *Gyrodactylus salaris* detections 1975 - 2006



Annual report 2006

# The surveillance and control programme for bacterial kidney disease (BKD) in Norway

Ole Bendik Dale Tore Håstein Liv Jorun Reitan Randi Faller Torbjørn Endal Marianne Heum Terje Steinum



Responsible institutions National Veterinary Institute Norwegian Food Safety Authority

# Introduction

*No BKD positive salmon were detected by the surveillance programme during 2005 and 2006.* 

Bacterial kidney disease (BKD) is a chronic, serious disease of salmonid fish. The name of the causal agent is *Renibacterium salmoninarum* which means "the little kidney bacterium of salmonids". BKD was first reported in wild Atlantic salmon in Scotland during the 1930s, but the most serious problems have been seen in the USA and Canada. With the exception of Australia, BKD has been found in all salmonid rearing countries.

Renibacterium can be transmitted vertically from one generation to the next through an infection inside the eggs. Disinfection of the eggs will not kill the bacteria inside the egg, and thus the egg trade has probably been a major factor in the spread of BKD. The bacterium is well adapted to its host, and under good, natural conditions BKD does not seem to pose a serious threat to the fish. However, in densely reared farm fish severe mortality may occur. Fish with overt BKD have a generalised infection and obvious lesions in the form of whitish nodules, especially in the kidney. In apparently healthy fish the infection can be sequestered by the host response to minute foci in any organ, but most commonly in the kidney. Such infections can be difficult to detect as there are either very few bacteria present in samples, or even none if the tissue sampled does not include a sequestered infection site. In many ways the disease BKD is similar to tuberculosis in mammals, but Renibacterium is a completely different bacterium which poses no threat to warm-blooded animals.

In 1980 the first five cases of BKD were found in Norway (Figure 1). Three cases were in commercial stocks, while two cases were in feral stocks reared for mitigation purposes. No imports to any of these farms are known, and feral brood fish were the probable sources of infection. BKD has been found in fish from altogether 17 different rivers in Norway, but not south-east of the mountain range along the north-western part of Norway. The National Veterinary Institute has diagnosed altogether 369 outbreaks of BKD from 1980 to 2006 (Figure 1). In the farming industry the number of cases peaked in 1990 when 60 sea-water farms had disease outbreaks. Since then a steady decrease of outbreaks has been achieved, and in 2006 no BKD cases were found. BKD gave in general chronic disease with highly unpredictable courses. It has been noted that populations with BKD may suffer especially high losses due to Infectious salmon anemia (ISA).

As there are no satisfactory treatments or vaccines, good control of BKD in Norway has been achieved by an avoidance strategy. The most essential step is to keep brood stock free of the infection. In brood stocks with a high frequency of infection as in parts of the USA, all brood fish are tested individually to select the least infected part of the population for breeding. This selection is not fail proof. In Norwegian salmon farming, the fortunate situation is that screening and selection can been done on a population level as there are uninfected populations available. Systematic disease surveillance through the life-span of the domesticated populations will reveal BKD. If BKD is found in a commercial brood stock, the whole stock is replaced. The cost is minimal if the infected stock can be Table 1. Brood fish screening for BKD by ELISA 1992-96

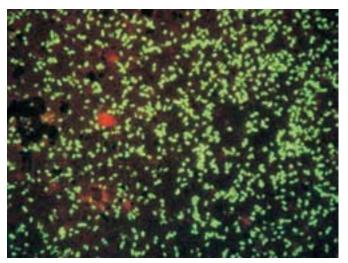
| Category   | Number screened | % pos (n) |  |
|------------|-----------------|-----------|--|
| Commercial | 1,041           | 0         |  |
| Feral*     | 4,048           | 0,15 (6)  |  |

\* 87 % Atlantic salmon, 12 % Rainbow trout, 1 % Arctic char.

slaughtered before disease or sexual maturation lowers the quality of the fish. A replacement brood stock can then be purchased in time for egg production. Only two commercial brood stocks have had to be culled since selection of BKD free stocks became a prime concern for the egg producers around 1990.

However, in order to have replacement stocks, a good overall disease control is required. Thus, smolts with BKD have not been allowed to be transferred to sea-water, and no movement of fish from seawater farms with BKD, other than for slaughter, has been allowed. Fallowing has been effective to curtail further BKD outbreaks in affected farms. The general hygienic standards that were introduced in the farming industry to stop ISA have also contributed significantly to the control of BKD.

In Norway feral stocks still represent a reservoir of infection which is difficult to eliminate. Fortunately the BKD prevalence in brood fish has been very low as shown by an earlier screening (Table 1). However, even this level has occasionally led to widespread infections in mitigation hatcheries. If fish from such hatcheries are released into the waterways, the BKD situation may deteriorate. To avoid this, screening individual, wild brood fish and checking for overt BKD in the hatchery before release is important in endemic areas. Preferably release of fish should be limited to the same watershed which the brood fish originated from. Thus, if low levels of infection are overlooked the disease will not be disseminated to other watersheds.



Immunofluorescense using a monoclonal antibody (4D3) on a kidney smear from a salmon with overt BKD.

Table 2. Results from BKD surveillance programme in Norway2005-2006

| County           | Sites | Pos | Neg   | Discarded* | Total |
|------------------|-------|-----|-------|------------|-------|
| Aust-Agder       | 1     | -   | -     | 30         | 30    |
| Vest-Agder       | 2     | 0   | 58    | 2          | 60    |
| Hordaland        | 5     | 0   | 274   | 0          | 274   |
| Sogn og Fjordane | 1     | 0   | 28    | 0          | 28    |
| Møre og Romsdal  | 4     | 0   | 240   | 0          | 240   |
| Sør-Trøndelag    | 11    | 0   | 368   | 4          | 372   |
| Nord-Trøndelag   | 12    | 0   | 329   | 1          | 360   |
| Nordland         | 18    | 0   | 590   | 40         | 630   |
| Total            | 54    | 0   | 1,887 | 77         | 1,994 |

\* Samples unfit for testing.

# Aim

The control and surveillance programme started in 2005 and is designed to provide documentation of the BKD situation in Norway in order to establish standards regarding import of live material to Norway.

# Materials and methods

The sampling is done by the Food Safety Authority, for the most part in conjunction with the sampling for VHS/IHN. The tissues sampled are predominantly from kidneys with the addition of the other internal organs from fingerlings. Extracts of the tissues are mostly tested individually by a commercially available (BiosChile) ELISA utilising monoclonal antibodies specific for a bacterial surface protein (MSA or p57) (1, 2, 3). This protein is regarded as an important virulence factor. Some pooling of samples from very small fish (maximum 5 fish) has been done. Positive and suspect samples in the ELISA are then tested for the presence of the gene coding for this protein by an in-house real-time PCR.

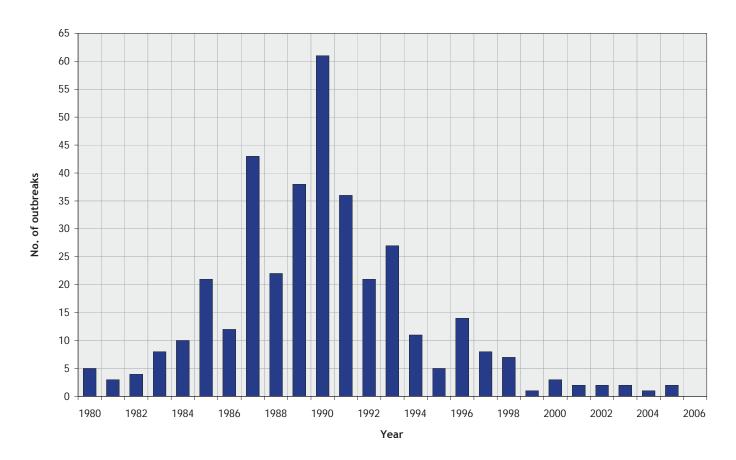


Figure 1. Number of BKD-outbreaks in Norway during the periode 1980-2006.

# Results

No BKD positive salmon were detected by the programme during 2005 and 2006 (Table 2).

This result is in keeping with the continuous disease diagnostics performed in the Norwegian aquaculture.

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Annual report 2006

The surveillance and control programme for bonamiosis and marteiliosis in European flat oysters (*Ostrea edulis* L.) in Norway

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Responsible institutions National Veterinary Institute Norwegian Food Safety Authority

# Introduction

# Bonamia *sp. or* Marteilia refringens *were not observed in any of the samples tested in surveillance 2006.*

Notifiable diseases of European flat oyster (Ostrea edulis L.) population in Norwegian waters (1, 2) have not been reported so far, including year 2006. This is in contrast to the situation in most other oyster producing European countries, where infectious diseases cause great losses in previously highly productive flat oyster populations (3). The protozoan parasites Bonamia ostreae and Marteilia refringens are identified as the main disease-causing organisms (4, 5) and bonamiosis has caused a collapse in flat oyster production in affected regions. B. ostreae has been detected as far north as Denmark. Bonamiosis was diagnosed in Limfjorden in 1980 but appears to have been successfully eradicated. In June 2006, B. ostreae was detected for the first time in Scotland and Wales. This led to the formation of two new surveillance zones. Bonamiosis and marteiliosis are classified as notifiable diseases by the OIE and as group A diseases in Norway.

In 2004 the entire coastline of Norway was recognized as an approved zone with regard to Bonamia ostreae and Marteilia refringens (6). The decision is based on the results of the surveillance and control programme for bonamiosis and marteiliosis which was initiated in the fall of 1995. The programme is based on directions given by the Commission Decision of November 6 2002 (7) referring to the OIE (International Office of Epizootics) "Manual of Diagnostic Tests for Aquatic Animals - 2003" (8), describing procedures for sampling and analysis of European flat oysters for bonamiosis and marteiliosis. The European flat oyster is found up to latitude 65 N in Norway, and wild populations are small and geographically limited due to climatic conditions. Since 1995, altogether 10 sites along the Norwegian coast have been included in the surveillance programme (Figure 1). However, not all sites have been included each year and selection of sampling sites has been based on the size of the wild populations and the structure of the oyster industry. In 2006, the sample sites were revised to ensure a more risk based approach. Two sites with no/very low activity were excluded, and a site with many transfers was included. In the spring of 2006, oysters were placed on this site. Analyses of this sample site were done for the first time in the autumn of 2006, and will be sampled both in autumn and in spring from 2007. Analysis of samples from this site will be prioritized. In 2006 a total of 6 sites were sampled.

The Norwegian Food Safety Authority is responsible for the programme, which involves inspection and sampling. The National Veterinary Institute in Bergen is responsible for laboratory procedures and analysis in accordance with the EU Decision, and also prepares the reports.

# Aim

The aim of the programme is to document the absence of *Bonamia ostreae* and *Marteilia refringens* in Nor-wegian flat oysters and maintain Norway's approved zone status.

# Materials and methods

## Sampling

The sites are inspected, and 30 oysters are sampled per site during spring and autumn by the Food Safety Authority District Offices, or by persons appointed by the District Offices. Live oysters are shipped to the National Veterinary Institute in Bergen.

# Analysis

Oyster shipments arrive at the laboratory within 24 hours of sampling. The oysters are prepared for histological examination according to section 3.1 of the OIE "Manual of Diagnostic Tests for Aquatic Animals – 2003" (8). A cross section, containing gills, mantle, and digestive gland, is cut from the specimen and fixed in Davidson's fixative. Fixed samples are processed for histology, sectioned and stained with Haematoxylin-Eosin.

*Bonamia* sp. are normally found in the gills, mantle and in the connective tissue surrounding the digestive gland. *Marteilia refringens* is normally found in the epithelium of the digestive gland and tubula.

# Results

During 2006, the National Veterinary Institute in Bergen received a total of 270 oysters from six sites (Table 1, Figure 1). All samples were examined. *Bonamia* sp. or *Marteilia refringens* were not observed. No observations of abnormal mortality have been reported for 2006.

Table 1. Number of oysters per sample site tested forbonamiosis and marteiliosis in 2006

| Sample site* | Spring 2006 | Autumn 2006 | Total 2006 |
|--------------|-------------|-------------|------------|
| 2            | 30          | 30          | 60         |
| 3            | 30          | 30          | 60         |
| 6            | 30          | 30          | 60         |
| 7            | -           | -           | 0          |
| 9            | 30          | 30          | 60         |
| 10           | 0           | 30          | 30         |
| Total: 6     | 120         | 150         | 270        |

\* Sites no 1, 4, 5 and 8 are no longer included

# Discussion

The results from the initial two-year period provide support for freedom from bonamiosis and marteiliosis in the Norwegian flat oyster population (9). A sample size of 30 gives a 95 % probability for detecting a prevalence of at least 10 % in an infected population.

The present sampling programme covers the geographical area in which commercial production and harvesting is possible. Sampling is judged to be representative and the results from the continued surveillance support the findings that *Bonamia ostreae* and *Marteilia refringens* are not present in the Norwegian flat oyster population.

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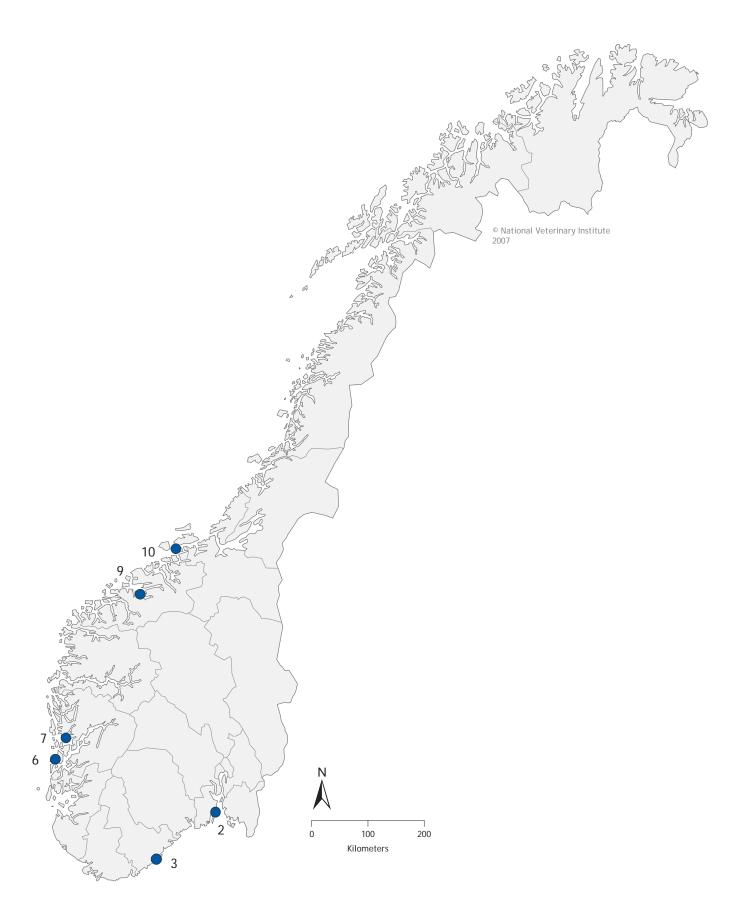


Figure 1. Geographical location of the sample sites in the surveillance and control programme for bonamiosis and marteiliosis in European flat oysters (*Ostrea edulis* L.) in 2006.



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