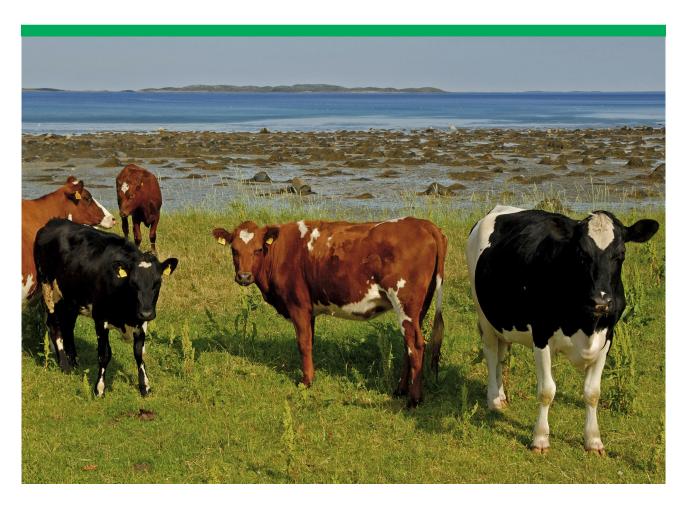
# The surveillance programme for paratuberculosis in Norway 2016







## The surveillance programme for paratuberculosis in Norway 2016

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#### **Authors**

Annette H Kampen, Lene Hermansen, Angelika Agdestein, Mette Valheim, Chiek Er

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#### Summary

In 2016, Mycobacterium avium subsp. paratuberculosis was not detected in Norway.

#### Introduction

Paratuberculosis in ruminants is a notifiable disease (List B) in Norway. *Mycobacterium avium* subsp. *paratuberculosis* infection was first diagnosed in cattle and goats in Norway in 1907 and 1934, respectively (1, 2). The control of this disease is enforced by government legislation and includes restrictions on animal movement. In goat herds, government restrictions combined with vaccination have previously been used to control paratuberculosis. However, the recent years a large proportion of the Norwegian goat herds has followed a disease eradication programme (3). In other ruminant species, *Mycobacterium avium* subsp. *paratuberculosis* was detected in sheep for the first time in Norway in 2002 and in alpaca in 2014 (4).

A national surveillance programme for paratuberculosis in cattle was established in 1996 (5, 6). The programme was extended to llamas and alpacas in 2000, goats in 2001, and sheep in 2002.

Descriptions of occurrence of the disease in Norway, control measures adopted until 1995, and results from the surveillance programmes from 1996 to 2001, can be found in the annual report for 2001 (6). The number of new infected herds detected since 1996 when the programme started is given in Figure 1.

#### Aim

The aim of the surveillance programme for paratuberculosis is to detect and control the infection in the Norwegian ruminant and camelid population.

#### Materials and methods

In 2016, cattle, goats, sheep, llamas and alpaca were included in the programme. Faecal samples were collected in farms and organs were collected at slaughterhouses by the Norwegian Food Safety Authority.

#### Active surveillance

Cattle

The target population consisted of all cattle herds delivering milk to dairies during the sampling period and all beef cattle herds receiving state support according to the state register in July 2015. Fifty dairy herds were selected for sampling by a risk-based strategy and fifty beef cattle herds were randomly selected. Faecal samples from the five oldest cows were collected.

#### Goats

Ninety herds were selected by a risk-based strategy for sampling from areas where paratuberculosis has previously been detected in goat herds, and thirty herds were selected from areas where paratuberculosis has not been registered. Faecal samples from the ten oldest goats were collected.

#### Sheep

Forty flocks from areas where paratuberculosis has been found in goat herds were randomly selected for sampling. Faecal samples from the ten oldest sheep were collected.

#### Llamas and alpacas

Llama and alpaca were introduced as new species to Norway in 1997-98. Since then, new animals are imported every year, and many originate from countries where paratuberculosis is endemic. Faecal samples from five animals older than four years of age are collected in each herd each year. If the herd has less than five animals, all animals are sampled. In addition, organ samples are collected at slaughter and from animals that die when older than four years.

#### Passive clinical surveillance

Clinical surveillance has been included in the programme since 2000. For cattle, samples are collected from animals with clinical signs like reduced milk production, weight loss, diarrhoea lasting more than 14 days, and animals that are older than three years of age.

#### Methods

Faecal samples - PCR

Faecal samples were investigated using a PCR technique. DNA extraction from faecal samples was performed on QIAcube using the QIAamp® DNA Mini Kit (Qiagen).

All samples were tested by real-time PCR using the ADIAVET ® REALTIME PARATB kit (bioMérieux). This test is based on gene amplification of the DNA segment IS900, specific for *Mycobacterium avium* subsp. *paratuberculosis*.

Faecal samples and organ samples - Bacteriological examination

Organ samples were analysed by bacterial culture. These samples were first decontaminated with 4% sodium hydroxide and 5% oxalic acid with 0.1% malachite green (7), and then 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. In case of faecal samples positive by PCR, further confirmation would have been performed by culture. However, as none of the samples from 2016 were positive by PCR, culturing of faecal samples was not performed in this period.

#### Organ samples - Histopathological examination

Histological examination of samples from jejunum, ileum, ileocecal valve, and mesenteric lymph nodes was performed. 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. Samples showing typical granulomatous lesions with acid-fast bacteria were considered positive for paratuberculosis.

#### **Results**

A total of 457 cattle, 1,113 goats, 350 sheep, and 686 camelids were examined in 2016. Details on type of samples and number of herds are described in Table 1.

Table 1. Number of samples collected for examination for Mycobacterium avium subsp. paratuberculosis in 2016.

| Species   | Type of sampling   | Number of animals (herds) |        |                |                |
|-----------|--------------------|---------------------------|--------|----------------|----------------|
|           |                    | Fecal                     | Organs | Total positive | Total analysed |
| Cattle    | Risk-based samples | 452 (91)                  |        | 0(0)           | 457 (92)       |
|           | Suspected cases    | 5 (1)                     |        |                |                |
| Goat      | Risk-based samples | 1 092 (116)               |        | 0 (0)          | 1 113 (118)    |
|           | Suspected cases    | 20 (1)                    | 1 (1)  |                |                |
| Sheep     | Random samples     | 328 (36)                  |        | 0 (0)          | 350 (40)       |
|           | Suspected cases    | 21 (3)                    | 1 (1)  |                |                |
| Camelides | Random samples     | 596 (187)                 | 12 (7) | 0 (0)          | (0) 686 (203)  |
|           | Suspected cases    | 71 (2)                    | 7 (7)  |                |                |

Since the surveillance programme for paratuberculosis started in 1996, *M. avium* subsp. *paratuberculosis* has been found in altogether 35 goat herds (some of these also positive before 1996), 11 cattle herds, 6 sheep flocks, and 2 alpaca herds (Figure 1). In the figure, only "newly infected" herds are presented. A herd that has previously been infected, can be counted as a new herd again if *M. avium* subsp. *paratuberculos* is found in a new ruminant species.

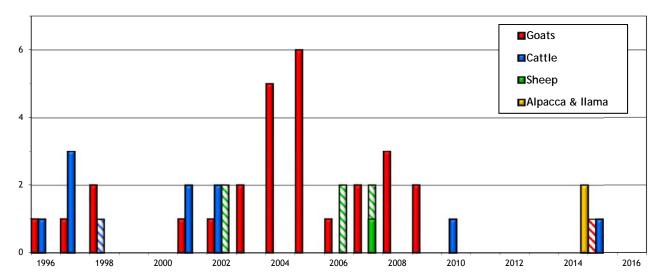


Figure 1. Number of new herds being reported positive for *Mycobacterium avium* subsp. *paratuberculosis* irrespective whether the samples were collected as part of the surveillance programme or not. Striped bars represents herds where another species in the same holding previously was reported positive.

#### Discussion

Paratuberculosis has been considered to occur among goats in six out of the 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 (three cattle herds, five sheep flocks). Importation of live cattle is limited and largely replaced by importation of semen and embryos.

The dairy organisation (TINE) and the Norwegian Goat Health Services have conducted an eradication programme named "Healthier goats", for caprine arthritis encephalitis, caseous lymphadenitis and paratuberculosis, three infectious diseases that were previously widespread in goats. The programme started in 2001 and initially concentrated on caprine arthritis encephalitis and caseous lymphadenitis. From 2004, paratuberculosis was added to the eradication programme to include herds in areas where paratuberculosis had been detected. In total 612 goat herds were included in the programme from 2001 to 2014 (3).

All goat milk herds in Norway have joined the eradication programme and have eradicated the three diseases. All known goat herds diagnosed with paratuberculosis have joined the eradication programme or have slaughtered their animals. Hence, Norway is in the unique position of having no known cases of paratuberculosis.

Even though the eradication programme has reduced the indigenous source of *M. avium* subsp. *paratuberculosis*, infected goat herds undetected so far may still represent a risk for spread of the infection to other ruminants in the coming years. Furthermore, the import of cattle, sheep, goats, llamas and alpacas presents a risk for new introduction of infected animals into the Norwegian ruminant population.

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www.vetinst.no

Bergen

post.vib@vetinst.no

Harstad

vih@vetinst.no

Tromsø

vitr@vetinst.no

Sandnes

vis@vetinst.no

Oslo

postmottak@vetinst.no

Trondheim

vit@vetinst.no

