

The surveillance and control programme
for *Echinococcus multilocularis* in
red foxes (*Vulpes vulpes*) in Norway.
Hunting season 2011-2012

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The surveillance and control programme for *Echinococcus multilocularis* in red foxes (*Vulpes vulpes*) in Norway. Hunting season 2011-2012

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Echinococcus multilocularis was not detected in any of the 614 red foxes (*Vulpes vulpes*) examined during the 2011-2012 licensed hunting season.

Introduction

Echinococcus multilocularis is endemic in large parts of the northern hemisphere, including eastern and central parts of Europe (1, 2). In 1999, *E. multilocularis* was detected in Denmark (3) and on the high-arctic Norwegian islands of Svalbard (4).

There was no evidence that this parasite had established in mainland Fennoscandia (5) prior to its detection in Sweden in February 2011 (6).

E. multilocularis has never been detected in mainland Norway, and anthelmintic treatment of imported dogs is compulsory to prevent introduction of the parasite through infected dogs. However, according to the EU Directive 998/2003/EC on pet movement, the maintenance of this national regulation post 2008 requires documentation of an *E. multilocularis* - free status within Norway.

Aim

The aim of the programme is to document freedom of *E. multilocularis* in mainland Norway.

Material and methods

Faecal samples collected from red foxes shot during the 2011-2012 licensed hunting season (from mid-July 2011 to mid-April 2012) were included in this year's program. All regions of Norway were represented in the sampling regime. Hunters were invited to participate based on the list of registered fox hunters. A standard form that included information on where and when the fox had been hunted, as well as the sex (male, female) and presumed age of the animal (juvenile, adult), was completed by each hunter.

The method used for the detection of *E. multilocularis* in the faecal samples was the newly developed DNA-fishing technique. This involves targeted DNA extraction from samples by applying specific DNA-hybridisation, followed by isolation using streptavidin coated magnetic beads (Mattson *et al*, in prep), then detection using a realtime PCR (Øines *et al*, in prep; Mattson *et al*, in prep). The new method is also capable of detecting DNA from adult worms, in addition to the eggs. These methods are aimed for use during the patent phase of the intestinal infection, more precisely when DNA from the eggs will be shed in the faeces. This period constitutes roughly two-thirds of the total infection period. The combination of the new methods were shown to be more sensitive than the previously used egg isolation using physical sieving followed by detection of parasite DNA using a multiplex PCR (7).

A total of 264 samples were run as pooled samples that consisted of 1 g faeces from each of 3 foxes, while the remaining 350 samples were run individually (3 g faeces per sample).

Although the diagnostic sensitivity between these two groups of samples will vary, a recent study showed that the new method used for the surveillance program (namely DNA-fishing and detection using realtime PCR) more than doubled the sensitivity compared the previous method used (egg isolation followed by multiplex PCR). In addition, the new and improved method was, contrary to the old sieving method, also able to detect DNA from other *E. multilocularis* parasitic stages, such as adult worms and proglotids.

The prevalence and corresponding confidence intervals were calculated in accordance with the EFSA harmonised schemes for the monitoring and reporting of *Echinococcus* in animals and

foodstuffs in the European Union (8) and we assumed a fox population of 70,000 (Olav Hjeljord, UMB, Ås, personal communication) and a test sensitivity of 63% for the last hunting season, and a sensitivity of 30% for the five last hunting seasons to account for the lower sensitivity of the test used until 2011.

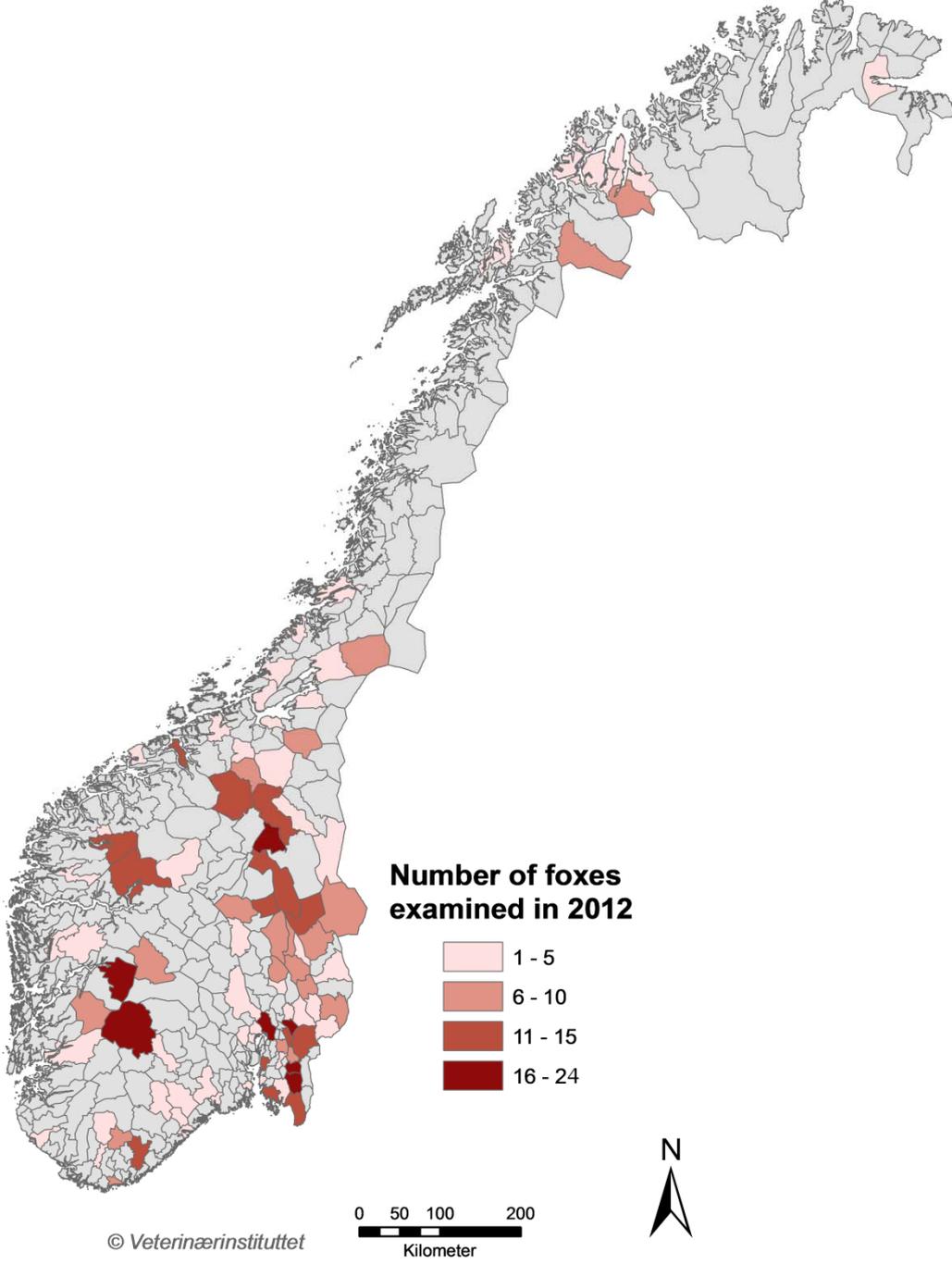
Results

A total of 671 fox samples were collected during the 2011-2012 hunting season, of which 614 were adequate for examination. All samples were negative for *E. multilocularis* giving an estimated prevalence of 0% (0 - 0.77%, 95% confidence interval). During the last five hunting seasons (from 2007-2008 to 2011-2012) a total of 1968 foxes have been examined. All foxes have tested negative giving an estimated prevalence of 0% (0 - 0.51%). In total, 2780 red fox faecal samples, from mainland Norway, have been tested for *E. multilocularis* between 2002 and 2012 (Table 1, Figure 1).

Table 1. Number and county of the red foxes sampled and examined for *Echinococcus multilocularis* in Norway during the red fox licensed hunting season from July to April, 2002-2012.

County	No. red foxes sampled		
	2002-2010	2011-2012	Total 2002-2012
Østfold	129	79	208
Akershus	223	86	309
Oslo	46	18	64
Hedmark	269	128	397
Oppland	183	33	216
Buskerud	90	12	102
Vestfold	47	2	49
Telemark	70	31	101
Aust-Agder	55	21	76
Vest-Agder	43	17	60
Rogaland	62	8	70
Hordaland	96	34	130
Sogn og Fjordane	161	32	193
Møre og Romsdal	82	16	98
Sør-Trøndelag	224	50	274
Nord-Trøndelag	111	18	129
Nordland	115	0	115
Troms	89	27	116
Finmark	71	2	73
Total	2166	614	2780

Figure 1. Map of Norway showing numbers and hunting municipality of red foxes sampled and examined for *Echinococcus multilocularis* during the red fox licensed hunting periods from July to April, 2002-2012.



Discussion

The 2011/2012 result is in agreement with the results from previous years with no positive samples detected. The cumulative sample size during the last five years is sufficient to confirm that the prevalence is less than 1%. This means that Norway fulfills the criteria, as given by EFSA (8), to document that *E. multilocularis* infection is absent from the national fox population. However, the criteria set by EFSA allow for samples to be collected over a five year period without taking into account the probability of introduction during the same period. Wahlström et al (5) showed that, even when taking into consideration the probability of introduction of infection, the number of samples collected until 2009 in Norway was sufficient to document that the prevalence was lower than 1%.

The detection of *E. multilocularis* in Sweden in 2011 and recently also in a new region in Denmark (9) have increased the risk of introduction of the parasite to Norway. As a consequence, an annual surveillance programme is necessary to document a continuous disease free status. Our findings support the maintenance of the national regulation for compulsory anthelmintic treatment of imported dogs to minimise the risk of *E. multilocularis* introduction to Norway.

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