

The occurrence and prevalence of potentially zoonotic enteropathogens in semi-domesticated reindeer

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Abstract: The information about pathogens excreted by semi-domesticated reindeer (*Rangifer tarandus tarandus*) that might represent a health risk to humans and animals is insufficient. The objectives of this study are to find the occurrence and prevalence of important potentially enteropathogenic, zoonotic bacteria and parasites in reindeer. Faecal samples from clinically healthy, semi-domesticated reindeer ($n=2243$) from northern regions of Finland and Norway were examined for important potentially enteropathogenic bacteria (*Campylobacter* spp., *Enterococcus* spp., *Escherichia coli*, *Salmonella* spp. and *Yersinia* spp.) and parasites (*Cryptosporidium* spp.) following standard procedures. *Escherichia coli* were isolated in 2123 (94.7%), *Enterococcus* spp. in 2084 (92.9%), *Yersinia* spp. in 108 (4.8%) samples and *Campylobacter* sp., identified as *C. hyointestinalis*, in one sample only (0.04%). Neither *Salmonella* spp. nor *Cryptosporidium*-oocysts were detected. This study clearly shows that *E. coli* and *Enterococcus* spp. belong to the normal intestinal flora of healthy reindeer. However, only few of the isolated *E. coli*-strains possess genes encoding *stx1* (0.14%), *stx2* (0%), *eae* (0.52%) and *bly*_{HEC} (0.99%), detected by PCR, that have the ability to cause health problems in humans and also animals. The isolated *Yersinia* spp. were further analysed for virulence factors, but examinations revealed no pathogenic strains. The public health risk due to excretion of important enteropathogenic microorganisms from reindeer has to be considered very low at present but a putative epidemiological threat to human health might arise when herding conditions are changed towards intensification and crowding. This study was performed as part of the EU-project RENMAN (www.urova.fi/home/renman/).

Key words: *Campylobacter*, *Enterococcus*, *Escherichia coli*, *Rangifer tarandus*, *Salmonella*, *Yersinia*, zoonosis.

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Introduction

Free-ranging animals may serve as sentinels or reservoirs for diseases in domestic livestock and man. Transmission of infectious agents to man may occur through direct contacts to free-ranging animals including cervids (Fanning & Edwards, 1991), by contamination of the environment through faecal shedding (Aavitsland & Hofshagen, 1999) or by consumption of venison (Keene *et al.*, 1997). In contrast,

to domestic animals, however, the epidemiological situation in free-ranging animals and in their habitat is difficult to assess. Bacteria, such as *Campylobacter* spp., *Enterococcus* spp., *Escherichia coli*, *Salmonella* spp., *Yersinia* spp. and the parasites *Cryptosporidium* spp. are among the most important agents in causing zoonosis like enteric and other severe diseases and have been isolated from healthy and diseased domestic ruminants (De Rycke *et al.*, 1986; Munoz *et*

al., 1996; Busato *et al.*, 1998 and 1999; Tham *et al.*, 1999). In *E. coli*, the ability to cause severe illness in humans and animals is associated to the occurrence of several virulence factors like the production of shigatoxins. Therefore the presence of shigatoxin genes can be an allusion for the virulence of certain strains, also known as shigatoxin producing *E. coli* (STEC). Besides STEC, *Campylobacter* spp., *Salmonella* spp. and *Yersinia* spp. are a problem for meat production with a high infection risk for humans consuming contaminated products. In addition, these bacteria are known to cause illness especially in young animals. Thus, a certain importance of these pathogens as well in reindeer production may not be excluded. *Salmonella* spp. (Kuronen *et al.*, 1998) have been found associated to mortality in reindeer in Finland and Sweden. *Campylobacter* spp., that have not been associated to mortality in reindeer, may also occur (Kobayashi *et al.*, 1999; Lahti *et al.*, 2001). A new genotype of *Cryptosporidium* closely related to *Cryptosporidium* (*Cr.*) *serpentis*, *Cr. muris* and *Cr. andersoni*, was isolated from 3 out of 49 examined caribous in Canada (Siefker *et al.*, 2002). Regarding *Cryptosporidium* spp. in northern European reindeer, no data is available. Ruminants, including reindeer, may be a reservoir for all these pathogens and more knowledge is required to better protect man and animals against outbreaks of diseases caused by these infectious organisms. This is of special importance as corralling of reindeer for winter feeding is increasing, eventually highering the prevalence of infectious microorganisms and the incidence of infectious diseases in reindeer.

Material and methods

In the RENMAN project, 2243 faeces samples from healthy reindeer, adults and calves, of both genders were examined for the occurrence of *Campylobacter* spp., *Enterococcus* spp., *E. coli*, *Salmonella* spp., *Yersinia* spp., and in addition, for the occurrence of the parasites *Cryptosporidium* spp. The samples were taken in the course of one year (June 2001 - April 2002) from Finnish and Norwegian free-ranging and corralled reindeer herds, considering parameters such as degree of intensification of husbandry, location and season. Samples were taken off the ground or per rectum from slaughter animals, sent to Kiel, Germany, directly after collection and were kept frozen (-4 °C) until further processed within max. one week.

The examination for *Campylobacter* spp. was done by inoculating 1 g faecal material into 9 ml Preston broth (Oxoid, Wesel, Germany). After 24 h incubation in a microaerophilic atmosphere (5% oxygen, 10% carbondioxide, 3% hydrogen and 82% nitrogen) at 37 °C, a loopful of the enriched suspension was plated on Preston agar (Oxoid) and incubated for 48 h under the above described conditions. *Campylobacter*-like colonies were analysed by Gram-staining, catalase and oxidase tests, and further biochemical reactions (ApiCampy, bioMérieux, Nürtingen, Germany).

To detect *Enterococcus* spp., 1 g faecal material was diluted in 9 ml glucose-azide broth (Merck, Darmstadt, Germany) and incubated for 48 h at 37 °C. A loopful broth was then spread both on kanamycin-aesculin-azide agar (Merck) and Slanetz and Bartley agar (Oxoid). After 48 h at 37 °C suspicious colonies were Gram-stained and their biochemical reactions were analysed further by catalase and oxidase tests.

Escherichia coli was isolated by adding 1 g faeces to 9 ml Gram-negative broth (Difco, Becton and Dickinson, Franklin Lakes, USA). After 24 h of incubation at 37 °C a loopful of broth was then plated onto Endo-c agar (Merck) and incubated under the above mentioned conditions for 24 h. Typical metallic shiny colonies were subcultured on blood agar (Oxoid), incubated for 24 h at 37 °C and tested for their biochemical reactions with API 20E (bioMérieux). PCR was used to detect the occurrence of shigatoxin1 and 2 genes (*stx1*, *stx2*), the intimin gene (*eae*) and EHEC-hemolysin gene (*hly_{EHEC}*). Primers were developed with help of the European Molecular Biological Library database and the oligo 6.0 software (Molecular Biology Insight, Cascade, USA) and manufactured commercially (Invitrogen, Karlsruhe,

Table 1. Primers used in PCR to amplify specific fragments from *E. coli* *stx1*, *stx2*, *eae* and *hly_{EHEC}* genes

Primer	Oligonucleotide sequence (5'- 3')	Size of amplified product
stx1 a	TGT AAC TGG AAA GGT GGA GTA TAC A	210 bp
stx1 b	GCT ATT CTG AGT CAA CGA AAA ATA AC	
stx2 a	GTT TTT CTT CGG TAT CCT ATT CC	484 bp
stx2 b	GAT GCA TCT CTG GTC ATT GTA TTA	
eae a	ATT ACC ATC CAC ACA GAC GGT	397 bp
eae b	ACA GCG TGG TTG GAT CAA CCT	
hly a	ACG ATG TGG TTT ATT CTG GA	166 bp
hly b	CTT CAC GTC ACC ATA CAT AT	

Germany). Base sequences and determined sizes of amplified products are shown in Table 1.

A loopful of *E. coli* colonies was diluted in 1 ml of treated saline solution (0.85 %) and heated in a thermoblock for 15 min at 100 °C. The samples were inserted in an ultrasonic bath for 3 min at 190 W and 50-60 Hz and centrifuged at 9875 g for 30 s. Five μ l of the supernatant were added to one ready-to-go-bead (Amersham Pharmacia Biotech, Buckinghamshire, UK) dissolved in 18 μ l sterile double distilled water. *EHEC EDL 933* (*stx1,2* positive) was used as a positive control and *E. coli ATCC 11 229* (*stx1,2* negative) was included as negative control. The conditions for the PCR were 95 °C for 12 min 30 s for initial denaturation, followed by 35 cycles of 95 °C for 20 s (denaturation), 57 °C for 30 s (primer annealing) and 72 °C for 40 s (DNA synthesis) and 5 min of final extension at 72 °C performed with a thermal cycler (Perkin-Elmer, Norwalk, USA). The amplified products were analysed by electrophoresis in a 2% agarose gel and were visualised following ethidium bromide staining (100 μ l/100 ml gel) (Sigma-Aldrid, Steinheim, Germany) at UV-light and photographed (AlphaInnotech, Biozym, Hessisch Oldendorf, Germany) using the Alphamager 1220 software (Biozym).

For the selective enrichment of *Salmonella* spp. 1 g faeces was inoculated into 14 ml of tetrathionate broth (Merck) and incubated for 24 h at 37 °C. One ml of this enriched broth was brought into tetrathionate broth the next day and incubated for another 24 h at 37 °C. This enrichment step was repeated a further time. On the fourth day one loopful of the cultured medium was plated both on *Salmonella-Shigella* agar (Difco) and Leifson agar (Merck). After 24 h of incubation at 37 °C presumptive *Salmonella* spp.

colonies were Gram-stained and tested by API 20E (bioMérieux).

Cultural examination of *Yersinia* spp. was performed by adding 1 g faeces into 9 ml of Gram-negative broth and incubating for 48 h at 21 °C. One loopful of broth was then plated on *Yersinia*-selective agar (Difco) and incubated for another 48 h at 21 °C. Colonies with the typical bull's-eye-appearance were subcultured on blood agar and Gram-stained and biochemical tests were subsequently carried out with API 20E (bioMérieux) and Micronaut (Merlin, Bornheim-Hersel, Germany). To detect various *Yersinia*-genes, PCR was performed using the primers listed in Table 2. The PCR conditions consisted of an initial 94 °C denaturation step of 10 min followed by 30 cycles of 94 °C for 1 min, 57 °C for 1 min, and 72 °C for 1 min. The final cycle was followed by a 72 °C incubation for 10 min. Amplified DNA fragments were visualized as mentioned for the *E. coli* virulence genes.

For the detection of *Cryptosporidium* oocysts, immunomagnetic separation was applied using Dynabeads anti-*Cryptosporidium* (Dyna, Oslo, Norway). Twenty μ l of the immun concentrate were used for a direct immunofluorescence test (*Cryptosporidium*-Antigen-IFT, medac, Wedel, Germany) (das Graças C. Pereira *et al.*, 1999).

Cryptosporidium parvum oocysts from a calf (Iowa isolate, USA) served as the positive control. Using a fluorescence microscope at x400–x1000 magnification *Cryptosporidium* oocysts appear as 6-10 μ m in size, round or oval in shape with bright green fluorescence.

For statistical analyses, the data was evaluated with the Statistica 5.0 software (StatSoft GmbH, Hamburg, Germany), following the instructions of

Table 2. Primers used in the PCR to amplify *Yersinia*-genes.

Primer	Oligonucleotide sequence (5'-3')	Size of amplified product	Identified gen	Reference
SP1 SP3	GAA TAT TGC ACA ATG GGC GCA AAC AAA CCG CCT GCG TGC GC	233 bp	16S rRNA	(Neubauer <i>et al.</i> , 1999)
Adh2 Adh3	CAG GCG TTA ATT CTG TTG GTG TCC AAT GGC AAC AGA G	191 bp	yadA	(Blais & Phillippe, 1995)
V1 V2	CCT ACG AAC AAA ACC CAC AA GGA TTT ATC ATG GAT ATT TAT GG	524 bp	V-antigen	(Price <i>et al.</i> , 1989)
Ye1 Ye2	AAT ACC GCA TAA CGT CTT CG CTT CTT CTG CGA GTA ACG TC	330 bp	16S rRNA	(Neubauer <i>et al.</i> , 2000)

Trampisch & Windeler (1997). For all analyses, differences were considered significant at $P \leq 0.05$.

Results

In 2224 (99.2%) out of the total number of 2243 faecal samples one or more of the examined bacteria species were isolated. *Campylobacter* sp, identified as *C. hyointestinalis*, was detected in one sample only (0.04%). *Enterococcus* spp. were isolated in 2084 (92.9%) samples. *Escherichia coli* were isolated in 2123 (94.7%) samples. There was no evidence of the occurrence of *Salmonella* spp. nor *Cryptosporidium* spp.. Table 3 shows the results for the detection of the *E. coli* toxin genes by PCR.

Table 3. Occurrence of *E. coli* toxin genes in 2123 isolated strains (prevalences in parentheses)

	<i>stx1</i>	<i>stx2</i>	<i>eae</i>	<i>bly</i> _{EHEC}	<i>eae</i> + <i>bly</i> _{EHEC}
<i>E. coli</i> <i>n</i> =2123	3 (0.14%)	0	11 (0.52%)	21 (0.99%)	2 (0.09%)

One hundred and eight (4.8%) strains of *Yersinia* spp. were isolated, consisting of *Y. enterocolitica* bio-group 1A (29), *Y. intermedia* (2), *Y. kristensenii* (72), *Y. mollaretii* (3) and *Y. rhodei* (2). Fig. 1 illustrates the prevalences of *Yersinia* spp. and the other isolated bacteria in the examined reindeer faeces.

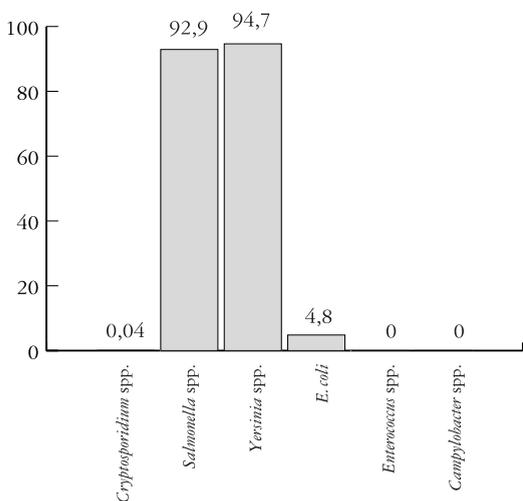


Fig. 1. Prevalences of analysed pathogens in faeces of reindeer ($n = 2243$)

Discussion

All bacteria investigated in this study may be found in northern Europe in the environment in aquatic, terrestrial and animal reservoirs (Kapperud, 1981) and were isolated from the intestinal tract of healthy or diseased ruminants worldwide (Adesiyun *et al.*, 1998; Busato *et al.*, 1998). Even though most of the isolated bacteria strains do not have the potential to cause severe human or animal health problems, certain strains might be a risk, especially for immunosuppressed, old or very young persons and animals. Therefore one has to regard the epidemiological impact of transmission of these infectious agents from the environment to reindeer and man and vice versa.

In reindeer, *Enterococcus* spp. and *E. coli* occurred in very high prevalences, showing the affiliation of these two species to the normal intestinal flora of healthy reindeer. Concerning *E. coli*, there are only few reports on diseases caused by shigatoxin-producing bacteria in ruminants (Sherwood, 1985; Mainil, 1999), however these bacteria are of extreme importance in causing severe diseases in humans (Griffin & Tauxe, 1991). As the genes encoding *stx1*, *eae* and *bly*_{EHEC} were detected only in very low numbers of the isolated *E. coli*-strains, the human health risk due to *E. coli* excreted by reindeer can be considered very low. These results comply with another study detecting no *E. coli* O157:H7 in 1387 faecal and 421 meat samples from reindeer (Lahti *et al.*, 2001). It is however known that STEC virulence factors are mobile within bacterial populations (Yamamoto *et al.*, 1984; Pupo *et al.*, 1997).

Yersinia spp. was isolated in 108 samples. The identified species *Y. intermedia*, *Y. kristensenii*, *Y. mollaretii* and *Y. rhodei* have been isolated before from various environmental samples (fresh water, soil, *etc.*), food, healthy animals and healthy and diseased humans (Baier & Puppel, 1981; Sulakvelidze, 2000). Even though these species are widely distributed in nature, their actual impact on human health is a matter of controversy. But as they are isolated from persons with gastrointestinal disorders, the role of these species should not be disregarded (Sulakvelidze, 2000). The isolated *Y. enterocolitica* strains belonged to Bio-group 1A, which embraces the nonpathogenic European *Y. enterocolitica* strains, often isolated from environmental samples, foods, animal and human faeces (Bottone, 1997).

Campylobacter hyointestinalis was isolated from one sample only. As the cultivation of *Campylobacter* spp. is exceedingly difficult, the real prevalence might be higher. Hitherto *Campylobacter hyointestinalis* has been associated only sporadically with human gastrointestinal disorders (Edmonds *et al.*, 1987; Gorkiewicz *et*

al., 2002). Even though the prevalence for *Campylobacter* spp. in this study was very low, it shows that reindeer can be carriers. This is approved by another study detecting *Campylobacter hyointestinalis* in a prevalence of 6% in Finnish reindeer faeces (Hänninen et al., 2002). It is surprising that neither *Salmonella* spp. nor *Cryptosporidium* oocysts were detected in reindeer in this study as both pathogens have been isolated from the environment, farm animals and man in Fennoscandia (Refsum et al., 2002; Horman et al., 2004), as was *Salmonella* spp. from reindeer in Finland as well (Kuronen et al., 1998).

Conclusion: The examined enteropathogens were either not detected at all (*Salmonella* spp. and *Cryptosporidium* spp.), in very small numbers (*Campylobacter* spp.) or if detected, their virulence and pathogenicity was very low (*E. coli* and *Yersinia* spp.). The potential human and animal health risk by reindeer, excreting various important enteropathogenic bacteria and *Cryptosporidium* spp., should be considered very low.

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References

- Aavitsland, P. & Hofshagen, T. 1999. *Salmonella* utbrudd i Herøy kommune. – *Nytt fra miljø og samfunnsmedisin* 3: 12.
- Adesiyun, A. A., Seepersadsingh, N., Inder, L. & Caesar, K. 1998. Some bacterial enteropathogens in wildlife and racing pigeons from Trinidad. – *J. Wildl. Dis.* 34: 73-80.
- Baier, R. & Puppel, H. 1981. Enteritis durch „atypische“ *Yersinien*. – *Dtsch. Med. Wschr.* 106: 208-210
- Blais, B. W. & Phillippe, L. M. 1995. Comparative analysis of *yadA* and *ail* polymerase chain reaction methods for virulent *Yersinia enterocolitica*. – *Food Control* 6: 211-214.
- Bottone, E. J. 1997. *Yersinia enterocolitica*: the charisma continues. – *Clin. Microbiol. Rev.* 10: 257-276.
- Busato, A., Lentze, T., Hofer, D., Burnens, A., Hentrich, B. & Gaillard, C. 1998. A case control study of potential enteric pathogens for calves raised in cow-calf herds. – *Zentralbl. Veterinarmed.* (B) 45: 519-528.
- Busato, A., Hofer, D., Lentze, T., Gaillard, C. & Burnens, A. 1999. Prevalence and infection risks of zoonotic enteropathogenic bacteria in Swiss cow-calf farms. – *Vet. Microbiol.* 69: 251-263.
- das Graças C. Pereira, M., Atwill, E. R., Jones, T. 1999. Comparison of Sensitivity of Immunofluorescent Microscopy to that of a Combination of Immunofluorescent Microscopy and Immunomagnetic Separation for Detection of *Cryptosporidium parvum* Oocysts in Adult Bovine Feces. – *Appl. Environ. Microbiol.* 65: 3236-3239.
- De Rycke, J., Bernard, S., Laporte, J., Naciri, M., Popoff, M. R. & Rodolakis, A. 1986. Prevalence of various enteropathogens in the feces of diarrheic and healthy calves. – *Ann. Rech. Vet.* 17: 159-168.
- Edmonds, P., Patton, C. M., Ward, G. E. & Griffin, P. M. 1987. *Campylobacter hyointestinalis* associated with human gastrointestinal disease in the United States. – *Clin. Microbiol.* 25: 251-263.
- Fanning, A. & Edwards, S. 1991. *Mycobacterium bovis* infection in human beings in contact with elk (*Cervus elaphus*) in Alberta. – *Canada Lancet* 228: 1253-1255.
- Gorkiewicz, G., Feierl, G. & Zechner, R. 2002. Transmission of *Campylobacter hyointestinalis* from a pig to human. – *J. Clin. Microbiol.* 40: 2601-2605.
- Griffin, P. M. & Tauxe, R. V. 1991. The epidemiology of infections caused by *Escherichia coli* O157:H7, other enterohemorrhagic *E. coli*, and the associated haemolytic uremic syndrome. – *Epidemiol. Rev.* 13: 60-98.
- Hänninen, M. L., Sarelli, L., Sukura, A., On, S. L. W., Harrington, C. S., Matero, P. & Hirvelä-Koski, V. 2002. *Campylobacter hyointestinalis* subsp. *hyointestinalis*, a common *Campylobacter* species in reindeer. – *J. Appl. Microbiol.* 92: 717-723.
- Hormann, A., Rimhanen-Finne, R., Maunula, L., von Bonsdorff, C., Torvela, N., Heikinheimo, A. & Hänninen, M. L. 2002. *Campylobacter* spp., *Giardia* spp., *Cryptosporidium* spp., noroviruses, and indicator organisms in surface water in southwestern Finland. – *Appl. Environ. Microbiol.* 70: 87-95.
- Kapperud, G. 1981. Survey on the reservoirs of *Yersinia enterocolitica* and *Yersinia enteocolitica*-like bacteria in Scandinavia. – *Acta. Pathol. Microbiol. Scand.* (B). 89: 29-35.
- Keene, W. E., Sazie, E., Kok, J., Rice, D. H., Hancock, D. D., Balan, V. K., Zhao, T. & Doyle, M. P. 1997. An outbreak of *Escherichia coli* O157:H7 infections traced to jerky made from deer meat. – *J. Am. Med. Ass.* 277: 1229-1231.
- Kobayashi, H., Pohjanvitra, T., Hirvelä-Koski, V. & Pelkonen, S. 1999. Prevalence and characteristics of *stx* and *eae* positive *Escherichia coli* in reindeer (*Rangifer tarandus*) and gulls (*Larus ridibundus* and *L. argentatus*). Poster. – In: *Pathogenicity and virulence. Verotoxi-genic E. coli in Europe. An EU Concerted Action Project*, Liège, Belgium. November 8 - 10, 1999.
- Kuronen, H., Hirvelä-Koski, V., Nylund, M. 1998. *Salmonella* poronvasoissa (Salmonellosis in reindeer). – *Poromies.* 4 - 5: 54 (in Finnish).
- Lahti, E., Hirvelä-Koski, V. & Honkanen-Buzalski, T. 2001. Occurrence of *Escherichia coli* O157 in reindeer (*Rangifer tarandus*). – *Vet. Rec.* 19: 633-634.

- Mainil, J. 1999. Shiga/Verocytotoxins and Shiga/Verotoxigenic *Escherichia coli* in animals. – *Vet. Res.* 30: 235-257.
- Munoz, M., Alvarez, M., Lanza, I. & Carmenes, P. 1996. Role of enteric pathogens in the aetiology of neonatal diarrhoea in lambs and goat kids in Spain. – *Epidemiol. Infect.* 117: 203-211.
- Neubauer, H., Reischl, U., Kostler, J., Aleksic, S., Finke, E.J. & Meyer, H. 1999. Variations in the 16S rRNA gene sequence of *Yersinia enterocolitica* isolates influence the specificity of molecular identification systems. – *Zentralbl. Bakteriol.* 289: 329-337.
- Neubauer, H., Hensel, A., Aleksic, S. & Meyer, H. 2000. Identification of *Yersinia enterocolitica* within the genus *Yersinia*. – *System. Appl. Microbiol.* 23: 58-62.
- Price, S. B., Leung, K. Y., Brave, S. S. & Straley, S. C. 1989. Molecular analysis of *lcrGVH*, the V antigen operon of *Yersinia pestis*. – *J. Bacteriol.* 171: 5646-5653.
- Pupo, G. M., Karaolis, D. K. R., Lan, R. & Reeves, P. R. 1997. Evolutionary relationships among pathogenic and nonpathogenic *Escherichia coli* strains inferred from multilocus enzyme electrophoresis and *mdh* sequence studies. – *Infect. Immun.* 65: 2685-2692.
- Refsum, T., Heir, E., Kapperud, G., Vardund, T. & Holstad, G. 2002. Molecular epidemiology of *Salmonella enterica* serovar *rypbimurium* isolates determined by pulsed-field gel electrophoresis: comparison of isolates from avian wildlife, domestic animals, and the environment in Norway. – *Appl. Environ. Microbiol.* 68:5600-5606.
- Siefker, C., Rickard, L. G., Pharr, G. T., Simmons, J. S. & O'Hara, T. M. 2002. Molecular characterisation of *Cryptosporidium* sp. isolated from Northern Alaskan caribou (*Rangifer tarandus*). – *J. Parasitol.* 88: 213-216.
- Sherwood, D., Snodgrass, D. R. & O'Brien, A. D. 1985. Shiga-like toxin production from *Escherichia coli* associated with calf diarrhea. – *Vet. Rec.* 116: 217.
- Sulakvelidze, A. 2000. *Yersinia* other than *Y. enterocolitica*, *Y. pseudotuberculosis*, and *Y. pestis*: the ignored species. – *Microbes Infect.* 2000: 497-513.
- Tham, W., Bannerman, E., Bille, J., Danielsson-Tham, M. L., Eld, K., Ericsson, H., Gavier-Widen, D., Rocourt, J. & Morner, T. 1999. *Listeria monocytogenes* subtypes associated with mortality among fallow deer (*Dama dama*). – *J. Zoo Wildl. Med.* 30: 545-549.
- Trampisch, H. J. & Windeler, J. 1997. *Medizinische Statistik*. Springer Verlag, Berlin: 133-147.
- Yamamoto, T., Honda, T., Miwatani, T. & Yokota, T. 1984. A virulence plasmid in *Escherichia coli* enterotoxigenic for humans: intergenetic transfer and expression. – *J. Infect. Dis.* 150: 688-698.

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Abstract in Norwegian / Sammendrag:

Det er mangelfull kunnskap om hvorvidt det i reinmøkk kan finnes mikroorganismer som kan representere en helserisiko for dyr og mennesker. Hensikten med denne studien var å undersøke forekomsten av mulige sykdomsfremkallende mikroorganismer i reinmøkk. Prøver av reinmøkk ble samlet fra 2243 klinisk friske tamrein i nordre deler av Finland og Norge. Prøvene ble undersøkt for bakteriene *Campylobacter* spp., *Enterococcus* spp., *Escherichia coli*, *Salmonella* spp., *Yersinia* spp. og parasitten *Cryptosporidium* spp. ved bruk av standardiserte laboratoriemetoder. *E. coli* ble funnet i 2123 prøver (94,7%), *Enterococcus* spp. i 2084

prøver (92,9%) og *Yersinia* spp. i 108 prøver (4,8%). *Campylobacter* spp., identifisert som *Campylobacter hyointestinalis*, ble bare funnet i én prøve (0,04%). *Salmonella* spp. og *Cryptosporidium* spp. ble ikke påvist. Videre undersøkelser av *E. coli* viste at bare svært få (<1%) av isolatene hadde gener som kodet for mulige sykdomsfremkallende toksiner. Videre undersøkelser av *Yersinia* spp. viste at ingen av disse isolatene var sykdomsfremkallende. Studien viser at helserisikoen knyttet til de undersøkte mikroorganismene i reinmøkk må betraktes som svært liten.