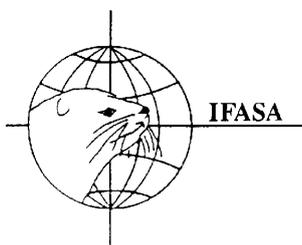
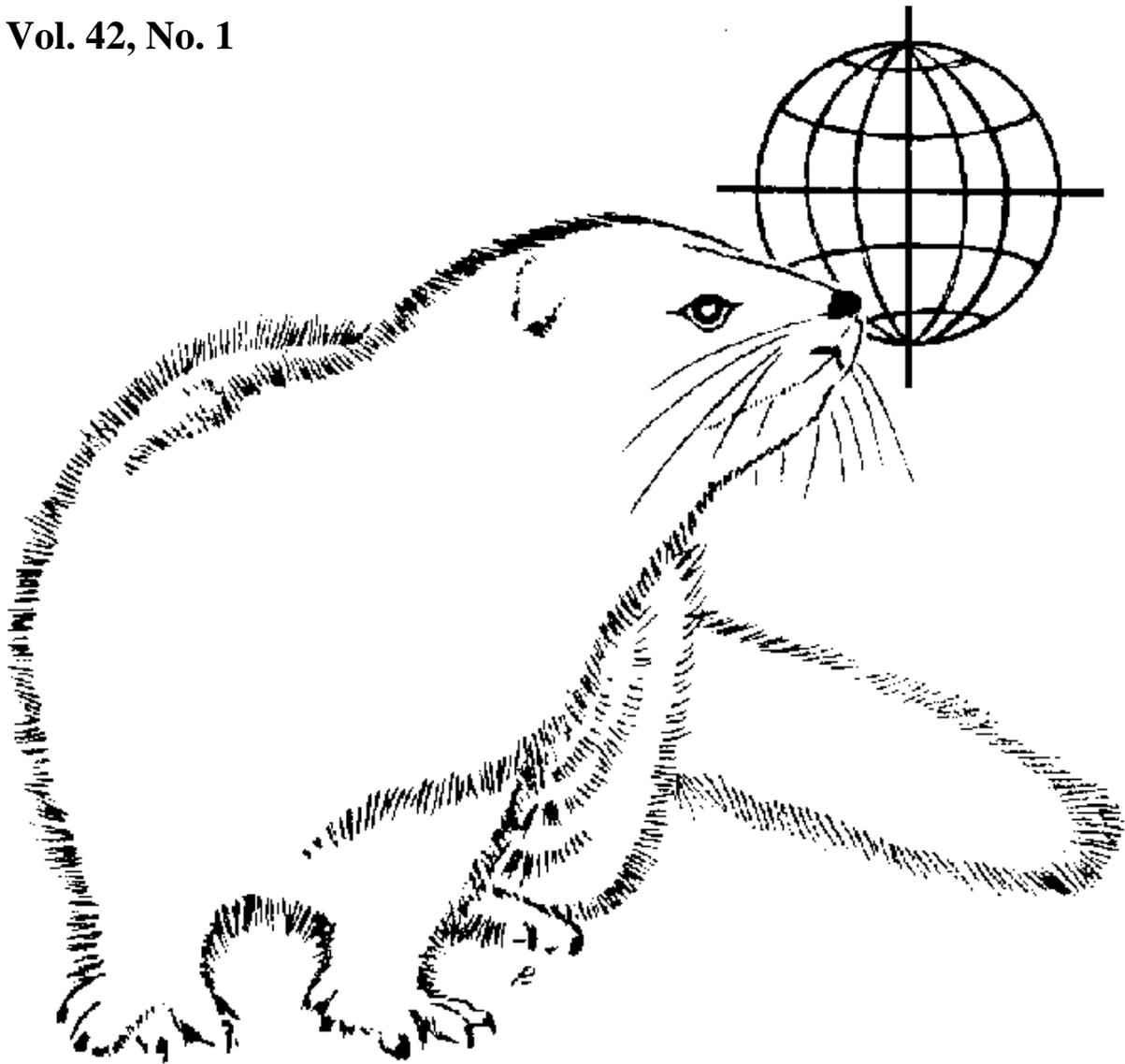


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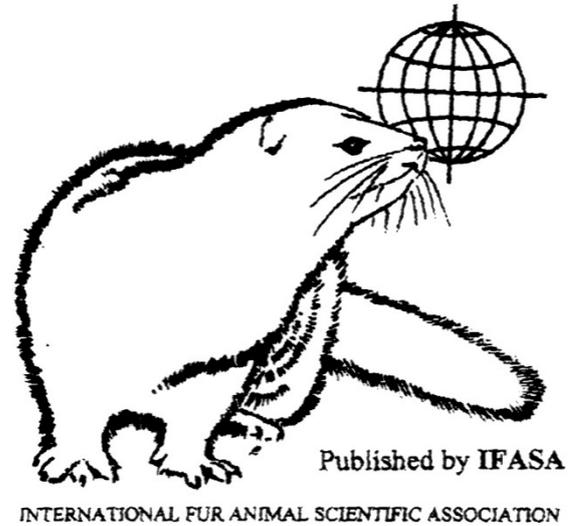
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Notes from the Editor

With the development of new, better and cheaper sequencing technologies, genome sequences have now been available from several species. This includes genome sequences for dog, cat and ferret. An abstract from the publication of the first draft mink genome is presented in this issue of *Scientifur*. The estimated size of the mink genome is 2.7 Gb which is similar to the size of the ferret genome. In total, 21,053 protein-coding genes were identified in the mink genome assembly. The draft reference genome allows for new genomic research in mink. This includes studies of the evolution in Carnivora. Importantly for the industry, it provides a basis for improved breeding for e.g. fur quality traits. Future generation of single nucleotide polymorphism (SNP) markers covering the whole genome makes genome-wide association studies (GWAS) feasible and by combining the linkage map, the location of fur quality genes and detected fur quality Quantitative Trait Loci (QTLs), genetic variants contributing to fur quality traits can be identified and used in breeding. In addition, genomic selection, which is widely used in other livestock species, is a potential new tool in mink breeding.

Two studies using gene expression to identify genes for fur quality traits are presented. The studies point at candidate genes for coat colour (black and white) in mink and for development of fur in rex rabbits.

Tracking Aleutian Mink Disease Virus (AMDV) is crucial for combatting the disease. A new phylogenetic study shows that while partial NS1 sequencing can distinguish virus strains belonging to major clusters, detailed information on farm to farm transmission requires full genome sequencing.

It is a pleasure to bring an abstract of a recent Finnish PhD-thesis: "Fur Animal Epidemic Necrotic Pyoderma – Pathology, Etiology and Epidemiology". The study deals with a severe disease named *Fur animal epidemic necrotic pyoderma* (FENP) detected in Finland in 2007 in mink, foxes and finnraccoon. The disease is ascribed to the use of seal byproducts in feed for mink. The study finds the bacterium *Arcanobacterium phocae* as a likely causative pathogen for FENP. The study was initiated by University of Helsinki (UH), the Finnish Fur Breeders' Association (FFBA), and the Finnish Food Safety Authority. The thesis was defended at Department of Veterinary Biosciences, Faculty of Veterinary Medicine, University of Helsinki, Finland.

Vivi Hunnicke Nielsen

Editor *Scientifur*

BREEDING, GENETICS AND REPRODUCTION

The first draft reference genome of the American mink (*Neovison vison*)

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The American mink (*Neovison vison*) is a semiaquatic species of mustelid native to North America. It's an important animal for the fur industry. Many efforts have been made to locate genes influencing fur quality and color, but this search has been impeded by the lack of a reference genome. Here we present the first draft genome of mink. In our study, two mink individuals were sequenced by Illumina sequencing with 797 Gb sequence generated. Assembly yielded 7,175 scaffolds with an N50 of 6.3 Mb and length of 2.4 Gb including gaps. Repeat sequences constitute around 31% of the genome, which is lower than for dog and cat genomes. The alignments of mink, ferret and dog genomes help to illustrate the chromosomes rearrangement. Gene annotation identified 21,053 protein-coding sequences present in mink genome. The reference genome's structure is consistent with the microsatellite-based genetic map. Mapping of well-studied genes known to be involved in coat quality and coat color, and previously located fur quality QTL provide new knowledge about putative candidate genes for fur traits. The draft genome shows great potential to facilitate genomic research towards improved breeding for high fur quality animals and strengthen our understanding on evolution of Carnivora.

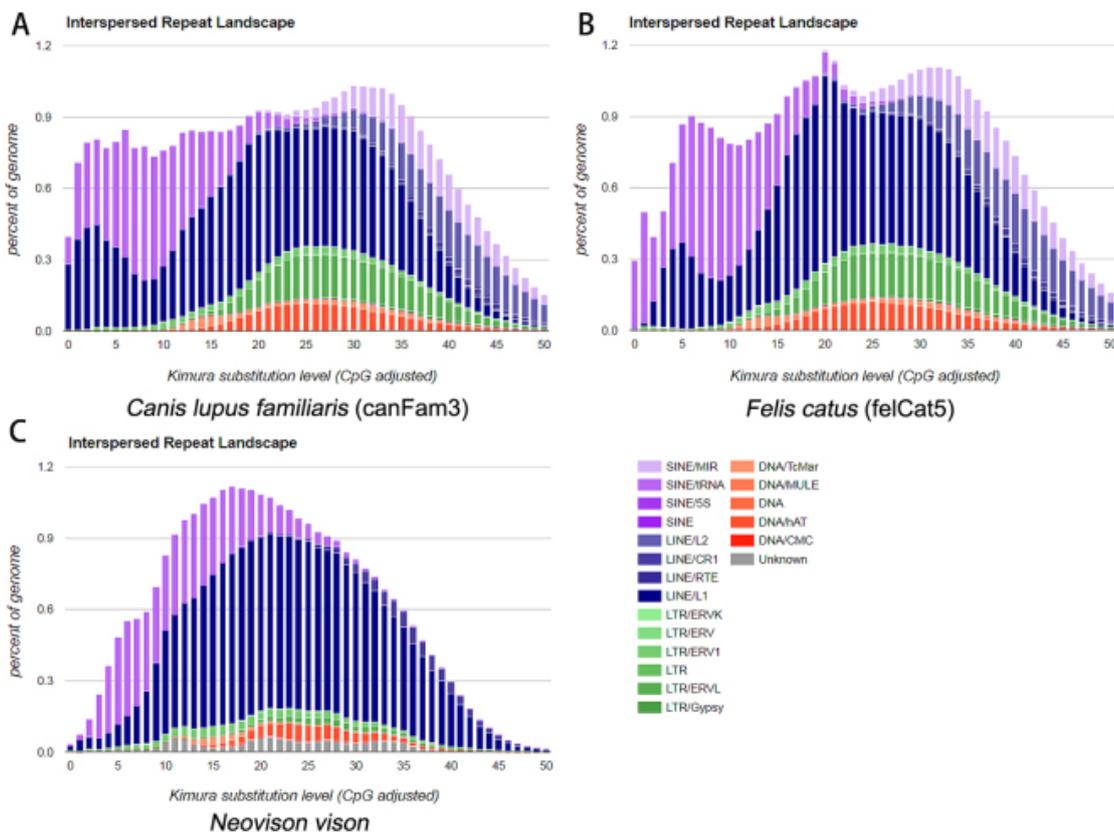


Fig. 1. Interspersed Repeat Landscape of the (A) dog, (B) cat and (C) mink genome.

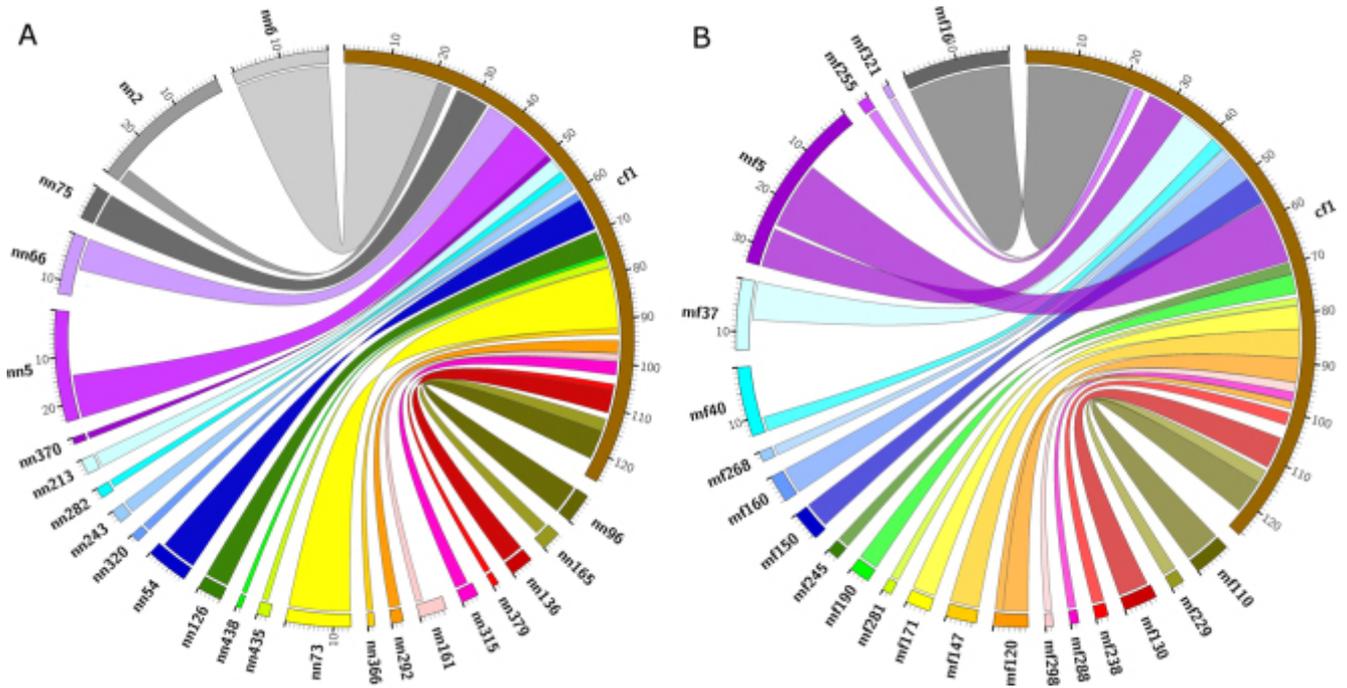


Fig. 2. Genome alignment of (A) mink genome and (B) ferret genome to dog chromosome 1 (cf1). The scaffold of mink start with 'nn' and ferret scaffold start with 'mf'. The first seven Mb of mink scaffold 66 (nn66) and ferret scaffold 37 (mf37) can be aligned to position 33 Mb to 40 Mb of dog chromosome 1 (cf1).

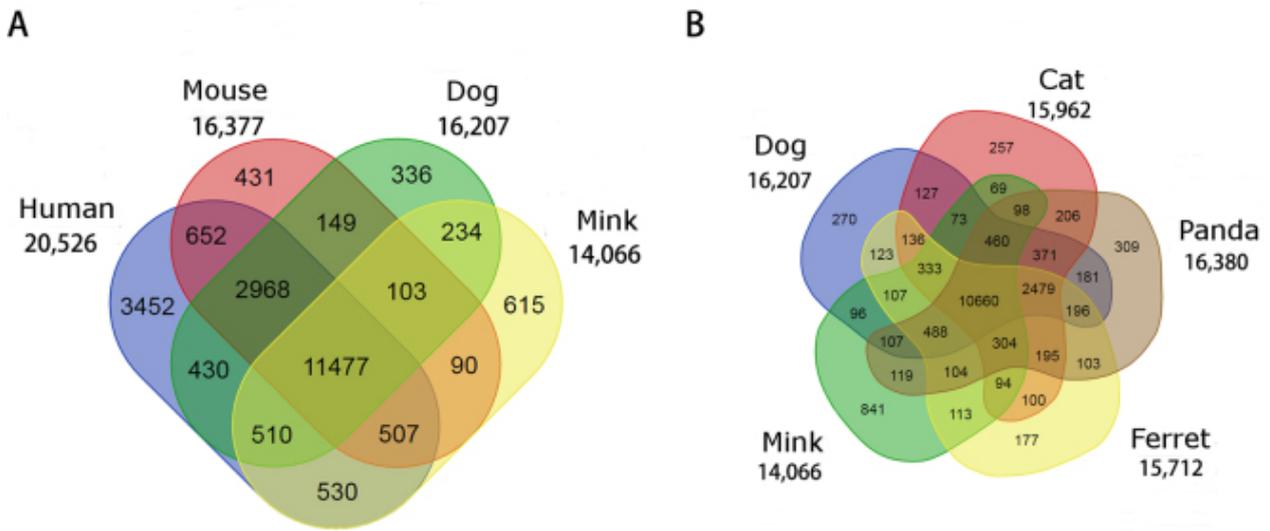


Fig. 3. Unique and shared gene families between (A) the human, mouse, dog and mink genomes; (B) dog, cat, panda, ferret and mink genomes. Numbers in Venn diagram represent the number of gene family and number under each species represent total number of gene.

Sci Rep. 2017 Nov 6; 7(1): 14564.
 Doi: 10.1038/s41598-017-15169-z.

Species Identification of Fox-, Mink-, Dog-, and Rabbit-Derived Ingredients by Multiplex PCR and Real-Time PCR Assay

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Various detection methods have been developed to date for identification of animal species. New techniques based on PCR approach have raised the hope of developing better identification methods, which can overcome the limitations of the existing methods. PCR-based methods used the mitochondrial DNA (mtDNA) as well as nuclear DNA sequences. In this study, by targeting nuclear DNA, multiplex PCR and real-time PCR methods were developed to assist with qualitative and quantitative analysis. The multiplex PCR was found to simultaneously and effectively distinguish four species (fox, dog, mink, and rabbit) ingredients by the different sizes of electrophoretic bands: 480, 317, 220, and 209 bp. Real-time fluorescent PCR's amplification profiles and standard curves showed good quantitative measurement responses and linearity, as indicated by good repeatability and coefficient of determination $R^2 > 0.99$. The quantitative results of quaternary DNA mixtures including mink, fox, dog, and rabbit DNA are in line with our expectations: R.D. (relative deviation) varied between 1.98 and 12.23% and R.S.D. (relative standard deviation) varied between 3.06 and 11.51%, both of which are well within the acceptance criterion of $\leq 25\%$. Combining the two methods is suitable for the rapid identification and accurate quantification of fox-, dog-, mink-, and rabbit-derived ingredients in the animal products.

Appl Biochem Biotechnol. 2017 Oct 25.
Doi: 10.1007/s12010-017-2621-2.

Comparative Transcriptome Analysis of Mink (*Neovison vison*) Skin Reveals the Key Genes Involved in the Melanogenesis of Black and White Coat Colour

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Farmed mink (*Neovison vison*) is one of the most important fur-bearing species worldwide, and coat colour is a crucial qualitative characteristic that contributes to the economic value of the fur. To identify additional genes that may play important roles in coat colour regulation, Illumina/Solexa high-throughput sequencing technology was used to catalogue the global gene expression profiles in mink skin with two different coat colours (black and white). RNA-seq analysis indicated that a total of 12,557 genes were differentially expressed in black versus white minks, with 3,530 genes up-regulated and 9,027 genes down-regulated in black minks. Significant differences were not observed in the expression of MC1R and TYR between the two different coat colours, and the expression of ASIP was not detected in the mink skin of either coat colour. The expression levels of KITLG, LEF1, DCT, TYRP1, PMEL, Myo5a, Rab27a and SLC7A11 were validated by qRT-PCR, and the results were consistent with RNA-seq analysis. This study provides several candidate genes that may be associated with the development of two coat colours in mink skin. These results will expand our understanding of the complex molecular mechanisms

underlying skin physiology and melanogenesis in mink and will provide a foundation for future studies.

Sci Rep. 2017 Sep 29; 7(1):12461. Doi: 10.1038/s41598-017-12754-0.



Fig. 1. Two coat colour phenotypes of mink representing 2 Chinese cultivated breeds, (a) BLM_S: Jinzhou black mink, black coat colour over the entire body with dark brown nose and black eyes. (b) WHM_S: Jilin white mink, white coat colour, where the red eyes can noticed, used in this study.

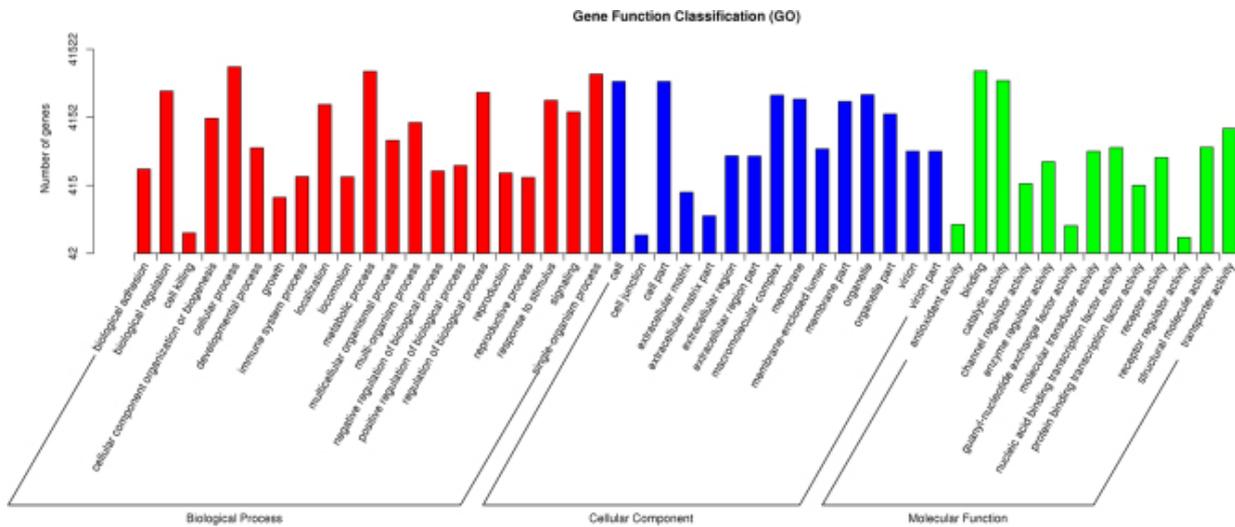


Fig. 2. Function classification of the assembled unigenes based on Gene Ontology (GO) categorization. The 41,522 unigenes were summarized in three main GO categories: biological processes (BP), cellular components (CC) and molecular functions (MF). The X axis indicates the next level of term in the three major categories of GO, and the Y axis indicates the number of unigenes annotated to the term.

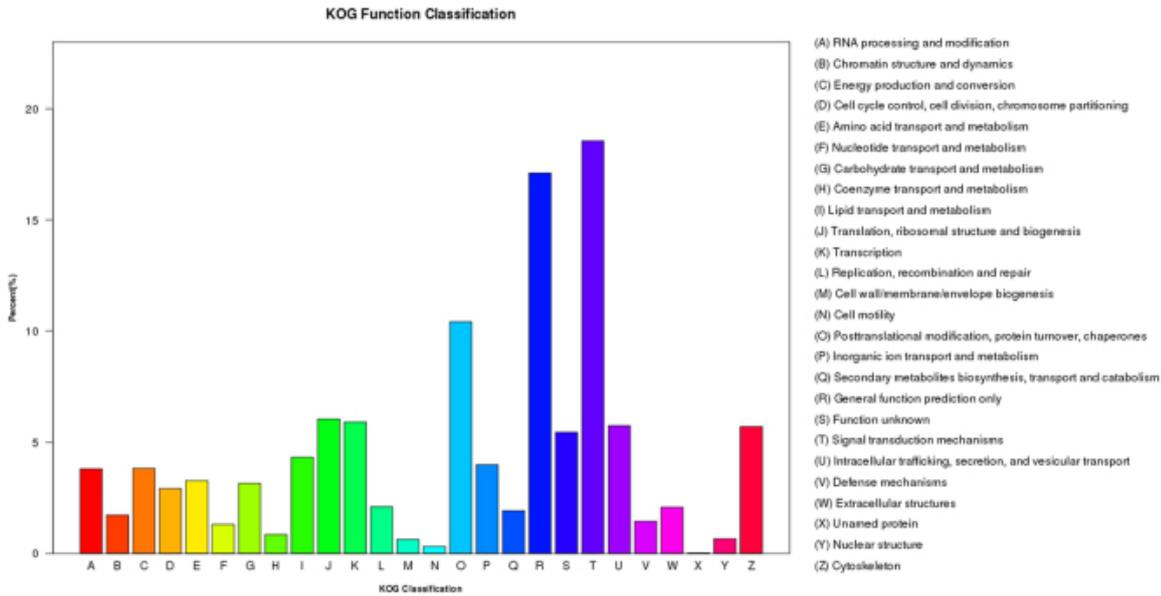


Fig. 3. Histogram of Clusters of eukaryotic Orthologous Groups (COG) classification. The 14,445 unigenes were aligned to the COG database to predict and classify possible functions. The X axis indicates the name of 26 group in COG categories, and the Y axis indicates the percentage of the total number of annotated genes.

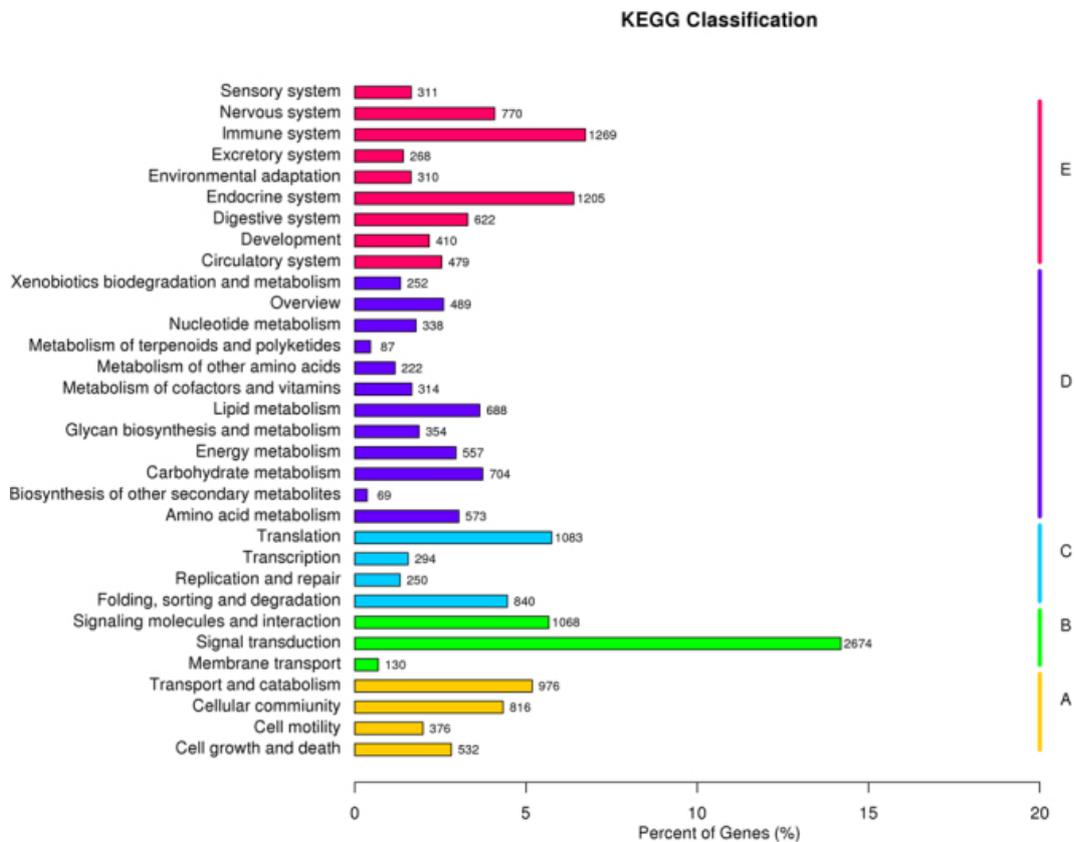


Fig. 4. Pathway assignment based on the Kyoto Encyclopedia of Genes and Genomes (KEGG) database. 19,330 unigenes were assigned into 5 KEGG biochemical pathways. (A), Cellular Processes (CP); (B), Environmental Information Processing (EIP); (C), Genetic Information Processing (GIP); (D), Metabolism (M); (E), Organismal Systems (OS).

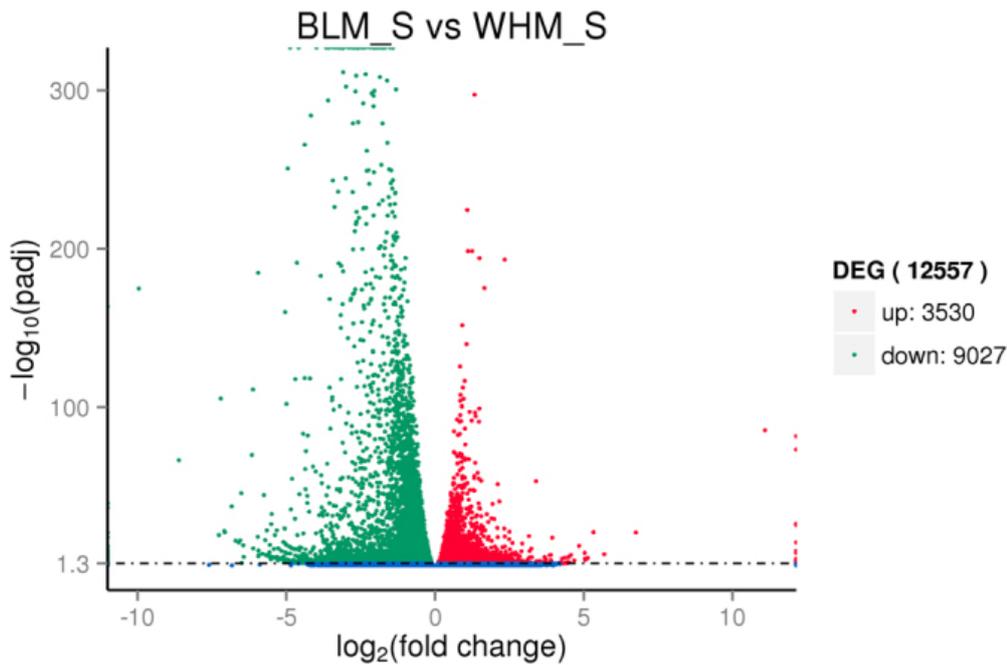


Fig. 5. Comparison of expression patterns of differential unigenes identified between two mink skins with black and white coat colour. The X axis indicates gene expression changes in different samples, and the Y axis indicates the significant degree of gene expression changes. Scattered points represent each gene, the red dots represent differentially up-regulated genes, the green dots represent differentially down-regulated genes, and the blue dots represent no significant difference gene. In total, 12,557 unigenes were identified as differentially expressed between skins with two coat colours, including 3,530 genes that were up-regulated and 9,027 down-regulated genes.

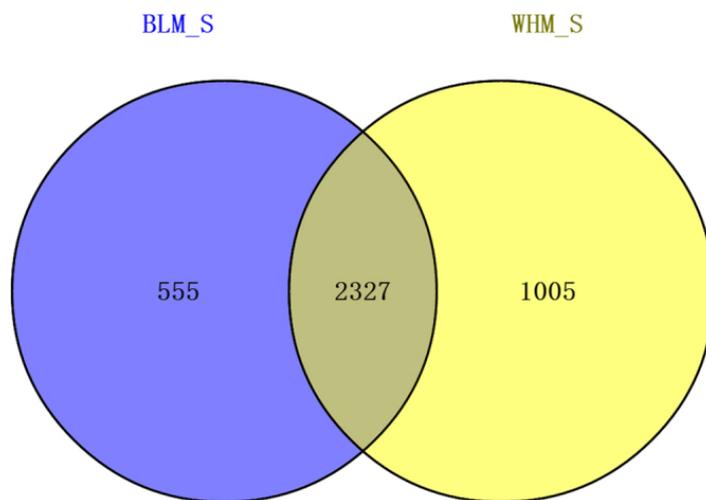


Fig. 6. The differentially expressed genes (DEGs) that are unique or shared between BLM_S and WHM_S. BLM_S refers to the black coat colour group, and WHM_S refers to the white coat colour group. The numbers in each section of the figure indicate the number of DEGs in the indicated comparison ($|\log_2\text{Foldchange}| > 1$ and $\text{padj} < 0.05$).

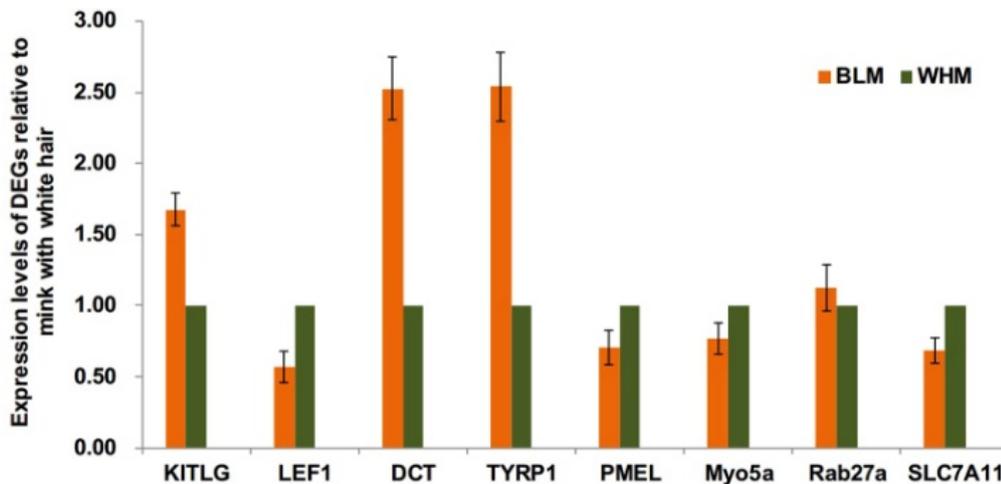


Fig. 7. Quantitative real time PCR (qRT-PCR) validation of DEGs in mink skin with two different coat colours. Abundance of target genes was normalized relative to abundance of β -actin gene. Bars in each panel represent the mean \pm standard (n = 3).

Characteristics of Structure and Nucleotide Polymorphism of the American Mink (*Neovison vison*) Growth Hormone Gene

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The nucleotide sequence, variation and gene-structure of the American mink (*Neovison vison*) growth hormone gene was analyzed. For this purpose set of 3 amplicons were sequenced, including 5'-UTR and all exons and introns. The study involved 389 animals. The sequencing results revealed the presence of 5 exons (10 bp, 161 bp, 117 bp, 162 bp, 201 bp) and 4 introns (245 bp, 171 bp, 176 bp, 290 bp), with a total length of 1745 bp. Fourteen polymorphic variable sites were identified: 12 SNP substitutions, one single nucleotide deletion and one ins/del polymorphism. The assessment of informativeness and conclusiveness of the identified variable sites shows that the highest ability to differentiate the American mink growth hormone gene genotypes has tri-allelic polymorphism g.1219C>G (PIC = 0.3039), and three diallelic polymorphisms - g.616G>C, g.846A>G and g.931C>T (PIC>0.2400). The recognized SNPs enrich the database of single nucleotide polymorphisms for American mink.

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Doi:10.17582/journal.pjz/2017.49.4.1293.1299

Y-Chromosome Markers for the Red Fox

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The de novo assembly of the red fox (*Vulpes vulpes*) genome has facilitated the development of genomic tools for the species. Efforts to identify the population history of red foxes in North America have previously been limited by a lack of information about the red fox Y-chromosome sequence. However, a megabase of red fox Y-chromosome sequence was recently identified over 2 scaffolds in the reference

genome. Here, these scaffolds were scanned for repeated motifs, revealing 194 likely microsatellites. Twenty-three of these loci were selected for primer development and, after testing, produced a panel of 11 novel markers that were analyzed alongside 2 markers previously developed for the red fox from dog Y-chromosome sequence. The markers were genotyped in 76 male red foxes from 4 populations: 7 foxes from Newfoundland (eastern Canada), 12 from Maryland (eastern United States), and 9 from the island of Great Britain, as well as 48 foxes of known North American origin maintained on an experimental farm in Novosibirsk, Russia. The full marker panel revealed 22 haplotypes among these red foxes, whereas the 2 previously known markers alone would have identified only 10 haplotypes. The haplotypes from the 4 populations clustered primarily by continent, but unidirectional gene flow from Great Britain and farm populations may influence haplotype diversity in the Maryland population. The development of new markers has increased the resolution at which red fox Y-chromosome diversity can be analyzed and provides insight into the contribution of males to red fox population diversity and patterns of phylogeography.

J Hered. 2017 Sep 1; 108(6):678-685.
Doi: 10.1093/jhered/esx066.

Gene expression profiling analysis reveals fur development in rex rabbits (*Oryctolagus cuniculus*)

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Fur is an important economic trait in rabbits. The identification of genes that influence fur development and knowledge regarding the actions of these genes provides useful tools for improving fur quality. However, the mechanism of fur development is unclear. To obtain candidate genes related to fur development, the transcriptomes of tissues from backs and bellies of Chinchilla rex rabbits were compared. Of the genes analyzed, 336 showed altered expression in the two groups (285 upregulated and 51 downregulated, $P \leq 0.05$, fold-change ≥ 2 or ≤ 0.5). Using GO and

KEGG to obtain gene classes that were differentially enriched, we found several genes to be involved in many important biological processes. In addition, we identified several signaling pathways involved in fur development, including the Wnt and MAPK signaling pathways, revealing mechanisms of skin and hair follicle development, and epidermal cell and keratinocytes differentiation. The obtained rabbit transcriptome and differentially expressed gene profiling data provided comprehensive gene expression information for SFRP2, FRZB, CACNG1, SLC25A4, and SLC16A3. To validate the RNA-seq data, the expression levels of eight differentially expressed genes involved in fur development were confirmed by qRT-PCR. The results of rabbit transcriptomic profiling provide a basis for understanding the molecular mechanisms of fur development.

Genome. 2017 Dec; 60(12):1060-1067.
Doi: 10.1139/gen-2017-0003.
Epub 2017 Aug 29.

NUTRITION, FEEDING AND MANAGEMENT

Use of polychlorinated biphenyl and toxic equivalent concentrations in scat from mink (*Neovison vison*) fed fish from the upper Hudson River to predict dietary and hepatic concentrations and health effects

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Environ Toxicol Chem. 2017 Oct 6.
Doi: 10.1002/etc.3997.

Serum Concentration of Macro-, Micro-, and Trace Elements in Silver Fox (*Vulpes vulpes*) and Their Interrelationships with Morphometric, Densitometric, and Mechanical Properties of the Mandible

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The optimal content of macro-, micro-, and trace elements in tissues ensures proper systemic growth and development and optimal health status in animals and humans. However, very little is known on the elemental content in the plasma compartment in Silver fox. The aim of this study was to determine the content of selected elements in serum obtained from 8-month-old female (N=8) and male (N=7) silver foxes. Moreover, relationships of the evaluated elements with the morphological, densitometric, and mechanical parameters of the mandible were determined. Serum content of 12 different elements was measured using inductively coupled plasma-atomic emission spectrometry. The morphometric and densitometric properties of the mandible were determined using quantitative computed tomography method, while mechanical endurance was tested using a three-point bending test. Serum concentration of calcium was significantly higher by 20% in male foxes (P = 0.01), while manganese concentration was significantly lower in males by over 17% (P = 0.03). Positive correlations of serum concentration of calcium, phosphorus, and magnesium with the morphological traits of the mandible such as weight, length, and bone volume were stated (P < 0.05). In the group of elements playing regulatory functions, the positive relationships between serum concentrations of selenium, chromium, manganese, copper, and cobalt were found (P < 0.05). The elaborated experimental model may serve for further studies on foxes, especially focused on nutritional factors affecting elemental homeostasis, whole-body metabolism, and systemic growth and development. Daily diet formulation and precise delivery for farm foxes, together with relatively large animal population maintained at the same environmental conditions, regularly subjected to slaughter procedure, enable economical experimentation with various dietary and pharmacological manipulations.

Biol Trace Elem Res. 2017 Dec 20.
Doi: 10.1007/s12011-017-1221-x.

BEHAVIOUR AND WELFARE

Mate choice screening in captive solitary carnivores: The role of male behavior and cues on mate preference and paternity in females of a model species, American mink (*Neovison vison*)

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Mate choice studies suggest that choosy females benefit from increased fecundity, litter size, and offspring survival. Thus, providing females with the opportunity to choose among potential mates, deemed genetically suitable based on studbook data, might improve breeding management in production and zoo animals and thereby the sustainability of captive populations. Investigating mate preference via odor from potential mates before animal transfer is a proposed strategy for incorporating mate choice into breeding management. In this study, we test whether olfactory cues and signals from males can be used to assess and measure female mate preference in American mink. Eighteen females were subjected to a 4-day stimulus test in which females showed a preference for one of two males' urine and feces. Subsequently, each female was subjected to a 10-day mate preference test involving the same two males of the first test. Paternity tests revealed that 13 females had offspring, which could be assigned to only one male, suggesting that these females performed a mate choice. In nine of these females preference during the stimulus test was directed toward the male that fathered their offspring. Our results suggest that even though there was a preference difference in scent stimulus trials from potential mates this preference was not predictive of eventual mate preference or paternity. Other factors such as aspects of male behavior seem to play a role, when the mates are introduced. Our study supports that mate preference and mate choice are complex matters influenced by multiple cues and signals.

Zoo Biol. 2017 Dec; 36(6):367-381.
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HEALTH AND DISEASE

The American mink (*Neovison vison*) is a competent host for native European parasites

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The American mink (*Neovison vison*) is a mustelid native to North America that was introduced in Europe and the former USSR for fur farming. Throughout the last century, accidental or deliberate escapes of mink from farms caused the establishment of stable feral populations. In fact, the American mink is considered an invasive alien species in 28 European countries. The present study evaluates the gastrointestinal and cardiopulmonary helminth fauna of the American mink in Galicia (NW Spain) to understand its role as a potential reservoir for parasites affecting other autochthonous mustelids. In the period 2008-2014, fifty American mink (35 males and 15 females) of different ages (22 immature and 28 adults) from the provinces of Lugo, Ourense and Pontevedra were captured and sacrificed. Eight parasite species were found (6 nematodes and 2 trematodes) with the following prevalences: *Molineus patens* (68%), *Aonchotheca putorii* (54%), *Crenosoma melesi* (10%), *Aonchotheca annulosa* (8%), *Angiostrongylus daskalovi* (6%), *Aelurostrongylus* spp. (2%), *Troglostrongylus acutum* (2%) and an unidentified trematode (2%). Eighty-two per cent of the mink harboured helminths, including 15 animals (30%) infected by only one parasite species, 19 (38%) by two species, 5 (10%) by three species and 2 mink (4%) by four species. All helminth species identified are native to European mustelids. Statistical models were used to evaluate if animal characteristics (age, sex and

weight), date and capture area influenced the prevalence, intensity or parasite richness. Statistical differences were detected only in models for intensity of *M. patens*, *A. putorii* and *C. melesi*. This is the first report of *Angiostrongylus daskalovi*, a cardiopulmonary nematode, and *A. annulosa*, a gastrointestinal nematode specific of rodents, in American mink. Moreover, although the fluke *T. acutum* has already been cited in American mink, to our knowledge, the present study represents the first report of this trematode in the lung.

Vet Parasitol. 2017 Nov 30; 247:93-99.
Doi: 10.1016/j.vetpar.2017.10.004.
Epub 2017 Oct 10.

Identification and characterization of a novel B-cell epitope on Aleutian Mink Disease Virus capsid protein VP2 using a monoclonal antibody

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Aleutian mink disease is caused by a highly contagious parvovirus (Aleutian mink disease virus, AMDV). This disease is one of the most commercially important infectious disease worldwide and causes considerable economic losses to mink farmers. The capsid protein VP2 is the major immunogenic antigenic protein of AMDV, and is involved in viral tropism, pathogenicity, and host selection. However, few reports have described the use of VP2-specific monoclonal antibodies (mAbs) in B-cell epitope identification and immunological detection. In this study, we produced a specific mAb, 1G5, against AMDV VP2 protein (amino acids:

200 ~ 588) and characterized its specificity and relative affinity. Six partially overlapping truncated recombinant proteins and seven synthesized peptides were used to identify the epitopes recognized by 1G5. The results indicate that mAb 1G5 can distinguish AMDV, MEV and CPV2 with high affinity ($K_a = 5.37 \times 10^9$), and the minimal linear epitope is located in amino acid residues ⁴⁵⁹EEEGWPAASGTHFED⁴⁷³. Sequence alignments demonstrated that the linear epitope was completely conserved among most Amdoparvoviruses except the bat parvovirus, where three substitutions (⁴⁶³W-⁴⁶³F, ⁴⁶⁶A-⁴⁶⁶G and ⁴⁷¹F-⁴⁷¹Y) were noted. Our results reveal that the identified epitope might be a common B-cell epitope of AMDV antibodies, and the 1G5 mAb can be used to identify the cleavage of the capsid proteins during AMDV infection. This is also the first report of a B-cell epitope on AMDV capsid protein VP2 (VP2: 459-473) using a mAb. These findings have potential applications in the development of new diagnostic tools for AMDV.

Virus Res. 2017 Dec 23. pii: S0168-1702(17) 30526-9.

Doi: 10.1016/j.virusres.2017.12.008.

Global phylogenetic analysis of contemporary Aleutian mink disease viruses (AMDVs)

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Background

Aleutian mink disease has major economic consequences on the mink farming industry worldwide, as it causes a disease that affects both the fur quality and

the health and welfare of the mink. The virus causing this disease is a single-stranded DNA virus of the genus Amdoparvovirus belonging to the family of Parvoviridae. In Denmark, infection with AMDV has largely been restricted to a region in the northern part of the country since 2001, affecting only 5% of the total Danish mink farms. However, in 2015 outbreaks of AMDV were diagnosed in all parts of the country. Initial analyses revealed that the outbreaks were caused by two different strains of AMDV that were significantly different from the circulating Danish strains. To track the source of these outbreaks, a major investigation of global AMDV strains was initiated.

Methods

Samples from 13 different countries were collected and partial NS1 gene was sequenced and subjected to phylogenetic analyses.

Results

The analyses revealed that AMDV exhibited substantial genetic diversity. No clear country wise clustering was evident, but exchange of viruses between countries was revealed. One of the Danish outbreaks was caused by a strain of AMDV that closely resembled a strain originating from Sweden. In contrast, we did not identify any potential source for the other and more widespread outbreak strain.

Conclusion

To the authors knowledge this is the first major global phylogenetic study of contemporary AMDV partial NS1 sequences. The study proved that partial NS1 sequencing can be used to distinguish virus strains belonging to major clusters. The partial NS1 sequencing can therefore be a helpful tool in combination with epidemiological data, in relation to outbreak tracking. However detailed information on farm to farm transmission requires full genome sequencing.

Virol J. 2017 Nov 22; 14(1):231.

Doi: 10.1186/s12985-017-0898-y.

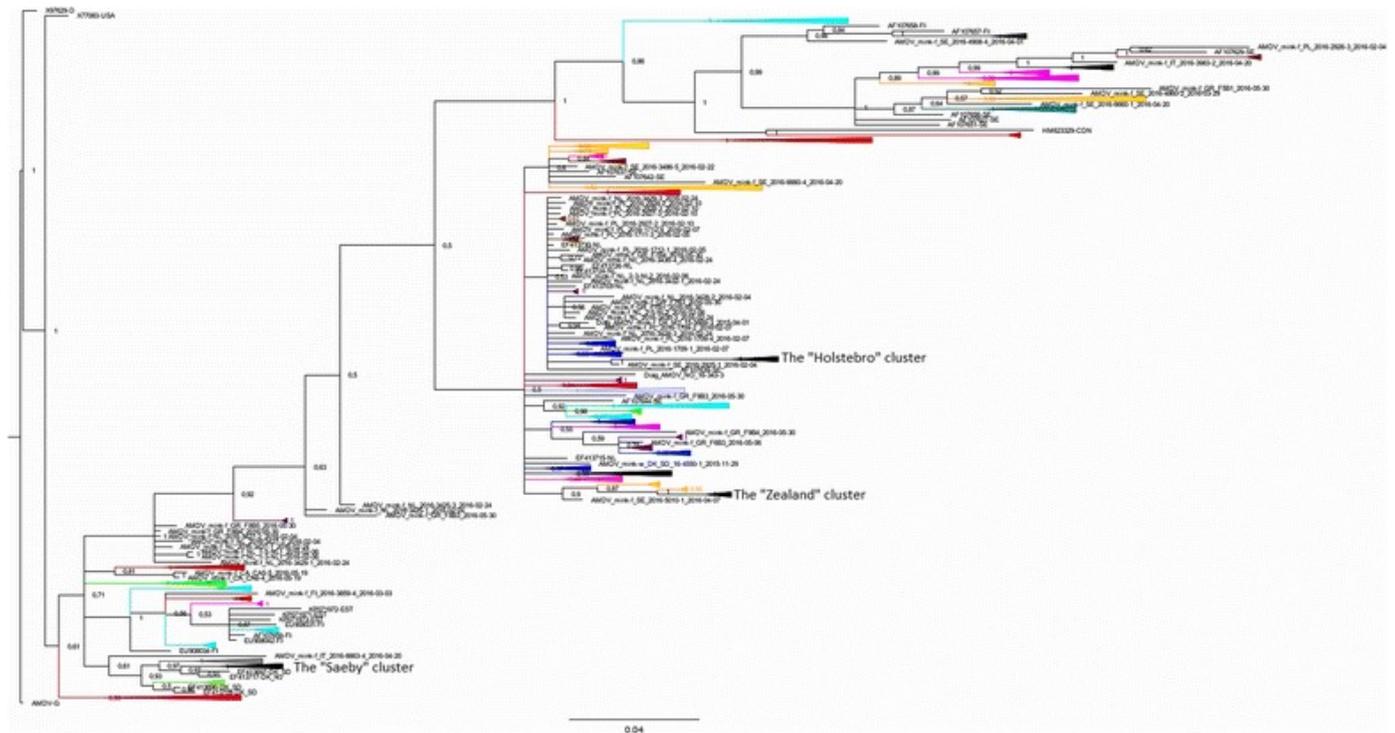


Fig. 1. Bayesian tree of the global distribution of partial NS1 sequences. Color coding of the tree given by the clusters: brown: Poland, dark green: Iceland, green: USA, turquoise: Finland, blue: The Netherlands, purple: Italy, yellow: Estonia, grey: Latvia, lavender: China, orange: Sweden, pink: Lithuania, red: Canada, dark grey: Spain, aubergine color: Greece and black: Denmark. Single sequences outside clusters are named as mentioned in the text: AMDV_mink-f/w_country code_unique identifier_sampling date. "F" or "w" indicates if the mink was farmed or wild. Samples from the routine diagnostics were named with prefix "Diag". Sequences from GenBank are named with their accession number and country code as suffix. A detailed tree can be found in the Additional file [1](#)

Comparative molecular analysis of strains of the Aleutian Disease Virus isolated from farmed and wild mink

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Introduction and objective

Aleutian Disease is a significant biological factor causing substantial losses in mink farming. The virus inducing the disease also infects wild populations which may constitute an asymptomatic reservoir. To compare genetic variants of the AMD virus occurring

in wild and farmed mink populations, an analysis was performed on a fragment of the VP2 protein sequence of the virus infecting both populations, taken from different living environments.

Material and methods

Genetic material was isolated from 11 farmed animals in which anti-AMDV antibodies had been detected and from 20 wild animals. The DNA obtained was amplified using primers specific for the fragment encoding the VP2 protein. The product obtained was sequenced and bioinformatic analysis was performed.

Results

Viral material was detected in 11 farmed and 7 free-living animals. Similarity of sequences averaged 99% within groups and 94% between groups. The sequencing results made it possible to identify characteristic changes for each group. In the isolates from the wild animals, the following changes were observed in the epitope region with respect to the reference sequence: C3704T, G3710A, T3722C, T3746C and A3749G. In the isolates from the farmed animals

a G3779A transition was noted. Phylogenetic analysis showed that the variants infecting the two groups occupy separate branches of the phylogenetic tree.

Conclusions

The variants of the virus infecting the two groups may have a common origin, but at present they constitute two separate groups, with characteristic differences making it possible to recognize their genotype.

Ann Agric Environ Med. 2017 Sep 21; 24(3):366-371.

Doi: 10.26444/aaem/75688.

Epub 2017 Jul 5.

Arcanobacterium phocae infection in mink (*Neovison vison*), seals (*Phoca vitulina*, *Halichoerus grypus*) and otters (*Lutra lutra*)

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Background

Infectious skin disorders are not uncommon in mink. Such disorders are important as they have a negative impact on animal health and welfare as well as on the quality and value of the fur. This study presents the isolation of *Arcanobacterium phocae* from mink with severe skin lesions and other pathological conditions, and from wild seals and otters.

Results

In 2015, *A. phocae* was isolated for the first time in Denmark from outbreaks of dermatitis in mink farms. The outbreaks affected at least 12 farms. Originating from these 12 farms, 23 animals cultured positive for *A. phocae*. The main clinical findings were necrotizing pododermatitis or dermatitis located to other body sites, such as the lumbar and cervical regions. *A. phocae* could be isolated from skin lesions and in nine animals also from liver, spleen and lung, indicating a systemic spread. The bacterium was also, for the first time in Denmark, detected in dead seals

(n = 9) (lungs, throat or wounds) and otters (n = 2) (throat and foot).

Conclusions

An infectious skin disorder in mink associated with *A. phocae* has started to occur in Danish farmed mink. The origin of the infection has not been identified and it is still not clear what the pathogenesis or the port of entry for *A. phocae* infections are.

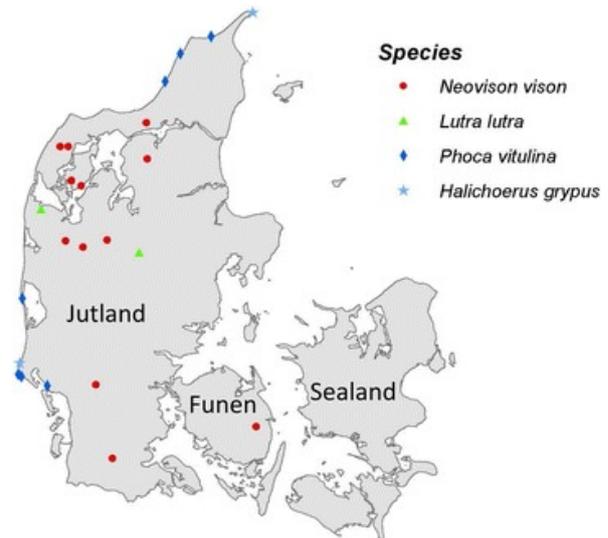


Fig. 1. Distribution of mink, seals and otters infected with *Arcanobacterium phocae*. The majority of infected mink farms are located in Jutland and one farm is at the island of Funen. All seals were located on the coast of Jutland and otters were found in the countryside in Jutland

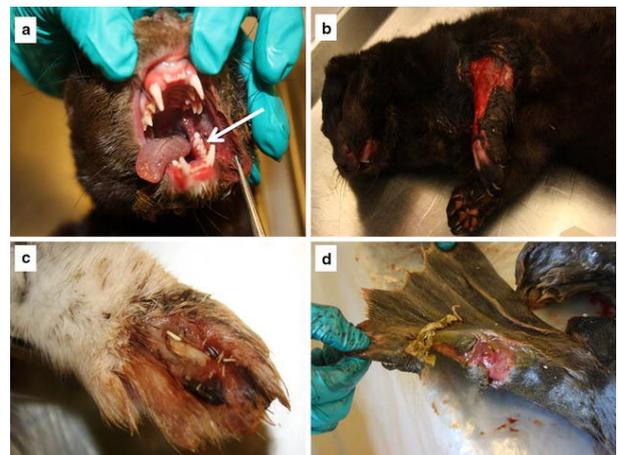


Fig. 2. *Arcanobacterium phocae* associated lesions in mink and seals. **a** Mink with stomatitis of the buccal mucosa (arrow) and suppurative dermatitis of the cheek. *A. phocae* was cultured from the lesions. **b** Severe profound necrotizing dermatitis on the forelimb of a mink. *A. phocae* was cultured from pus. **c** Exudative pododermatitis caused by *A. phocae*. **d** Ulcer on rear flipper of a seal. *A. phocae* was cultured from the ulcer

Acta Vet Scand. 2017 Oct 26; 59 (1): 74.

Doi: 10.1186/s13028-017-0342-8.

Genome Sequence of a Porcine Bocavirus Detected in Feces of Domestic Minks in China

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We report here the genome sequence of porcine bocavirus strain PBOV-JZ08, which was isolated from mink feces in China. Sequence analysis implied that PBOV-JZ08 clustered with three porcine bocaviruses.

Genome Announc. 2017 Nov 9; 5(45). pii: e01170-17.

Doi: 10.1128/genomeA.01170-17.

Molecular detection and genetic characterizations of Cryptosporidium spp. in farmed foxes, minks, and raccoon dogs in northeastern China

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Cryptosporidium spp. are common intestinal protozoa causing diarrhea in humans and a variety of animal species. With the recent development of fur industry, a large number of fur animals are farmed worldwide, especially in China. The existence of identical Cryptosporidium species/genotypes in humans and fur animals suggests zoonotic potential. In order to assess the presence of zoonotic Cryptosporidium species and/or genotypes in farmed fur animals, 367 fecal specimens were collected from 213 foxes, 114 minks and 40 raccoon dogs farmed in Heilongjiang, Jilin, and Liaoning provinces, northeastern China, during the period from June 2014 to October 2016. By PCR and sequencing of the partial small subunit (SSU) rRNA gene of Cryptosporidium, 20 of

367 (5.4%) animal samples were found to be infected, corresponding to 12 of 213 fox samples (5.6%) and 8 of 114 mink samples (7.0%) screened. Three Cryptosporidium species/genotypes were identified: C. canis (n=17), C. meleagridis (n=1) and Cryptosporidium mink genotype (n=2). Two host-adapted C. canis types (C. canis dog genotype and C. canis fox genotype) were found. By PCR and sequencing of the partial 60 kDa glycoprotein (gp60) encoding gene, one mink genotype isolate was successfully subtyped as XcA5G1R1. The three Cryptosporidium species/genotypes identified in this study have been previously reported in humans suggesting that fur animals infected with Cryptosporidium spp. may pose a risk of zoonotic transmission of cryptosporidiosis, especially for the people working in fur animal farming and processing industry.

Parasitol Res. 2017 Nov 25.

Doi: 10.1007/s00436-017-5686-5.

Hepatitis E virus detected from Chinese laboratory ferrets and farmed mink

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A novel genotype of hepatitis E virus (HEV) has been discovered in ferrets and mink worldwide. However, the situation in China is unknown. Diverse HEV RNA belonging to HEV genotype C2 was detected in this study from 17.5% (18/103) of the laboratory ferrets and 1.9% (2/107) of farmed mink in China. Phylogenetic analysis showed that the ferret HEV strains clustered into three groups with known strains detected from the United States and the Netherlands. Attempted transmission of ferret HEV to two rabbits was not successful. This study confirmed that the C2 genotype of HEV is circulating in China and succeeded in infecting laboratory ferrets with this genotype of HEV. Further studies are required to determine whether the C2 genotype of HEV can infect humans.

Transbound Emerg Dis. 2017 Sep 22.

Doi: 10.1111/tbed.12720.

Serotype and virulence genes of *Klebsiella pneumoniae* isolated from mink and its pathogenesis in mice and mink

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In the study, 15 *K. pneumoniae* strains were isolated from the mink experiencing respiratory distress in mideastern Shandong province, China, and the prevalence of *K. pneumoniae* in the sampled mink was 11.9% (15/126). Fourteen (93.33%) of the 15 *K. pneumoniae* isolates were identified as serotype K2 and hypermucoviscosity phenotype. The 12 virulence-associated genes of the *K. pneumoniae* isolates

were tested. The prevalence of the *wabG* gene for the isolates were 100% (15/15), the *ureA* gene 100% (15/15), the *rmpA* gene 93.33% (14/15), the *aerobactin* gene 93.33% (14/15), the *uge* gene 93.33% (14/15), the *IucB* gene 80% (12/15) and the *ybtA* gene 13.33% (2/15). But the other five genes, *fim*, *iroNB*, *wcaG*, *alls* and *kfuBC*, gave a negative PCR reaction in the 15 isolates, respectively. The animal experiments using *K. pneumoniae*-SD-12 and *K. pneumoniae*-SD-21 demonstrated that the serotype K2 was high virulence for mice and mink. These finding implied there exist potential threat that *K. pneumoniae* pathogens could transmit to human, especially the fur animal farm workers and residents lived near the fur animal farms. Therefore, the etiology and epidemiological surveillance of *K. pneumoniae* in mink should be strengthened for people's public health.

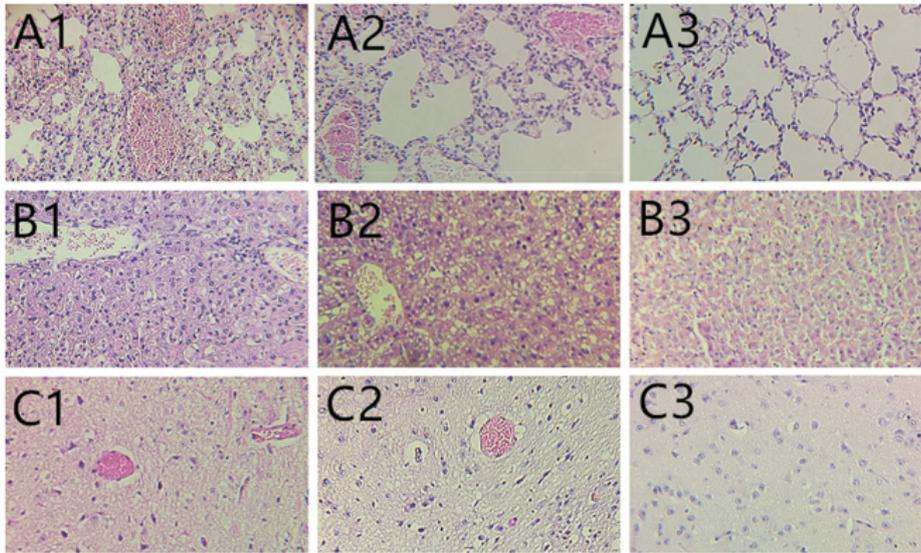


Fig. 1. Histopathologic appearance of the tissues of the experimental mice. **(A1)** Lung tissue taken from a mouse died from *K. pneumoniae*-SD-12 infection on days 4 p.i., characterized by bleeding of the lung breakage. **(A2)** Lung tissue taken from a mouse died from *K. pneumoniae*-SD-21 infection on days 4 p.i., characterized by bleeding of the lung breakage. **(A3)** Lung tissue taken from a euthanized mouse inoculated with 0.9% NaCl solution on days 4 p.i. **(B1)** Liver tissue taken from a mouse died from *K. pneumoniae*-SD-12 infection on days 4 p.i., characterized by congesting and steatosis of the liver breakage. **(B2)** Liver tissue taken from a mouse died from *K. pneumoniae*-SD-21

infection on days 4 p.i., characterized by congesting and steatosis of the liver breakage. **(B3)** Liver tissue taken from a euthanized mouse inoculated with 0.9% NaCl solution on days 4 p.i. **(C1)** Brain tissue taken from a mouse died from *K. pneumoniae*-SD-12 infection on days 4 p.i., characterized by light bleeding and edema of the brain breakage. **(C2)** Brain tissue taken from a mouse died from *K. pneumoniae*-SD-21 infection on days 4 p.i., characterized by light bleeding and edema of the brain breakage. **(C3)** Brain tissue taken from a euthanized mouse inoculated with 0.9% NaCl solution on days 4 p.i. HE stain. Original magnification was $\times 200$ for all images.

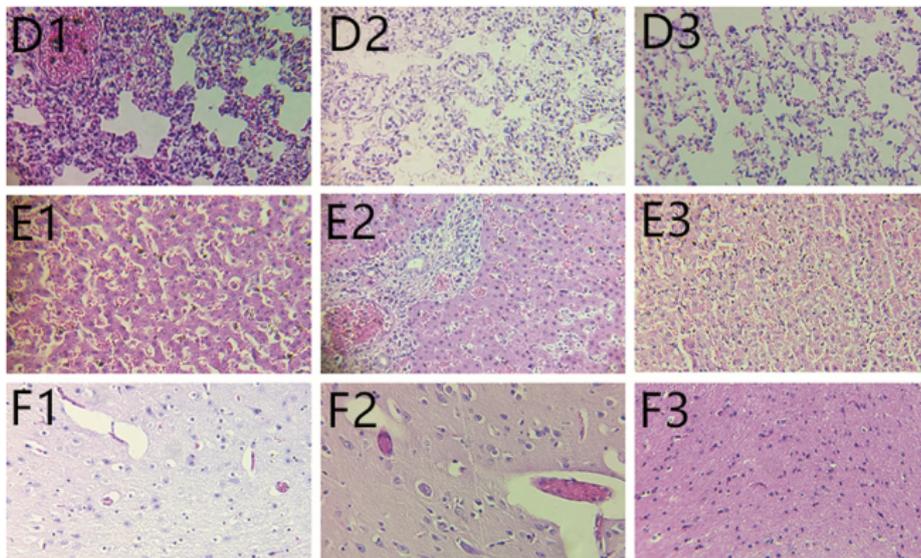


Fig. 2. Histopathologic appearance of the tissues of the experimental mink. **(D1)** Lung tissue taken from a mink died from *K. pneumoniae*-SD-12 infection on days 5 p.i., characterized by bleeding and congesting of the lung breakage. **(D2)** Lung tissue taken from a mink died from *K. pneumoniae*-SD-21 infection on days 5 p.i., characterized by bleeding of the lung breakage. **(D3)** Lung tissue from a euthanized mink inoculated with 0.9% NaCl solution on days 5 p.i. **(E1)** Liver tissue taken from a mink died from *K. pneumoniae*-SD-12 infection on days 5 p.i., characterized by congesting and steatosis of the liver breakage. **(E2)** Liver tissue taken from a mink died from *K. pneumoniae*-

SD-21 infection on days 5 p.i., characterized by congesting and steatosis of the liver breakage. **(E3)** Liver tissue from a euthanized mink inoculated with 0.9% NaCl solution on days 5 p.i. **(F1)** Brain tissue taken from a mink died from *K. pneumoniae*-SD-12 infection on days 5 p.i., characterized by light bleeding and edema of the brain breakage. **(F2)** Brain tissue taken from a mink died from *K. pneumoniae*-SD-21 infection on days 5 p.i., characterized by light bleeding and edema of the brain breakage. **(F3)** Brain tissue from a euthanized mink inoculated with 0.9% NaCl solution on days 5 p.i. HE stain. Original magnification was $\times 200$ for all images.

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Doi:10.1038/s41598-017-17681-8.

High-resolution phylogeny providing insights towards the epidemiology, zoonotic aspects and taxonomy of sapoviruses

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The evolution, epidemiology and zoonotic aspects of Sapoviruses (SaV) are still not well explored. In this study, we applied high-resolution phylogeny to investigate the epidemiological and zoonotic origins as well as taxonomic classification of animal and human SaV. Bayesian framework analyses showed an increase in porcine SaV (PoSaV) population dynamics and genetic diversity between 1975 and 1982, resulting in a SaV gene flow and generation of new strains among porcine and human populations. Our results also show the contribution of different animal populations involved in SaV epidemiology and highlight zoonotic aspects, as exemplified by the crucial role that swine, dogs, mink and humans play in SaV spread. Additionally, phylogenetic analysis suggests that bats may play key role in SaV epidemiology. According to our hypothesis, these animals may act as reservoirs or intermediate host species, contributing to viral spread in zoonotic and other epidemiological scenarios and facilitating the generation of new SaV genogroups and genotypes through recombination events. Data from large-scale phylogeny partition based on patristic distance, did not show a correlation between transmission clusters on generation of SaV genogroups, nevertheless we present both important findings about SaV taxonomy and important considerations useful for further taxonomical studies.

Infect Genet Evol. 2017 Dec; 56:8-13.
Doi: 10.1016/j.meegid.2017.09.024.
Epub 2017 Sep 23.

Canine angiostrongylosis in Sweden: a nationwide seroepidemiological survey by enzyme-linked immunosorbent assays and a summary of five-year diagnostic activity (2011-2015)

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Background

For the first time in Sweden, *Angiostrongylus vasorum* was detected on the island of Sydöster in foxes and dogs in 2003. After sporadic detection of the parasite in foxes in southern Sweden, the first positive canine faecal sample on the mainland was found in 2011. Since then a total of 2882 faecal samples have been analysed with the Baermann test at the National Veterinary Institute (SVA) during the years 2011-2015; 20 of them being positive. Contemporaneously, of over 525 fox necropsies, only three were found to be infected. To gather a more accurate knowledge of *A. vasorum* occurrence in Sweden, a large scale seroepidemiological survey was performed and totally 3885 serum samples from dogs were tested for both the presence of circulating antigens and of specific antibodies to *A. vasorum*.

Results

In total, 0.10% (n = 4, 95% Confidence Intervals, CI 0.03-0.26%) of the dogs were positive for both antigen and antibodies, whereas 0.51% (n = 20, CI 0.31-

0.79%) of the tested dogs were only antigen positive and 0.88% (n = 34, CI 0.61-1.22%) only positive for specific antibodies. Seropositive animals, as well as the majority of *A. vasorum*-positive faecal samples tested during the same period, were spread over central and southern Sweden. Annual prevalence of positive faecal dog samples and of necropsied *A. vasorum* positive foxes (coming from southern Sweden) varied from 0.3 to 0.9% (overall: 0.7%, CI 0.4-1.1%) and 0.0 to 1.4% (overall: 0.3%, CI 0.1-0.9%), respectively.

Conclusions

The findings confirmed that *A. vasorum* has become established in various geographical areas of central and southern Sweden. Veterinarians and dog owners should be aware of the potential risks of infection in large areas of the country, since canine angiostrongylosis may be a fatal disease if left untreated.

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Doi: 10.1186/s13028-017-0351-7.



Oslo 2017

**NJF SEMINAR 498
AUTUMN MEETING IN
FUR ANIMAL RESEARCH 2017**

**Radisson Blu Plaza Hotel in Oslo
18 – 20 October 2017**

BREEDING AND GENETICS SESSION

Variation in Body length in mink at 28 weeks of age is largely established before the age of 10 weeks

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Body length (BL) in mink is correlated to pelt size (PS). BL in adult brown type mink shows a large variation (45 cm to 55 cm in males, 35 to 45 cm in females). Edelveen Research farm has developed a practical system to measure the BL using a transparent tube. In 2016 kits from line mating were measured for BL and body weight (BW) at 10, 14, 18, 22 and 25 weeks of age and post-mortem at 28 weeks of age. At 10 weeks of age the BL varied from 33 to 41 cm in males and from 30 to 37 cm in females, respectively 75% and 85% of the average adult BL. Between 10 and 28 weeks of age 75% of the males grew at least 12 cm and 85% of the females grew at least 6 cm. During this period very little additional variation in BL was developed. The PS is already significantly correlated with the BL at 10 weeks of age and this correlation hardly gets any stronger at 28 weeks of age. Variation of BL in mink appears to originate in the lactation period. In accordance with this, we found that measurements of BL from as early as 10 weeks of age gives a good indication of both the expected BL and PS of individual mink.

Stretchability of mink skin appears to be genetic correlated

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Pelt size (PS) after processing of mink is on average twice the body length (BL), although the exact PS is hard to predict due to individual differences in stretchability. Using regression formulas the PS can be calculated using the BL, the body weight (BW) and the sex of the mink ($R^2=0.7$). We define the difference in stretchability (DS) as the measured PS minus this calculated PS. To evaluate if DS is hereditary a two-year project was started with line mating of 368 female mink housed in 46 blocks of 1 male and 8 females each. Their kits were housed in pairs or groups with littermates with a feeding regime aimed at 22 hours of access to

food per day. DS didn't vary a lot within each litter, while there was marked variation between litters. Often the nests in which all minks had a low DS and the nests in which all kits had a high DS were tied to specific male breeders. When comparing litters with high DS to those with low DS the PS of mink with the same BL and BW is up to 5 cm longer for males and 3.5 cm for females, explaining why PS is hard to predict when not accounting for DS. Stretchability of the mink skin appears to be hereditary making it a prime, but so far neglected, trait in breeding programmes aimed at achieving maximum PS.

Relocation of mink kits the first days after birth and survival during nursing period – studied using microchips

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The survival of relocated brown mink kits was investigated by microchipping the kits when they were relocated and following them for the rest of the nursing period. The results showed that the survival of the relocated largest kits was as good as survival of the foster mother's own kits, but it was best for the whole litter that the female only received one kit extra. The survival of relocated kits was not dependent on whether they came from a young female or a second year female. The timing of relocation within the first five days after birth did not affect kit survival, but the weight was significant so that the heaviest kits had the best chance of survival. A large proportion of the kits that came from litter size 1 died, but kits from litters greater than 10, had very low mortality. The best foster mothers were the second year females.

A Roofless Nest Box in Breeding Blue Fox Vixens

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In Finland a noticeable cub mortality in blue foxes may be related to insufficient whelping environment. Some blue fox farmers are using nest boxes without a roof based on their experience that it is a good whelping environment for vixens. However, the use of roofless nest

boxes has been criticized and it has been questioned whether they provide sufficient shelter for young, new-born cubs. The effect of using roofless nest boxes on cub mortality was studied in Kannus Research Farm Luova Ltd. in Spring 2017. The study revealed that there is no major differences on cub mortality between nest boxes with and without the roof. Under such conditions, the standard sized nest box without the roof can be as good whelping environment as the standard nest box with the roof.

Inbreeding and effective population size in Finnish blue fox

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¹*Natural Resources Institute Finland (Luke), Green technology, Biometrical genetics, 31600 Jokioinen, Finland.*

Management of genetic diversity of a blue fox population is essential for the sustainable use of genetic resources and genetic improvement. Therefore, significant effort is made to control inbreeding through a breeding program. Due to new national breeding scheme, it is expected that the selection of blue foxes will be more efficient in the future, and it may increase the kinship and inbreeding level of animals. The objectives of this study were to examine the average inbreeding and relationship coefficients, generation interval and the effective population size of the Finnish blue fox population. The average inbreeding coefficients were very low (<1.5%) among the production and breeding animals, which belong to the national breeding scheme. The average increase in inbreeding coefficients of the breeding animals was 0.050% per year. Kinship of the animals has increased gradually, and coefficient of relationship was 6.1% in 2016. The mean generation interval was 1.97 years and effective population size was estimated to be at least 150. The results showed that the effective population size has slightly decreased during the past ten years, but it is still fairly high. Kinship, inbreeding and rate of inbreeding have risen steadily, but they were nonetheless low. As a conclusion, it can be said that inbreeding depression is not a common problem in the current blue fox population.

VETERINARY & PATHOLOGY SESSION

Effects of Aleutian disease on mink semen and reproductive success

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The aim of this study was to investigate whether the Aleutian disease status of the male mink and amount of antibodies in the blood would affect males' semen quality, and thus effect the whelping result of the mink females. During the mating season semen samples were taken from both AD-free and AD-positive mink males. The samples were analysed by the Proiser ISASv1 analyser, and were divided into groups according to quality. The males were used in line mating to females in different known AD-groups, and the whelping result of these females was calculated according to normal routines on the farm. The stillborn kits were also counted. The AD-status of the females showed to have a greater effect on breeding result than did the AD-status of the male or the semen quality seen in the semen analysis.

Control of fleas on farmed mink

Kim Spholt Larsen¹ and Martin Sciuto¹

¹*KSL Consulting*

Mink farmers are confronted with the problem of flea infestations in mink and the need of adequate treatment against fleas. The squirrel flea, *Ceratophyllus sciuroorum*, is the most common flea on farmed mink and a heavy infestation of this flea can cause anemia, poor growth and sometimes death of very young mink kits. The fleas have also been demonstrated to be vectors of pathogenic organisms, e.g. the Aleutian Mink Disease. The only insecticides registered for flea control on mink farms in Denmark at present are dimilin and permethrin products. Treatments are made both preventively and when flea infestations are present on the farms. Failure of controlling the fleas has been experienced on more farms. The unsuccessful control seems to be due to the presence of permethrin resistance, but poor or changing management practice on the farms is also part of the problem. Lately new flea control products have been tested along with the development of "a

farm management flea control cycle". The results from these studies and an all year flea management cycle will be presented.

NUTRITION SESSION

Effects of plan of feeding on foot condition and animal welfare in blue foxes

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Natural Resources Institute Finland (Luke), Green Technology, ¹Kokkola, ²Maaninka, ³Jokioinen, ⁴Luova Research Farm, Kannus, ⁵Finnish Fur Breeders' Association, Vantaa.

Pronounced bending of forelegs is an escalating phenomenon in blue foxes (*Vulpes lagopus*) which has been recognized on farms and measures to understand its background, etiology and welfare implications has been initiated. The aim of the present study was to find out to which extent feeding intensity (ad libitum vs restricted) influences on foot condition and welfare in growing-furring blue foxes. The subjects were 200 sibling pairs divided into two groups: (1) Ad libitum (Ad Lib) and (2) Restricted feeding (Restr). Feeding arrangements were started on July 29. Restricted portion was raised from Sept 22 onwards to ad libitum level. The statistical analyses were done by using the Fisher's exact test, Tukey's test, Spearman correlations, Logistic regression and Mixed model of the SAS software for Windows. Initial body weights of groups were similar. From Aug 11 onwards body weights of Ad Lib remained significantly ($P<0.001$) higher compared to Restr ones during the restricted period. Thereafter, compensatory growth occurred in Restr groups ($P<0.05$). Final body weights at pelting (Dec 8) were significantly higher ($P<0.001$) in Ad Lib than Rest animals. Body condition score (BSC) followed the same pattern. Body length ($P<0.01$) as well as waist measure ($P<0.001$) and neck circumference ($P<0.01$) were longer in Ad Lib animals. Foot condition was better in Restr than Ad Lib both at the end of restricted period as well as at pelting. Moving difficulties, patellar luxation and veering were slight in both groups. Angulation of rear angle was more pronounced in Ad Lib animals ($P<0.001$). Weight of right adrenal gland was significantly heavier ($P<0.01$) in Ad Lib than Restr groups. Weights of thymus were heavier in Restr than Ad Lib animals ($P<0.01$). Animals in Restr group, particularly females, used platforms typically significantly more than animals in Ad Lib group ($P<0.01$). The conclusion of the

present study is that foot bending is the best method to evaluate foot condition even in large, fat animals. Prolonged high level feeding may decline foot condition and animal welfare.

Metabolic changes in male mink fed diets without vitamin B during the growing period

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An experiment was conducted to study the effect of feeding mink either a diet with recommended addition of vitamins and minerals or a diet without addition of vitamins and minerals. The mink were weighed four times during the study from July to pelting and blood and urine samples were collected in September and November. The samples were analyzed using non-targeted LC-MS based metabolomics. The weight of the mink did not differ in July, August, and September, but mink fed the diet without added vitamins and minerals had a significantly higher growth from September to pelting than mink fed a diet with added vitamins and minerals. In the urine samples, it was found that mink excreted riboflavin, niacin, pantothenic acid, and pyridoxine in the urine regardless of diet, showing that these vitamins are in excess even in a feed with no vitamins added. Furthermore, mink fed a diet without added vitamins and minerals had a lower excretion of amino acid metabolites which correlated with improved growth in this group indicating that more amino acids were available for deposition, this, however, requires further investigations.

Fat soluble vitamins in mink fed with and without vitamins and minerals during the growing period

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The effect of vitamin and mineral supplementation according to requirement was compared to an unsupplemented group of male mink in the growing and furring

period with respect to growth rate and content of fat soluble vitamins in plasma and liver, heart, lung, brain and adipose tissue. The mink showed no difference in growth, but the liver in the unsupplemented group was slightly but significant larger. Vitamin A and D did not differ in plasma between the two groups of mink, but the liver storage of vitamin A differed by a factor of five. The vitamin E content and the distribution of the stereoisomers of α -tocopherol varied between plasma and the analysed tissues. In general, there was a significant effect of vitamin E supplementation and a general preference for RRR- and RRS- α -tocopherol. Overall, the mink grow well without vitamin supplementation, but the storage reserves of vitamin A and E was diminished, while vitamin D status was unaffected.

Performance, nutrient digestibility and nitrogen utilization in blue fox (*Vulpes lagopus*) fed low-protein diets supplemented with DL-methionine and L-histidine

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Effect of low-protein diets supplemented with DL-methionine (MET) and L-histidine (HIS) on growth, pelt size and pelt quality was studied with 400 blue fox in two performance trials in growing-furring period in October (trial 1) and November 2016 (trial 2) in Luova Ltd, Kannus, Finland. Nutrient digestibility and nitrogen (N) utilization was studied in digestibility trial (trial 3) with 20 blue fox males in October 2016 in Luova Ltd, Kannus, Finland. In trials 1 and 3, diets contain digestible crude protein (DCP) 24 (control), 20 and 16 % of metabolisable energy (ME) and in trial 2, 20 (control), 16.5 and 13 % of ME. In all trials, the middle protein level was fed with or without MET and the lowest protein level was fed with MET and with or without HIS. In trial 1, blue foxes had the greatest average daily gain (ADG) and male blue foxes had the best pelt quality on control diet. In trial 2, blue foxes had greater ADG with DCP 16.5 % and 13% of ME comparing to control group. In trial 2, male blue foxes had the best pelt quality and both males and females the biggest pelt size with DCP 13 % of ME. In digestibility trial, differences in nutrient digestibility and N retention between groups were minor.

Comparison of expected and determined apparent total tract digestibility coefficients of nutrients for Danish mink feed and which factors that affects these digestibility coefficients

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The purpose of this study is to compare expected and determined apparent total tract digestibility coefficients (ATTDC) of protein, fat, and carbohydrate in complete diets, to test if the assumption used during feed optimization of additivity of nutrient digestibility of single feed items holds. Further it is tested if BMI status, no addition of vitamin and mineral pre-mix, sex, or genetic background affects ATTDC. Results from eight different digestibility trials performed in 2016 and 2017 with mink fed complete diets from four different Danish feed kitchens and the feed kitchen at Kopenhagen Furs research facility were used. Results generally support the assumption of additivity of ATTDC of protein, fat, and carbohydrate for single feed items, but determined ATTDC of fat was lower for minks with low BMI status and for white males. For young males with extreme low BMI status, ATTDC of fat was decreased by 46.2 percent units compared to a control group of normal weight young males. White adult males also had significantly impaired ATTDC of fat compared to brown adult males by 2.5 percent units. There was no significant effect on ATTDC of not adding vitamin and mineral pre-mix to the diet. Sex did neither affect ATTDC of protein, fat, nor carbohydrate, when protein content of faeces was corrected for urea contamination.

Effect of different feeding strategies on growth of mink (*Neovision vison*)

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¹Kannuksen tutkimustila Luova Oy.

²Seredat Oy.

The quality of mink skins (size and fur quality) produced in Finland has been improved by e.g. importing new breeding animals. Feed and the quality of its raw materials has been known to affect the growth of mink as well as the size and quality of the skin. Additionally, one factor affecting growth is suspected to be that how often and when in the daytime mink is fed. There are individual fur farmers who have distributed mink feeding rates to more than one or two daily doses. In their view, changing the way of feeding can affect the

growth of mink positively. This idea for different feeding strategies for improving mink growth and skin quality was studied. An effect of a number of feeding times and the time when feeding is performed on the growth of mink from weaning to pelting was studied. Black mink kits were weaned in July 29th. The litters were divided into feeding groups: R1, R2 and R3, each with 100 litters. The litter size (3 or 4 kits/cage) and the ratio of male/female was kept as similar as possible in different feeding groups. The three feeding groups are as follows; R1: minks were fed from weaning to pelting 2 times per day at 8 am and at 3 pm; R2: minks were fed from weaning to the beginning of August 2 times per day at 6 am and at 9 pm and from September 12th until pelting minks were fed once a day at 3 pm; R3: minks were fed from weaning until September 12th 4 times a day between at 6 am and at 9 pm and from September 12th until pelting feeding took place 2 times a day between at 8 am and at 3 pm. The minks in all three feeding groups were fed with commercial mink feed required by the growing season. The feed was dosed according to the feeding pattern set for the Farmplanner® feeding program. The actual amount of feed given to the mink was calculated in R1, R2 and R3 as an average over a week-long period starting from weaning. The skin data of the feeding groups is collected by means of separate skin monitoring. Randomly selected litters (30 litters/ feeding group) were weighed from weaning to pelting in every two weeks for growth data analyses. The statistical analysis was done with the R-statistics software (<https://www.r-project.org/>). Statistical analysis was performed first in basic analysis and further for more detailed analysis in multivariate analysis of growth. The basic analysis of the data suggests that there are differences between the feeding groups R1, R2 and R3 at the end of growth. Therefore, growth was analyzed over the entire study period by splitting the growth into two-week periods. The multivariate analysis of growth was calculated. In addition, the group size and gender impact within the cage were to be examined in more detail by multivariate analysis. The multivariate analysis showed that there are many significant variables in the study. A number of different factors have a statistical significance on the interpretation of mink growth results: time (phase of growth), cage (place), group size and gender distribution within the cage. There was no significant difference in skin quality between the feeding groups. However, the male/female ratio in the cage had a significant effect on the growth. We can conclude based on the multivariate analysis that the feeding groups R1, R2 and R3 did not significantly differ by growth or skin quality from one another.

Effect of dietary fatty acid composition on fat digestibility and metabolism in male mink kits (*Neovision vison*) from 6 to 11 weeks of age

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The digestive tract and enzyme activity is not fully developed in mink kits and this may affect the digestibility of nutrients. Previous studies have shown that high contents of fat in the diet to mink kits may decrease the overall fat digestibility and thereby the metabolizable energy (ME) content in the diet. Our objectives were to investigate fat and fatty acid digestibility of diets with different fatty acid compositions fed to mink kits from 6 to 11 weeks of age, with a calculated distribution of ME from protein fat and carbohydrates of 35, 50 and 15%, respectively. Furthermore, to investigate the effect of dietary fatty acid composition on ME intake, growth, organ development and fat soluble vitamin concentrations in plasma and tissues. Eighty mink kits, divided into 4 dietary treatment groups, were studied from 6 to 11 weeks of age in order to determine fat and fatty acid digestibility of four different fat sources. Ninety percent of the dietary fat consisted of soy oil (mainly C18:1n-9, C18:2n-6), sunflower oil (mainly C18:1n-9, C18:2n-6), coconut oil (mainly C8:0-C12:0) and Lipitec® (mainly C16:0, C18:0) in groups 1, 2, 3 and 4 respectively. It can be concluded that diets based on fat sources with a high proportion of unsaturated fatty acids have higher digestibility than such with a high content of saturated fatty acids and that especially the content of C18:0 is determining for the overall fat digestibility. Diets with low content of ME resulted in higher feed intake which shows that the feed intake in male kits aged 6 to 11 weeks is mainly regulated to a similar ME intake. In addition, the intestine weight in relation to body weight and length was also affected by dietary fatty acid composition where high dietary content of saturated fatty acids resulted in a longer and heavier intestine than kits fed diets with high content of unsaturated fatty acids. Vitamin A, D and E concentration in plasma increased with age, whereas vitamin A in hepatic tissue and vitamin E in adipose tissue decreased with age. The higher content of vitamin E in adipose tissue and vitamin A in hepatic tissue in the mink fed Lipitec® was most likely caused by the higher

feed intake resulting from the high content of poorly digestible saturated fat.

Calcium sources and calcium levels in diets for mink

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Eggshell was compared with limestone as calcium (Ca) source for mink in the lactation period (6 females per group, 6 weeks) and in the growth period (10 males per group, 23 weeks). The current recommendation for Ca is 0.6 % and 0.4% of DM in the lactation and growth period, respectively. The Ca level in two periods were planned to be slightly below requirement, but Ca in the lactation period was 0.8-1.0 of DM and adjusted to as low as 0.15-0.35% of DM in the growth period. Ca from the limestone and eggshell accounted for about 50 and 30 % of total dietary Ca content of the lactation and growth period, respectively. In the lactation period, females given the eggshell diet weaned kits with higher BW than those in the limestone group. Apparent Ca digestibility was higher ($P < 0.06$) in six week old kits with the eggshell supplement, 66 % versus 54% with limestone. BW, body length and length of femur was higher with the eggshell diet, thus showing that Ca from eggshell may have a better availability than Ca from limestone. The result of the study in the growth period confirm the current minimum recommendation of 0.4% Ca per kg DM.

Hydrolysed proteins from poultry by-products in diets for silver foxes

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Enzymatically produced protein hydrolysate made from poultry by-products containing 32 % crude protein, 4 % ash and 1 % crude fat after evaporation was examined as protein source for silver foxes in the growth period (Sept-Dec 91 d). Protein from the hydrolysate accounted for 60 and 30 % of dietary protein

in two experimental diets. Control diets with 60 % of protein from chicken filet or from poultry meal was included in the study. Protein digestibility was high for all diets (>80%). Amino acid content of the complete diets were similar, except that Trp was lower with hydrolysate diets. Feed intake and body weight gain did not differ among diets, and body composition, which was determined to evaluate if body fat deposition was lower with the hydrolysate, was similar (37-40% body fat) irrespective of protein source. The hydrolysate increased serum L-carnitine, which is involved in fat oxidation, but no effect was observed on body fat deposition. Small intestinal health parameters were not affected by protein source, but the poultry meal diet showed the best results concerning villi height and absorption area. The study show that the hydrolysate have good protein quality and suitable as ingredient for fur animals.

Taurine deficiency in silver foxes

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Taurine (Tau) is a beta aminosulfonic acid, which is essential for cats, but not for dogs and foxes, provided a sufficient dietary supply of methionine and cysteine (sulphur containing amino acids). Tau is not incorporated into body proteins but is one of the most abundant free amino acids in the body. It is concentrated in the heart, brain and eye tissue. One major function of Tau is conjugation with bile acids in the liver as well as other essential compounds to make them more hydrophilic such as vitamin A. Taurine is produced from cysteine in most species, but the production is very slow in strict carnivores. Deficiency of Tau reveal several symptoms since it is involved in fetal development, growth, reproduction, sight, hearing, heart function fat emulsification, immune response and bile acid (NRC, 2006).

Dilated cardiomyopathy and retinal degeneration is the pronounced symptoms of taurine deficiency in cats. Cats do not produce Tau at sufficient amounts because of low hepatic cysteinesulfinic acid carboxylase activity. Cats also use Tau as the only conjugator to produce bile acids and cannot apply glycine for the same purpose if Tau levels are low.

Concerning dilated cardiomyopathy in cats, the reason for these symptoms of Tau deficiency is not clarified. Except that Tau is found abundantly in the heart muscle and therefore most probably has a role cardiac contractility. Dilated cardiomyopathy has also been observed in farmed silver foxes in USA (Moise et al. 1991) with low plasma Tau. The symptoms of the heart problems was mainly in young growing foxes. The dietary Tau levels were not examined in the study of the foxes, which came from different farms, but study could not exclude that the cause could be both low dietary Tau levels, or genetically disposition for low Tau production from Met and Cys (Moise et al, 1991).

In Norway in 2017, some young silver foxes have shown symptoms of dilated cardiomyopathy after autopsy. Several young individuals have died in July/August after showing typical symptoms like shortness of breath before dying after a few days. The autopsies have indicated signs of dilated heart muscle. As the heart chambers dilate, the heart muscle does not contract normally and cannot pump blood very well. Analyses of the diets have not revealed very low levels of Tau, and normally animal by-products of which fur animal feed is made from, contain high levels of Tau. The requirement in growing and adult cats is 24 mg Tau per MJ and the feed applied for the foxes contained 96 mg per MJ. The feed also contained sufficient amounts of the building blocks Met and Cys, 0.30 and 0.16 g/MJ, respectively. Fish is a richer source of Tau than source from land animals, one may therefore speculate if the last years lower inclusion levels of fish by-products and higher levels of chicken by-products has reduced the Tau supply from the feed. Analyses of Tau from serum samples of silver foxes with symptoms will probably give more information if the diagnosis is low Tau or not.

Weight-for-length index as a measure of obesity in blue foxes

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The blue fox experiment for studying the variation in weight, feed intake and computed feed efficiency was utilized to assess the use of relative weight in estimating the fatness of animals. The relative weight is formed from the length based standard weight. The relative weight together with body length explains almost

70% of the variation in feed intake and has a high heritability. The relative weight as an indicator for fatness would require a detailed comparison with body composition measurements.

ETHOLOGY & WELFARE SESSION

Do juvenile Finnraccoons (*Nyctereutes procyonoides ussuriensis*) rest on the platform?

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Resting platforms are used as resting site in fur animals, like foxes (*Vulpes vulpes* and *V. lagopus*) and mink (*Neovison vison*), but also in the Finnraccoon (*Nyctereutes procyonoides ussuriensis*). In contrast to other fur animal species, the scientific knowledge of platform use is limited in the Finnraccoon. The aim of the present study was to document the use of the platform in the Finnraccoon, with special attention to resting behaviour. Sixteen sister-pairs of Finnraccoons were housed in 1.2 m² cages from weaning. Each cage was equipped with a slightly V shaped resting platform (30 × 115 cm, width × length), made of plastic coated wire mesh, mounted 40 cm from the cage floor in the 70 cm high cage. In order to demonstrate the effect of not having the platform available, the access to the platform was denied (deprivation) for two weeks (14 days) from eight pairs in two occasions; i.e. in the middle of September and from the end of November to mid-December. These sister-pairs were labelled as Deprivation group, whereas those sister pairs having permanent access to the platform were labelled as Control group. The resting behaviour of Finnraccoons was video recorded 24 hours for six times: before each deprivation period, 12th day of each deprivation and after each deprivation. The behaviour was analysed from the video recordings by using instantaneous sampling with a five minutes sampling interval. The data were analysed by using Linear Mixed Model (SPSS). The Finnraccoons rested 67-74% of the daily observations, slightly increasing with advancing autumn ($F_{5,70} = 2.62$, $P = 0.031$), similarly in both groups (group × recording interaction: $F_{5,70} = 0.14$, $P = 0.983$). No difference between groups was found in the total time spent resting ($F_{1,14} = 0.73$, $P = 0.409$). Of the total resting, allohuddling on the cage floor was the most common type of resting (61-91% of the resting observations) increasing with the advancing autumn ($F_{5,70} = 3.61$, $P = 0.006$) and being more common in the Deprivation group than in

the Control group ($F_{1,14} = 5.52$, $P = 0.034$). Resting on the platform constituted 2-23% of the resting observations, without difference between groups ($F_{1,14} = 2.95$, $P = 0.108$). Resting on the platform tended to decrease with advancing autumn ($F_{3,42} = 2.60$, $P = 0.065$) similarly in both groups ($F_{3,42} = 0.54$, $P = 0.661$). Allohuddling on the platform was observed only occasionally in five out of the 16 sister pairs, and in two of these, only in September. When also activity was included, the platform was used for 2-17% of the day. Resting alone on the cage floor decreased with advancing autumn ($F_{5,70} = 4.34$, $P = 0.002$), constituting 9-20% of the resting observations. No difference between groups was found in the resting alone on the cage floor ($F_{1,14} = 1.30$, $P = 0.273$). The resting behaviour followed circadian rhythm: resting was the most common behaviour in the evening and night-time, whereas less resting was observed in early hours and during working hours. This general trend changed slightly as the autumn proceeded. Staying on the platform was more common during the working hours than during night-time. The results show that, juvenile Finnraccoons preferred allohuddling on the cage floor instead of resting on the platform. This preference was clear especially in the evening and night-time, whereas during the working hours, less resting was observed in general, and also other resting locations were used. However, Finnraccoons use the platform mainly for resting and not so much to active behaviours. Platform is used most commonly during the working hours, which suggest that the platform is used for surveillance of the human activities on the farm. The deprivation of the platform did not clearly affect the distribution of the resting, since the general level of resting on the platform was so low. Although the level of use is at a relatively low level, access to a platform may enhance the welfare of Finnraccoons through increased complexity of the cage and by providing choice

WelFur on-farm welfare assessment of foxes: development of behavioural tests

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Firstly, we studied associations between the two behavioural tests in the WelFur on-farm welfare assessment protocol for foxes. The Feeding Test (FT) is well validated and it measures human-animal relationship (HAR), whereas the more poorly validated Stick Test

(ST) is assumed to measure one component of positive emotional state, namely explorative behaviour. Our results from the breeding animals of one commercial farm with both blue foxes ($n = 690$ females and 43 males) and silver foxes ($n = 81$ females and 24 males) show, however, that also the ST is probably rather a HAR test than a test of explorative behaviour. Accordingly, the less-laborious ST could probably substitute for the more laborious FT in the WelFur-Fox protocol. On the other hand, quite obviously a new test for assessing exploratory behaviour or positive emotional state in WelFur-Fox is needed. Secondly, we compared the results of the FT also to the results of a potential new test, a Subjective Evaluation of HAR (SE). In the SE the assessor recorded on a six-point scale a fox's response to him/her (0 = approaches, 5 = withdraws) at the same time when recording other WelFur-Fox measurements from the animal and its cage. The SE is, thus, less-laborious than the FT. The SE score distributions overlapped to some extent between the eaters (confident towards human) and non-eaters (not confident towards human) in the FT. This indicates either i) that the SE requires further development aiming at increasing its discriminative power before it could replace the FT in the WelFur-Fox protocol, or ii) that the SE has more fine-tuned discriminative power than the FT.

Additional water for kits did not significantly increase animal welfare at farm level

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Mink kits are gradually eating solid feed from about four weeks of age. Several studies have shown positive effects of providing water which is easy accessible for mink kits on animal welfare, with less saliva licking from the commissure of the dam's lips, less injuries and higher growth rate in kits with early and easy access to water (Brink *et al.*, 2004; De Rond & Kleyn van Willigen, 2012; Hansen *et al.*, 2015; Jespersen *et al.*, 2014; Malmkvist *et al.*, 2016; Møller & Lohi, 1988; 1989 OR Møller 1991). The position of the water is found to be important. A trial at the research farm AU-Foulum found that the kits started to drink 11.1 days earlier with a water nipple positioned in the cage near the nest box entry, than if only the ordinary watering system was provided (Caille, 2016; Malmkvist *et al.*, 2016), but only a few days earlier if the normal watering system was made more accessible (Møller, 1991). We do therefore assume that a water nipple positioned near the

nest box entry will ease the kits access to water and significantly reduce the prevalence of injuries from four weeks post-partum till weaning.

The former studies on providing water that is easy accessible for mink kits have mainly been controlled studies on research farms. In these studies, the management procedures are highly controlled and no extra care is taken for the different groups within a study. In commercial farms, the farmer will typically pay more attention to large litters or litters that do not seem to thrive, e.g. with signs of injuries or reduced growth. We, therefore, decided to study the effect of providing easy accessible water for kits in commercial farms, where the farmers manage the animals as they would normally do. This way we can see whether easy accessible water can have a positive effect for the kits, above what proper management can compensate for.

Providing water which is easily accessible for kits is registered as positive and will give a better welfare score in the welfare assessment system WelFur-Mink in relation to the welfare criteria 'Absence of thirst' (Møller *et al.*, 2015). Due to the earlier findings of fewer injuries and lower mortality when easy accessible water supply is provided for the mink kits, we also expect that this will affect the WelFur criteria 'Absence of injuries' and 'Absence of disease'.

The aim of the study was to evaluate the effect of providing easy accessible water nipples for mink kits at the nest box entry on a number of selected animal-based welfare indicators as well as the potential changes in the aggregated WelFur-Mink score at farm level. In this article, we present result regarding kit injuries and kit loss, and we hypothesized that the easy accessible water for kits would decrease the prevalence of kits with injuries by the end of the nursing period, and reduce the kit loss from four weeks post-partum until weaning.

Status and perspectives of the relations between science and the full-scale implementation of WelFur-Mink in Europe

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Fur Europe decided to implement WelFur in Europe on a voluntary basis from January 2017, while the fur auctions have decided only to sell European pelts from

farms that are classified 'Acceptable' or better from 2020. More than 50 assessors from a private audit company have been trained by the WelFur researchers, and have assessed more than 1000 mink farms in the winter and nursing periods. The implementation of such a comprehensive protocol is a huge task that requires training and accommodation at all levels before the new tasks become routine. It also requires that work-flows, tools and programs are refined to the task. New methods for taking unbiased and representative samples on a mink farm and for assessing killing methods have been developed to assist the assessment. In order to secure quick action to welfare problems observed, all results are reported back to the farmer after assessment. Advisors from each country have been taught how to interpret the WelFur results and help farmers when needed. Despite obstacles on the way, WelFur seems to be the first example of a full scale implementation of a science based welfare assessment in an entire animal sector.

Background

Since 2009, researchers from Aarhus and 5 other European universities have been working on the development of a welfare assessment protocol for mink and foxes after the concept developed in Welfare Quality®. The WelFur-Mink protocol is based on a critical evaluation of validity, reliability and feasibility at measurement level. The protocol has been repeatedly tested in 12 European countries and developed to handle all conditions found in each of the three seasons that covers the strict annual cycle of mink production. The assessment results from the 22 welfare indicators are aggregated across the 3 seasons into 12 welfare criteria and 4 welfare principles, from which each farm is classified as either 'Best', 'Good', 'Acceptable' or 'Unacceptable' current practice. In 2016, the European Fur Breeders Associations in Fur Europe decided to implement WelFur in all of Europe on a voluntary basis from January 2017. At the same time, the fur auction houses in Europe and North Americas decided only to sell European pelts from farms that are classified 'Acceptable -' 'Good -' or 'Best current practice' regarding animal welfare from 2020. These decisions started a race against with time in order to prepare everything for a full-scale implementation of WelFur.

Aarhus University have been responsible for the development of the science based welfare assessment system 'WelFur-Mink'. Therefore, it has been natural for us also to take responsibility for the training of assessors from the independent 3rd party company that are conducting the on-farm assessments. There has, therefore, been a close interaction between the completion of the program, which is used to enter information on the

farms, the company making the welfare assessments, Fur Europe and the researchers from the AU, to keep everything together so that farms from all over Europe could be assessed in a uniform manner from January 2017. WelFur is the first example of implementation of a complete welfare assessment system for livestock production in a whole sector in Europe. It is therefore limited how many experiences we have been able to draw on in the process. In this presentation, we look at the status of the implementation of WelFur-Mink and the challenges that have been resolved along the way in order to ensure a uniform assessment of farms across Europe. The challenges have been to further develop and optimize the WelFur-Mink protocol, train assessors to conduct WelFur assessments on mink farms, and educate consultants to advise breeders about the results, in all European mink producing countries and, in particular, explain what WelFur is, what it shows and what it can be used for.

Access to multiple enriched cages increase confidence and exploration and reduce head twirls in juvenile silver foxes

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Providing animals with access to environmental enrichments represents an important method to improve their welfare. Even if foxes' have favourite objects, e.g. meat bones, access to several enrichments give more choices, potentially reinforcing activity and play. In this study we aimed to examine how access to a multiple enriched cage affected some welfare related parameters like fear towards humans, exploration, abnormal repetitive behaviours (head twirls) and weight gain in cubs. Sixty 3-month-old cubs, 30 males and 30 females, were housed in male-female sibling pairs separated in a treatment and a control group. Both groups had access to double standard housing conditions with a top nest box, a resting shelf and wooden gnawing sticks. The treatment group had additional access to a tube, a wooden plate, a hockey-puck, straw and a meat bone. To examine fearfulness and exploration a feeding test, a repeated titbit-test and a novel object test were conducted after 4 and 5 weeks. Stereotypic head twirls were recorded from video 30 minutes before feeding time after 6 weeks. The feeding test did not differentiate between the groups, but the titbit test revealed significant differences ($P \leq 0.05$) where more cubs from en-

riched cages accepted the titbit (53.3 % vs. 23.3 %) after two trials. Immediately after handling none of the controls accepted the titbit compared to 16.7 % of the enriched foxes. Significantly more cubs from the enriched cages contacted the novel object (96.6 % vs. 70.0 %; $P=0.012$). The latency to contact the object was shorter ($P \leq 0.003$) and the manipulation time was longer in the enriched group ($P=0.019$). The number of cubs with head twirls were lower in the enriched group (36.7 % vs. 73.3 %; $P=0.009$) and the average number of head twirls was also lower compared to the controls ($P=0.012$). After 5 weeks, control cubs gained more weight compared to enriched cubs but this levelled off after 9 weeks ($P=0.382$). The results show significant effects of early access to multiple enrichments on cubs' behaviour; reducing fearfulness towards humans and novel objects, after only 6 weeks of access. The lowered level of head twirls indicates the potency of multiple enrichments to reduce frustrated motivations underlying the development of behavioural abnormalities. Our results suggests addition of multiple enrichments as a useful means for improving foxes' housing conditions.

POSTER SESSION

Evaluation of temperament in finnraccoon (*Nyctereutes procyonoides ussuriensis*)

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Two different experimental set-ups were completed to assess various aspects of temperament in cage housed Finnraccoons (*Nyctereutes procyonoides ussuriensis*). In Experiment 1 performed in autumn, the connection between the Stick test, Feeding test and Subjective evaluation of the temperament (Subjective evaluation) were studied in 214 juveniles housed in various housing conditions (small vs large cage; traditional enrichment vs. increased enrichments, group size 2 vs. 4). In Experiment 2 completed during winter, the effect of various housing conditions on the temperament of 60 adult females (singly vs pair housed, in small and large cage; with or without a winter nest) was examined by using the Feeding test, Stick test, Subjective evaluation and Trapezov's hand test. The study further clarified whether different assessors rated the temperament in a similar way at different and same time points when using subjective evaluation of the temperament alone, in

pairs and in group. The statistical tests included Cohen's kappa, Fisher's exact test, Logistic regression and Mixed models (SAS, v 9.4). In Experiment 1, 57% of animals explored the stick in a confident way in the Stick test; 13% were aggressive and 3% fearful towards the stick. In the Feeding test, 79% of animals ate in the presence of a human, thus being interpreted as confident. The animals classified as aggressive (Odds ratio estimate (Logistic regression), OR= 30.5, P = 0.013) or confident (OR= 6.5, P = 0.0446) in the Stick test ate more probably in the Feeding test than those classified fearful. In the Subjective evaluation, the majority of the animals were scored confident, varying from 60% to 85% between the six assessors. The higher the confidence score, derived as a sum from the six subjective evaluations by different assessors, the more eagerly the animal ate in the Feeding test (OR=1.031, P<0.001). The inter-assessor agreement varied from poor to excellent (Cohen kappa from 0.39 to 0.85) between the assessors. The agreement between the Subjective evaluation and outcome of the Stick test was poor (Cohen kappa below 0.40). Housing environment did not affect ($F_{3,49} = 1.43$, $P = 0.245$, Mixed model) the temperament. In Experiment 2, 77 - 100% of animals ate in the Feeding test, without difference between housing groups ($P = 0.32$, Fisher exact test). In the Subjective evaluation, 73% - 97% were scored confident. The agreement between the assessors was generally higher when the Subjective evaluation was done in the very same moment in pairs or in groups (0.455 - 1.000) than when done in different time points (0.016 - 0.655). The housing conditions affected the outcome of the subjective evaluation of the temperament in adult females ($F_{3,44} = 4.75$, $P = 0.0059$); less confident animals were found amongst the ones housed with access to a winter nest. In the Trapezov's hand test, so called non-domesticated animals varied from 29 to 58%, without difference between the housing group ($P=0.236$, Fisher exact test). It can be concluded that the majority of Finnraccoons are curious and behave confident towards humans. Although, the temperament tests share some common properties, the results do not unambiguously correspond between tests. It seems that all tests measure the same aspects of fearfulness, but only the Feeding test and Subjective evaluation of the temperament measure the same aspects of confidence. The Trapezov's hand test is a more invasive test than the others, and therefore it may better capture true confidence towards humans, including aggressive behaviour.

Expression of mannose-binding lectin in mink

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Mannose-binding lectin (MBL) recognises a number of structural oligosaccharide components on the surface of pathogens. By binding to a pathogen, MBL functions as an opsonin facilitating phagocytosis or activates the lectin complement pathway, thereby promoting the killing of the pathogen and playing a major role in the innate immune defence. Human individuals deficient in MBL have been found to be more susceptible to viral and bacterial infections as well as to autoimmune diseases, and the MBL deficiency is due to differences in the genetic basis of different individuals. The protein has been isolated and characterised from several mammals, including humans and pigs, but so far not from mink. Two forms of the MBL gene have been described in most species, with the exception of humans and chickens, where only one form has been found. In humans, the MBL promoter is highly polymorphic, and three positions in particular have been associated with different levels of the protein in peripheral blood.

In this study, MBL expression in brown and aleutian mink is examined and expression levels of MBL in the liver were found to differ between individuals. In order to elucidate if the differences are owing to polymorphism in the gene, the DNA sequences of the gene were compared. Identification of MBL polymorphism in the mink MBL gene may provide new opportunities for selective breeding.

Field observations of the effects of increased ammonium chloride supplementation on urine pH and struvite urolithiasis incidence in mink

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On each of two farms a group of mink was given a high dose of ammonium chloride in the feed (2.5 to 4 kg/tonnes) while a control group was fed unsupplemented feed. Urinary pH was measured at day 0 (before supplementing), day 6, day 13, day 40 and day 70 after start of supplementation. To determine if commonly

used single-use test strips are reliable, 362 measurements were performed both with an electronic pH probe (Testo 205 pH) and with single-use test strips (Dosatest pH 5.2-6.8). Random mortality was selected for post-mortem analysis. Results show no consistent effect of treatment on urinary pH, and both treated and untreated mink had a mean pH varying from 6.02 to 6.35 (farm 1) and from 5.88 to 5.98 (farm 2) depending on the month. Twenty-five percent of sampled mortality suffered urolithiasis, and all sampled stones were 100% struvite. It is suggested that factors other than the average urinary pH may present a major risk factor for development of struvite uroliths in mink. Additionally, under field conditions the single-use pH testing strips used in this study are an acceptable alternative to an electronic pH probe for determining mink urinary pH.

The effect of housing condition on the fur quality and selling price of the pelt in the Finn raccoon

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The Finn raccoon (*Nyctereutes procyonoides ussuriensis*) is an important fur animal in Finland. The aim of the present study was to examine whether the housing conditions affect the fur quality and the selling price of the pelt in juvenile Finn raccoons. A total of 152 Finn raccoons were raised from weaning until pelting in four different housing conditions, differing in the cage size, animal density, group size and furnishing of the cage. The length and quality of the fur were evaluated and the selling price was registered. The data were analysed by using the Pearson Chi-square test and Linear Mixed Model (SPSS). The fur quality differed between groups with the poorest quality in the group of four animals housed in an enriched cage. No general difference between males and females was found. No difference between groups was found in the distribution of the pelts into the size categories. The size category and the quality of the fur were the main factors determining the selling price of the pelt. The selling price was higher in pelts of males than in those of females. This difference between males and females appeared only in the Finn raccoons housed in pairs, and not in those housed in groups. When only the auction where the majority of pelts were sold was considered, the selling price of the pelt was higher in females when housed in larger cages instead of small cages and in enriched cages instead of

barren cages. In conclusion, the larger cage size and increased complexity of the cage may increase the selling price of pelts in juvenile Finn raccoons, especially in females. Although, the increased cage complexity impairs the quality of the fur, this does not finally affect the selling price of the pelt.

Harri 1986 may affect these variables. Also, housing conditions may influence the fur quality and the final selling price of the pelt (Korhonen 1987, Kasanen et al. 2000, Ahola et al. 2004).

The aim of the present study was to examine the effect of group size, available area and environmental enrichment on fur quality and selling price of the pelt in juvenile Finn raccoons.

Grading traits improve the genetic evaluation of fertility traits in Finnish blue fox

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The Finnish blue fox breeding scheme has to find new methods to improve fertility and fur quality traits and simultaneously maintain the current animal size. Estimated breeding values (EBVs) of fertility traits were previously calculated with repeatability animal model. However, the genetic trends of fertility traits have been modest because of unfavourable genetic correlations to some of the grading traits. The aims of this study were to develop a multiple trait animal model and estimate the (co)variance components between the grading and fertility traits to be used in the national breeding schemes of blue fox in Finland and Norway. This was the first time when these genetic parameters were estimated simultaneously for the following eight traits: pregnancy rate, whelping success, first litter size, grading size, quality, guard hair coverage, colour darkness, and colour clarity. Data were a sample from nine farms, which had 42462 animals. The pedigree contained 47177 animals. Heritability estimates for the first litter size, pregnancy rate and whelping success were low (0.14 ± 0.01 , 0.05 ± 0.01 and 0.06 ± 0.01 , respectively). For grading traits, the highest heritability was obtained for colour darkness (0.64 ± 0.01) and lowest for colour clarity (0.21 ± 0.01). The grading size, guard hair coverage and overall fur quality had moderate heritability estimates (0.27 ± 0.01 , 0.23 ± 0.01 and 0.22 ± 0.01 , respectively). Grading size and quality had the highest antagonistic genetic correlations with the most important fertility traits; 1st litter size (-0.53 ± 0.05 and -0.54 ± 0.05 , respectively) and whelping success (-0.44 ± 0.07 and -

0.37±0.08). Guard hair coverage also had antagonistic relationship with all the studied fertility traits (from -0.24±0.08 to -0.36±0.06). Colour clarity had unfavourable genetic correlation with the first litter size (-0.32±0.06) and with whelping success (-0.36±0.09). Colour darkness had no genetic correlation with the studied fertility or grading traits except for guard hair coverage (0.25±0.03). All fur quality traits (guard hair coverage, clarity and overall quality) had high genetic correlations (0.71±0.03 to 0.72±0.03) with each other. Our results showed that large grading size and excellent fur quality had fairly high antagonistic genetic correlations with reproductive results. Because fertility traits have low heritability, litter size, pregnancy rate and whelping success evaluations are likely to benefit from the multiple-trait model where grading traits with highest genetic correlations (grading size and quality) to the fertility traits are included. They will bring more information to the calculation of EBVs and consequently, accuracies of the EBVs are improved.

Assessing confidence in farmed foxes and mink in Norway – educational films for fur farmers

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The level of confidence, also termed tameness, is a measure of an animal's motivation to avoid or approach humans. A confident animal behaves in an assertive and calm manner in close contact with people. It is an important behavioural feature of domestic animals because it facilitates handling and care of animals. In these tutorial films, which is based on scientific research and knowledge in fur animals, we demonstrate how to recognise confidence in silver foxes, blue foxes and mink. The type of behavioural responses associated with confidence are specifically emphasised. In addition, behaviours associated with fear and aggression, responses that are undesirable when choosing future breeding animals, are also demonstrated. Based on behavioural responses during handling of animals and methods originating from Russian breeding experiments in foxes and mink, a 5-point assessment scale for confidence is presented. These films are aimed at helping fur farmers to recognise behavioural expressions in future breeding animals with particular emphasis on confidence, both in everyday situations on the farm and during behavioural tests.

**FUR ANIMAL EPIDEMIC NECROTIC PYODERMA
PATHOLOGY, ETIOLOGY, AND EPIDEMIOLOGY**

PhD thesis by Heli Nordgren

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In 2007, Finnish farmers detected signs of a new disease in fur animals. Mink had severe necropurulent dermatitis in the feet and head region and foxes had aggressive keratoconjunctivitis that spread to the eyelid and also to facial dermatitis. Even Finnraccoons (a raccoon dog bred for the fur industry), which are generally resistant to many diseases, suffered from painful furuncles between the toes.

The symptoms were severe, even fatal, and the disease seemed to spread aggressively between and within the farms. Similar symptoms (designated as "pododermatitis") in mink had previously been detected in the early 1970s in USA and in 1996 in Canada. North American farmers, and later researchers, linked the onset of the new symptoms

to the start of feeding mink with feed containing seal byproducts. Today, the fur industries have detected similar signs of this disease in many countries including Denmark, the Netherlands, Iceland, Poland, Spain, Greece, Norway, and Sweden.

The disease severely compromises animal welfare and causes financial losses to the farmers and the fur industry. The University of Helsinki (UH), the Finnish Fur Breeders' Association (FFBA), and the Finnish Food Safety Authority—Evira initiated a collaborative project in 2009 to describe the pathology of the disease, identify the causative organism(s), and describe the epidemiology of the disease in Finland including identification of possible sources and risk factors of the disease. Hence, we conducted pathological, microbiological, epidemiological, and experimental studies.

Ninety-nine fur animals underwent necropsies with complete microbiological examinations, including: mink, foxes, and Finnraccoons. The groups comprised diseased animals (61), their healthy controls from non-affected farms (34), and clinically healthy animals from affected farms (4). Due to the common gross and histologic lesions, particularly the severe necrotizing pyoderma observed in diseased animals of all fur animal species the disease was named *Fur animal epidemic necrotic pyoderma* (FENP).

No specific viral pathogens have yet been identified via microbiological examination, however, a suspected causative agent was seen in bacteriological studies. The bacterium *Arcanobacterium phocae* was isolated and confirmed by PCR from the affected tissues from all diseased animals (61/61), but not from healthy controls (1/23*) derived from non-affected farms. The association to *A. phocae* infection is particularly interesting, because this bacterium causes purulent skin inflammations and abscesses in marine mammals such as seals,

and a temporal connection between the onset of the epidemic in mink and the use of seals as a feed source for mink had previously been found in North America.

This report is the first to discover *A. phocae* in the samples obtained from fur animals. In addition, a novel *Streptococcus* species also with a likely marine origin and closely related to *Streptococcus halicoeri*, was detected in the tissues of animals with FENP, especially in mink and Finnraccoons. Further investigation is required into the role of the novel *Streptococcus* sp., as well as other possible viral or bacterial pathogens, in the pathogenesis of FENP.

Infection of mink with *A. phocae* alone was shown here to cause FENP-like symptoms. Signs were evident when *A. phocae* was delivered as the only infectious isolate (23/34), and also when delivered in combination with the novel *Streptococcus* sp. (7/8 tested). The inoculates were successfully delivered intradermally/subcutaneously or via artificially wounded skin. No signs were detected in animals that had been inoculated perorally (0/24). Similarly, inoculating mink with tissue extract of an FENP-diseased animal intradermally/subcutaneously or via artificially wounded skin caused typical signs (4/10) of the disease, whereas no signs were detected using the peroral inoculation route (0/4). The experimental infection also showed that the bacterium was able to spontaneously spread within in the experimental facility, as low levels of *A. phocae* were detected in non-inoculated mink towards the end of the study, but not from samples taken from the control animals from the beginning of the study. The experimental infection study further indicated that weather conditions may influence the outcome of FENP, as severe signs developed more rapidly when the ambient temperatures were very low (below – 20 °C).

Our epidemiological study conducted as a mail survey among Finnish fur farmers showed that the disease had spread to all areas where fur farming is practiced in Finland during the study years 2009-2011. Signs of FENP were reported in all farmed species, and the number of affected farms increased each year: 40% of responding farms (92/239) reported signs of FENP during the study period. The study also clarified the specific clinical signs of FENP observed by the farmers in different fur animal species. The presence of FENP on a given farm in the study was associated with importation of fur animals from Denmark and/or Poland, and the spread of the disease between Finnish farms was connected with fur animal purchases from domestic sources. Some predisposing risk factors were identified, such as the farm type: FENP was detected most on mixed farms (farming more than one species of fur animals) and on larger

farms (as defined by the number of the breeding fur animals). The study also showed an association between the occurrence of FENP on the farm and the access of birds and other wildlife to the farm and animal housing.

The use of the escape-proof shelter buildings, which block the access of birds and other wildlife to the shelter buildings and thus prevent close contacts between the wildlife and fur animals, significantly diminished the risk of FENP. The study indicated that FENP can be associated with other diseases on the farm as a positive association to sticky kits and a negative association to plasmacytosis was observed. The disease was also found to be associated with the color type of the mink, nipple drinking water dispensers, and the type of the bedding material used in mink nests.

This thesis describes the clinical signs and gross and histopathological lesions of FENP in fur animals. It implicates *A. phocae* as a likely causative pathogen for FENP and indicates a possible additional role of a novel *Streptococcus* sp. as a potential cofactor or co-pathogen. The data provide a basis for the development of the diagnostics, treatment, or vaccines for FENP. The data also describe the epidemiologic aspects of FENP in Finland showing that FENP is a highly detrimental disease among Finnish fur animals. Some potential risk factors that may be involved in contributing to the spread of the disease are revealed and thus provide targets for control and mitigation of FENP.

*The farm selected for healthy control farm for Finnraccoons proved to be affected during the study, hence healthy Finnraccoon controls (11) are excluded. One healthy mink from non-affected farm had low numbers of *A. phocae* in the PCR study.

The complete thesis can be downloaded from the open access database of the University of Helsinki at the address: <http://urn.fi/URN:ISBN:978-951-51-3808-8>



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