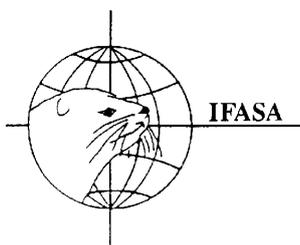
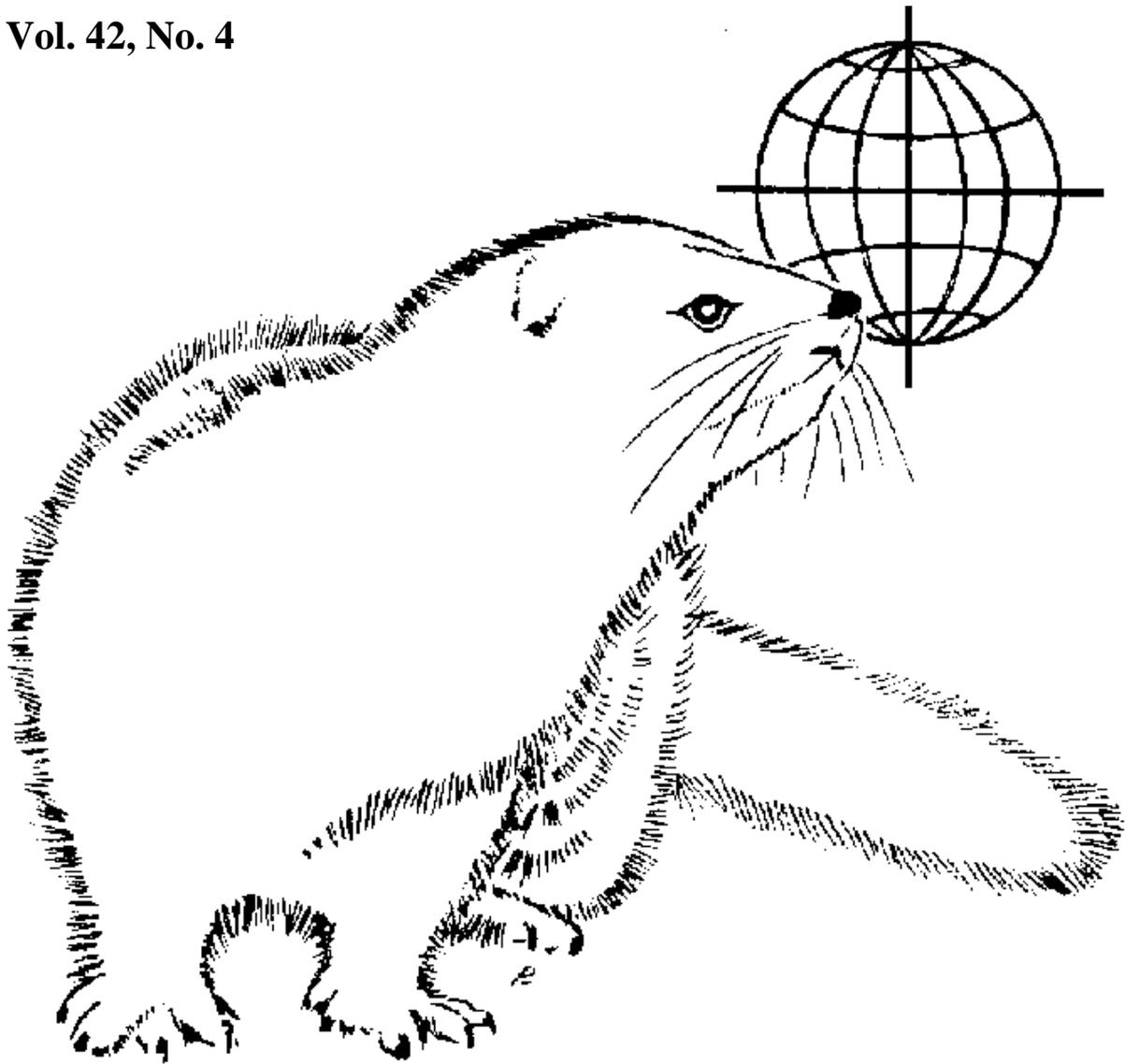


# SCIENTIFUR

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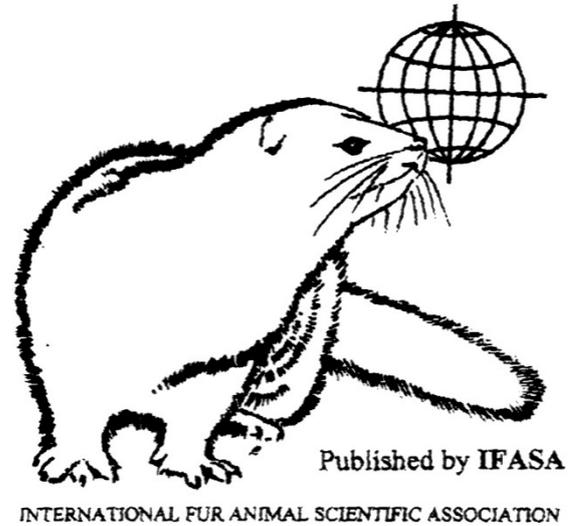
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## Notes from the Editor

On September 13 2018, the Rise Foundation (<http://www.risefoundation.eu/>) launched the report: “What is the Safe Operating Space for EU livestock?” (<http://www.risefoundation.eu/publications>)

The report deals with cattle, sheep, pigs and poultry production and meat, dairy and egg consumption and focus on e.g. human nutrition, pasture utilization, climate protection, and nutrient flows. The report suggests structural changes in farming for a better balance in the production, improvement of resource efficiency and reduction of leakage and waste. The report questions the present livestock production and its size.

While livestock production has been less exposed to criticisms hitherto, fur animal production has for a long time been challenged primarily due to welfare issues. Fur animal production faces environmental, economic and social challenges as well as the livestock production but has a large potential for sustainability and to be a part of the circular bioeconomy.

The final product, the fur, is highly durable. A mink coat can last for 40 years. With new fashion or a new owner, it is possible to redesign the fur coat. Finally, when not to be used anymore, the fur is biodegradable. Such sustainability should be considered in the fashion industry, which increasingly avoids fur in the production and which in opposition to sustainability focus on frequent launching of new collections and use of fake fur with a larger environmental footprint.

I am glad to publish in this issue of Scientifur an abstract from a master thesis in Agrobiography “Temperament in mink: the effect of coat colour, housing system and time” submitted and defended at Aarhus University in 2018.

Vivi Hunnicke Nielsen

Editor Scientifur



**BREEDING, GENETICS AND REPRODUCTION****Genomic responses to selection for tame/aggressive behaviors in the silver fox (*Vulpes vulpes*)**

Wang X.<sup>1,2</sup>, Pipes L.<sup>3</sup>, Trut L.N.<sup>4</sup>, Herbeck Y.<sup>4</sup>, Vladimirova A.V.<sup>4</sup>, Gulevich R.G.<sup>4</sup>, Kharlamova A.V.<sup>4</sup>, Johnson J.L.<sup>5</sup>, Acland G.M.<sup>6</sup>, Kukekova A.V.<sup>7</sup>, Clark A.G.<sup>1</sup>

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Animal domestication efforts have led to a shared spectrum of striking behavioral and morphological changes. To recapitulate this process, silver foxes have been selectively bred for tame and aggressive behaviors for more than 50 generations at the Institute for Cytology and Genetics in Novosibirsk, Russia. To understand the genetic basis and molecular mechanisms underlying the phenotypic changes, we profiled gene expression levels and coding SNP allele frequencies in two brain tissue specimens from 12 aggressive foxes and 12 tame foxes. Expression analysis revealed 146 genes in the prefrontal cortex and 33 genes in the basal forebrain that were differentially expressed, with a 5% false discovery rate (FDR). These candidates include genes in key pathways known to be critical to neurologic processing, including the serotonin and glutamate receptor pathways. In addition, 295 of the 31,000 exonic SNPs show significant allele frequency differences between the tame and aggressive populations (1% FDR), including genes with a role in neural crest cell fate determination.

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**Red fox genome assembly identifies genomic regions associated with tame and aggressive behaviours**

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Strains of red fox (*Vulpes vulpes*) with markedly different behavioural phenotypes have been developed in the famous long-term selective breeding programme known as the Russian farm-fox experiment. Here we sequenced and assembled the red fox genome and re-sequenced a subset of foxes from the tame, aggressive and conventional farm-bred populations to identify genomic regions associated with the response to selection for behaviour. Analysis of the re-sequenced genomes identified 103 regions with either significantly decreased heterozygosity in one of the three populations or increased divergence between the populations. A strong positional candidate gene for tame behaviour was highlighted: SorCS1, which encodes the main trafficking protein for AMPA glutamate receptors and neurexins and suggests a role for synaptic plasticity in fox domestication. Other regions identified as likely to have been under selection in foxes include genes implicated in human neurological disorders, mouse behaviour and dog domestication. The fox represents a powerful model for the genetic analysis of affiliative and aggressive behaviours that can benefit genetic studies of behaviour in dogs and other mammals, including humans.

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### **Construction of Red Fox Chromosomal Fragments from the Short-Read Genome Assembly**

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The genome of a red fox (*Vulpes vulpes*) was recently sequenced and assembled using next-generation sequencing (NGS). The assembly is of high quality, with 94X coverage and a scaffold N50 of 11.8 Mbp, but is split into 676,878 scaffolds, some of which are likely to contain assembly errors. Fragmentation and misassembly hinder accurate gene prediction and downstream analysis such as the identification of loci under selection. Therefore, assembly of the genome into chromosome-scale fragments was an important step towards developing this genomic model. Scaf-

folders from the assembly were aligned to the dog reference genome and compared to the alignment of an outgroup genome (cat) against the dog to identify syntenic sequences among species. The program Reference-Assisted Chromosome Assembly (RACA) then integrated the comparative alignment with the mapping of the raw sequencing reads generated during assembly against the fox scaffolds. The 128 sequence fragments RACA assembled were compared to the fox meiotic linkage map to guide the construction of 40 chromosomal fragments. This computational approach to assembly was facilitated by prior research in comparative mammalian genomics, and the continued improvement of the red fox genome can in turn offer insight into canid and carnivore chromosome evolution. This assembly is also necessary for advancing genetic research in foxes and other canids.

### RACA-Selected Scaffolds

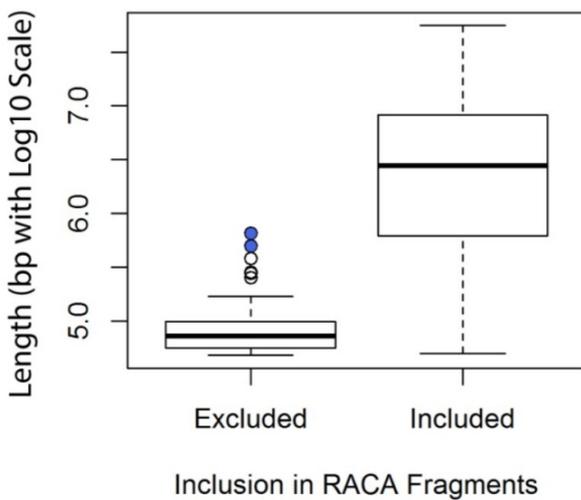
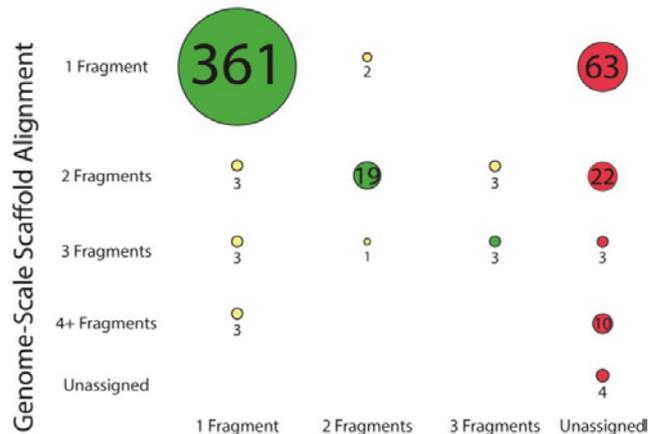


Fig. 1. Box plot indicating lengths of scaffolds included and excluded from Reference-Assisted Chromosome Assembly's (RACA's) assembled fragments. RACA was provided with the largest 500 scaffolds, which ranged from 48 to 55,683 Kbp. The scaffolds included in the assembly spanned approximately the full range of sizes (50 to 55,683 Kbp) whereas the excluded scaffolds tended to be smaller (48 to 656 Kbp). The outlier points shaded in blue represent scaffolds 292 and 310, which were experimentally demonstrated to contain red fox Y-chromosome sequence [], and therefore would not be expected to be included in RACA's assembled fragments.

### Syntenicity of Scaffolds to Dog Chromosomes



### RACA Syntenic Analysis

Fig. 2. The number of distinct dog chromosomes syntenic to each scaffold, as predicted with two complementary methods: genome-scale alignment to identify syntenic dog chromosome(s) for each scaffold [12], and the number of dog chromosomes syntenic to each scaffold as identified by RACA. Circle diameter is proportional to the number of scaffolds, which is presented inside of or beside the circle. Green indicates that the same number of syntenic chromosomes was predicted by both methods; yellow that the methods predicted different numbers; and red that the scaffolds were excluded from the RACA assembly. Unlike the prior analysis, RACA did not compare the scaffolds to the dog Y-chromosome.

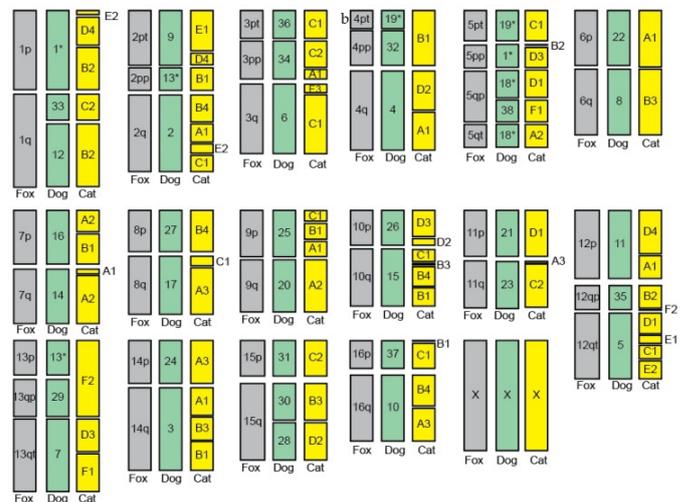


Fig. 3. The fox chromosomal segments shown alongside the corresponding syntenic chromosomes in dog and cat. Fox chromosomal fragments are ordered to approximate the full assembled chromosome, and chromosome numbers appear inside of or alongside fragments. Asterisks indicate dog chromosomes that map in more than one syntenic block to fox. Dog and fox fragments are to scale; cat fragments are approximately to scale. Syntenic blocks smaller than 500 Kbp (found only for the cat) are not shown.

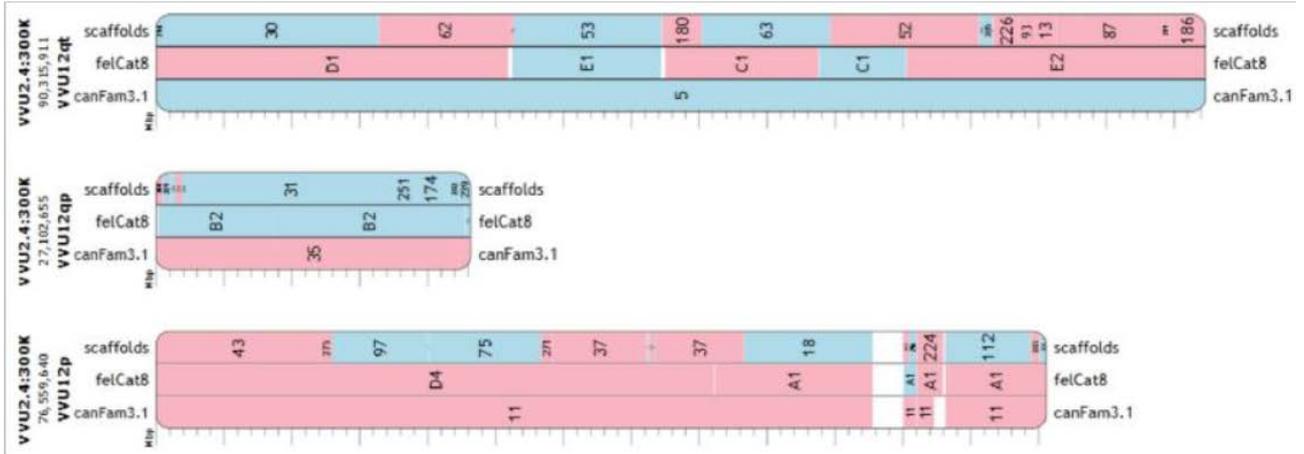


Fig. 4. View of fox, dog, and cat synteny along fox chromosome 12. Interspecies chromosome-scale synteny has been visualized in Evolution Highway [61]. The fox scaffolds comprising vv2.2 and the final 40 fragments comprising vv2.4 are included in the visualization. Red represents fragments that run negative relative to the fox chromosome fragments (i.e., opposite strand), whereas blue fragments run in the same direction.

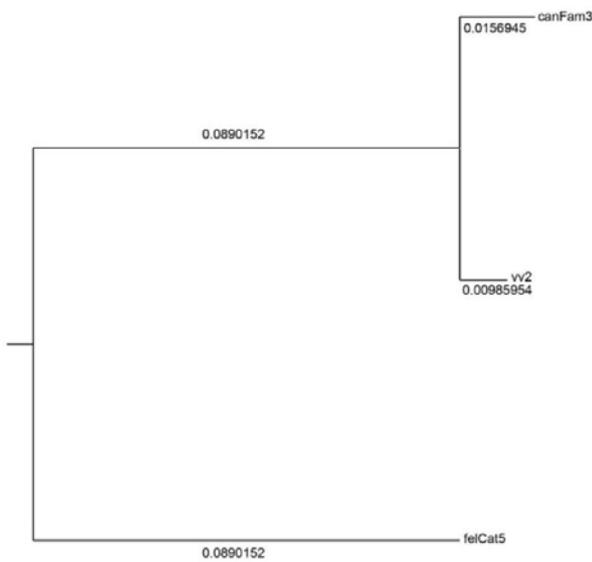


Fig. A1. Newick tree visualization including branch lengths.

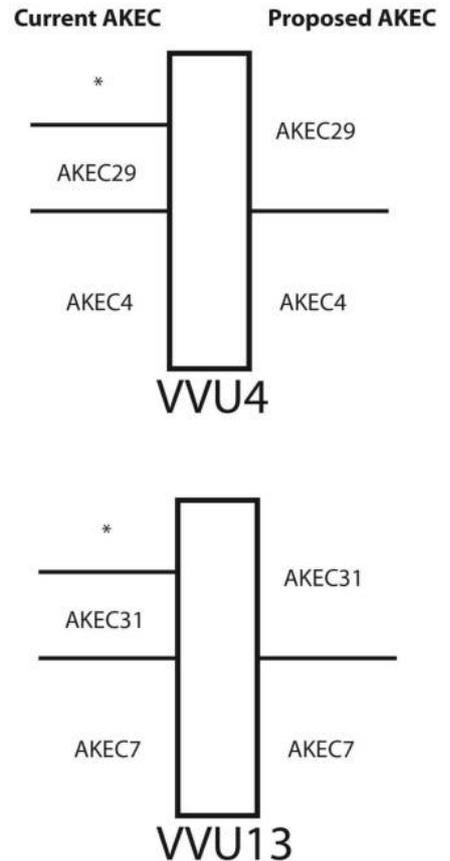


Fig. A2. Proposed updates to the Ancestral Karyotype of Extant Canids (AKEC). Asterisks denote regions of the fox chromosomes where the ancestral chromosome number was unknown in [39]. The original AKEC underestimates the coverage of VVU4 and VVU13 by the previously identified homologous ancestral chromosomes. The results of comparative genome mapping support a simpler homology between the ancestral chromosomes and the modern vulpine chromosomes, as indicated to the right.

*Genes (Basel)*. 2018 Jun 20;9(6). pii: E308.  
Doi: 10.3390/genes9060308.

### **Nuclear Genetic Analysis of the Red Fox across its Trans-Pacific Range**

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The red fox (*Vulpes vulpes*) occurs on multiple continents in diverse habitats, making it an informative system for evolutionary genomic research. However, its phylogeography remains unclear. Previously, mitochondrial DNA and small numbers of nuclear loci provided discordant views. Both markers indicated deep divergence (~ 0.5 million years [MY]) between Eurasian and southern North American populations but differed in the apparent continental affinity of Alaskan red foxes, implying some degree of gene exchange during secondary contact (~0.1 MY). We assayed >173000 nuclear genomic sites in 52 red foxes, along with 2 Rueppell's foxes (*Vulpes rueppellii*) and a gray wolf (*Canis lupus*) using the Illumina CanineHD BeadChip. We obtained 5107 single nucleotide polymorphisms (SNPs) in the foxes. Consistent with the Afro-Eurasian origins of red foxes, genetic diversity was higher in Eurasian than North American samples. Phylogenetic trees indicated that Alaskan and southern North American red foxes formed a monophyletic group nested within the Eurasian clade. However, admixture models suggested Alaskan red foxes contained up to 40% Eurasian ancestry. We hypothesize that North American red foxes either hybridized with Eurasian foxes in Beringia at the start of the last glaciation or merged with a Beringian population after the last glaciation. Future work is needed to test between these scenarios and assess speciation.

*J Hered*. 2018 Jun 27; 109(5):573-584.  
Doi: 10.1093/jhered/esy028.

### **Sequencing of Supernumerary Chromosomes of Red Fox and Raccoon Dog Confirms a Non-Random Gene Acquisition by B Chromosomes**

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B chromosomes (Bs) represent a variable addition to the main karyotype in some lineages of animals and plants. Bs accumulate through non-Mendelian inheritance and become widespread in populations. Despite the presence of multiple genes, most Bs lack specific phenotypic effects, although their influence on host genome epigenetic status and gene expression are recorded. Previously, using sequencing of isolated Bs of ruminants and rodents, we demonstrated that Bs originate as segmental duplications of specific genomic regions, and subsequently experience pseudogenization and repeat accumulation. Here, we used a similar approach to characterize Bs

of the red fox (*Vulpes vulpes* L.) and the Chinese raccoon dog (*Nyctereutes procyonoides procyonoides* Gray). We confirm the previous findings of the *KIT* gene on Bs of both species, but demonstrate an independent origin of Bs in these species, with two reused regions. Comparison of gene ensembles in Bs of canids, ruminants, and rodents once again indicates enrichment with cell-cycle genes, development-related genes, and genes functioning in the neuron synapse. The presence of B-chromosomal copies of genes involved in cell-cycle regulation and tissue differentiation may indicate importance of these genes for B chromosome establishment.

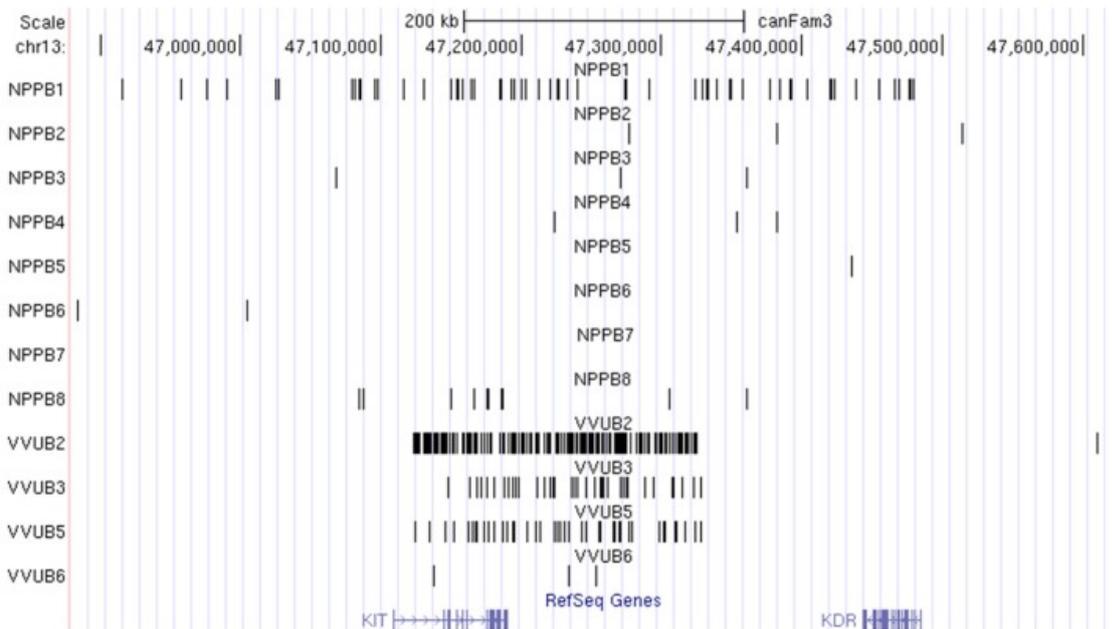


Fig. 1. The region encompassing protooncogene *KIT* is present on B chromosomes (Bs) of the fox (VVUB2, 3, 5 and 6) and raccoon dog (NPPB1-8) visualized in UCSC genome browser (<http://genome.ucsc.edu/>). Coordinates are given for the dog (CanFam3.1) genome.

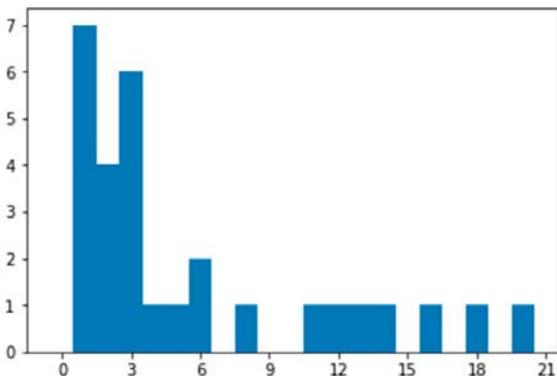


Fig. 2. Numbers of additional copies for B chromosomal regions (identified by isolated chromosome sequencing) estimated based on whole-genome sequencing of the red fox individual with three Bs. Region parts with different copy numbers counted separately. Regions with copy number below three were lost from some of Bs, while regions with copy number above three were amplified within Bs. X—number of additional copies, Y—counts of regions.

*Genes (Basel)*. 2018 Aug 10; 9(8). pii: E405.  
Doi: 10.3390/genes9080405.

### Determination of the Genetic Component of Fur-Chewing in Chinchillas (*Chinchilla lanigera*) and Its Economic Impact

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Fur-chewing is a common behavioral disorder developed by chinchillas kept in confinement that can indicate a past or present welfare problem. It also has a negative productive impact associated. The aim of this study was to determine the genetic component of fur-chewing, and the effect of this undesired behavior on fur price in a commercial fur-farming system of chinchillas (*Chinchilla lanigera*). The data for the analysis was derived from a commercial population of 10,196 chinchillas, recorded between the years 1990 and 2011. For determining differences in fur price according to presence of fur-chewing behavior, analysis of variance (ANOVA) was used, considering 3007 animals. For estimation of variance components of fur-chewing a sire-dam threshold (probit) mixed model was used, using data of 9,033 individuals, and then heritability on the underlying liability scale was calculated. The analysis revealed a significant negative impact on fur price from fur-chewing chinchillas ( $p$ -value < 0.05). In addition, the study showed that fur-chewing presents significant genetic variation, with an estimated heritability of 0.16. The presentation of fur-chewing should be taken into account when selecting broodstock in these systems, in order to reduce the number of affected individuals.

*Animals (Basel)*. 2018 Aug 21; 8(9). pii: E144.  
Doi: 10.3390/ani8090144.

### New Insights into the Melanophilin (*MLPH*) Gene Affecting Coat Color Dilution in Rabbits

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Coat color dilution corresponds to a specific pigmentation phenotype that leads to a dilution of wild type pigments. It affects both eumelanin and pheomelanin containing melanosomes. The mode of inheritance of the dilution phenotype is autosomal recessive. Candidate gene approaches focused on the melanophilin (*MLPH*) gene highlighted two variants associated with the dilution phenotype in rabbits: The c.111-5C>A variant that is located in an acceptor splice site or the c.585delG variant, a frameshift mutation. On the transcript level, the skipping of two exons has been reported as the molecular mechanism responsible for the coat color dilution. To clarify, which of the two variants represents the causal variant, (i) we analyzed their allelic segregation by genotyping Castor and Chinchilla populations, and (ii) we evaluated their functional effects on the stability of *MLPH* transcripts in skin samples of animals with diluted or wild type coat color. Firstly, we showed that the c.585delG variant showed perfect association with the dilution phenotype in contrast to the intronic c.111-5C>A variant. Secondly, we identified three different *MLPH* isoforms including the wild type isoform, the exon-skipping isoform and a retained intron isoform. Thirdly, we observed a drastic and significant decrease of *MLPH* transcript levels in rabbits with a coat color dilution ( $p$ -values ranging from  $10^{-03}$  to  $10^{-06}$ ). Together, our results bring new insights into the coat color dilution trait.

*Genes (Basel)*. 2018 Aug. 23; 9(9). pii: E430.  
Doi: 10.3390/genes9090430.

### Research advances on embryonic diapause in mammals

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Embryonic diapause occurs in over 130 species of mammals. In embryonic diapause, there is a reversible cessation phase in embryo development and subsequent reactivation and continuation of development with no adverse effects. This allows animals to prolong gestation and give birth to offspring at a more favorable time from a survival perspective. Embryonic diapause is divided into two types: facultative diapause which is induced by physiological conditions and obligate diapause which is induced by season of the year. The process of embryonic diapause includes three phases: onset, maintenance, and reactivation. With facultative diapause the suckling stimulus promotes secretion of prolactin, which suppresses development and secretion activity of the corpus luteum (CL). The decrease of progesterone, secreted by CL, subsequently results in embryos in the diapause state. The embryonic diapause that occurs in mink is of the obligate type and is regulated by photoperiod. The short daylight (<12 h) before the vernal equinox induces an increase in the release of melatonin from the pineal gland. The secretion of prolactin is suppressed and the production of progesterone from CL is reduced. During these phases, many others factors that regulate embryonic and uterine status, hormonal profile, polyamines, and uterine secretion, have been monitored. In recent times, the understanding of the role of different molecular events in embryonic diapause has been advanced. The present review summarizes the research advances related to embryonic diapause to enhance the understanding of this phenomenon and to be considered when there are future research endeavors related to embryonic diapause.

*Anim Reprod Sci.* 2018 Sep 20. pii: S0378-4320(18)30611-0.

Doi: 10.1016/j.anireprosci.2018.09.009.

Epub ahead of print.

### Collection and frozen storage of semen for artificial insemination in red foxes (*Vulpes vulpes*)

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This study was conducted on red foxes to determine the appropriate voltage in electroejaculation for semen collection from stud males, and to confirm whether frozen semen with bovine semen extender can be used for artificial insemination. The proper load voltage for electroejaculation was 3-4 V based on semen collection rates and concentrations of spermatozoa. Frozen semen was prepared according to the known procedure for cows. In frozen-thawed semen, a relatively high conception rate (81.3%) was obtained in vixens, in which the optimum insemination time was detected by vaginal electrical resistance. These findings demonstrated that the restricted condition for semen collection by electroejaculation with cryopreservation of semen using bovine semen extender can be applied to artificial insemination of red foxes.

*J Vet Med Sci.* 2018 Sep 21.

Doi: 10.1292/jvms.17-0433. Epub ahead of print.

### Infrared technology for estrous detection in *Chinchilla lanigera*

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Thermography is one of the most modern methods for non-invasive monitoring of animal behavior, health status and physiological state. The scarcity of scientific reports related to this subject in fur animals prompted the use of thermography in *Chinchilla lanigera*. The aim of the study was to evaluate the use of a thermal imaging camera to diagnose or confirm estrus in *Chinchilla lanigera*. The subjects included 100 female chinchillas from three different breeds, selected during ovulation after parturition, which were evaluated twice: measurements were taken over three successive days starting from the 2<sup>nd</sup> day post partum and again through the same time period 6 weeks later when the females were ready to mate. A picture of the external genital area was taken with a FLIR C2 thermal imaging camera and the temperature was determined and recorded from the selected area using the FLIR TOOLS computer program. There were distinct differences between chinchillas with and without a confirmed estrus. Chinchillas in estrus had ROI temperatures that were greater (2.33 °C) than non-estrous females. During estrus, the temperature of the external genital area clearly increases, which could be due to genital hyperemia. This fact can be used for non-invasive estrous diagnosis, which is especially important because of the absence of typical estrous behavior in this species. Monitoring the greater surface temperature of the vulval area allows determination of the optimal time to place a female with a male and expect effective copulation.

*Anim Reprod Sci.* 2018 Oct; 197:81-86.  
Doi: 10.1016/j.anireprosci.2018.08.012.  
Epub 2018 Aug 16.

## BEHAVIOUR AND WELFARE

### Revisiting the famous farm foxes: A psychological perspective

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Five decades ago, Dmitry Belyaev, Lyudmila Trut, and colleagues began a now-famous experiment, selectively breeding foxes based on one criterion: per-

ceived tame behavior. Over generations, the fox population changed in behavior (as predicted) but, intriguingly, also changed markedly in appearance—for example, many had wider mouths, curlier tails, different fur coloring, and floppy ears. These researchers concluded that the morphological changes that appeared in their foxes were a by-product of the researchers' selecting for genetic variants that are implicated both in behavior and in appearance. For decades, scientists have largely accepted this "shared genetic variants" interpretation to fully account for the co-occurrence of behavioral and morphological phenotypes in these foxes and in other domesticated animals. However, several decades of psychological research on human social cognition, human-canine interaction, and canine behavior strongly suggest that such an account may be incomplete. I forward a supplementary perspective, based on psychological research, that the covariation of appearance and behavior among these foxes may be partly an artifact of human psychological processes at play in selection. These processes include humans' tendency to infer individuals' traits based on their physical features; trait inferences, in turn, influence how humans treat those individuals. If accurate, this account bears on our understanding of these famous foxes, human-canine interactions, as well as humans' role in domestication.

*Learn Behav.* 2018 Jul 2.  
Doi: 10.3758/s13420-018-0333-2.  
Epub ahead of print.

## NUTRITION, FEEDING AND MANAGEMENT

### Epigallocatechin-3-Gallate Promotes the Growth of Mink Hair Follicles Through Sonic Hedgehog and Protein Kinase B Signaling Pathways

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**Background**

Hair follicles play an essential role in the growth of hair. Epigallocatechin-3-gallate (EGCG), a catechin polyphenol in green tea, has various bioactivities. The present study aims to evaluate the effect of EGCG on the growth of mink hair follicles and investigate the possible molecular mechanisms.

**Methods**

The length of hair follicles was recorded up to 6 days in presence of 0.1-5  $\mu\text{M}$  EGCG. Primary dermal papilla cells (DPCs) and outer root sheath cells (ORSCs) were treated with 0.25-4  $\mu\text{M}$  EGCG, and their growth was evaluated by MTT assay and cell cycle detection. The levels of key molecules in sonic hedgehog (Shh) and protein kinase B (AKT) signaling pathways were further assessed by quantitative real-time PCR, western blot and immunofluorescence. To determine the involvement of Shh and AKT pathways in EGCG-mediated growth-promotion of ORSCs and DPCs, Shh pathway inhibitors cyclopamine and GANT61 or AKT pathway inhibitor

LY294002 were employed, and then cell proliferation and cell cycle were analyzed.

**Results**

Data from *ex vivo* culture showed that, in presence of 0.5-2.5  $\mu\text{M}$  EGCG, the growth of mink hair follicles was promoted. *In vitro*, the proliferation of DPCs and ORSCs was enhanced by 0.5-4  $\mu\text{M}$  EGCG treatment. More cells entered S phase upon treatment of EGCG, accompanied with upregulation of cyclin D1 and cyclin E1. Furthermore, when exposed to EGCG, the Shh and AKT signaling pathways were activated in both hair follicles and primary DPCs and ORSCs. Inhibiting either of these two pathways partly reversed the effect of EGCG on proliferation and cell cycle of DPCs and ORSCs.

**Conclusion**

EGCG promotes the growth of mink hair follicles at concentrations of 0.5-2.5  $\mu\text{M}$ . This growth-promoting effect of EGCG may be associated with the increased proliferation of DPCs and ORSCs through activating Shh and AKT signaling pathways.

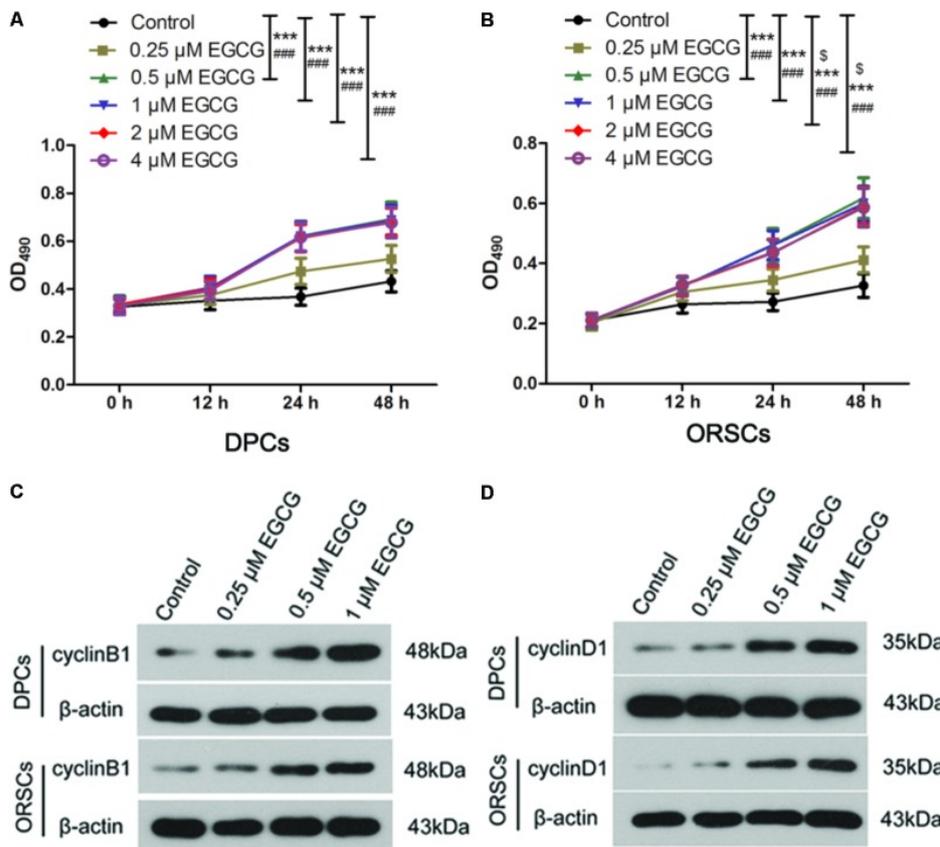


Fig. 1. Epigallocatechin-3-gallate (EGCG) accelerates the growth of DPCs and ORSCs. (A,B) After treatment with 0.25, 0.5, 1, 2, and 4  $\mu\text{M}$  EGCG for 12, 24, and 48 h, the cell viability of DPCs and ORSCs was assessed by MTT as-

say.  $^S p < 0.05$  for 12 h,  $^{***} p < 0.001$  for 24 h,  $^{###} p < 0.001$  for 48 h, all compared with the control group. (C) After treatment with different concentrations of EGCG, the protein level of cyclinB1 in DPCs and ORSCs was detected by western blot.  $\beta$ -actin served as the internal reference. (D) Western blot was performed to detect the protein level of cyclinD1 in DPCs and ORSCs after treatment with EGCG. All experiments were repeated three times. The results are presented as mean  $\pm$  SD.

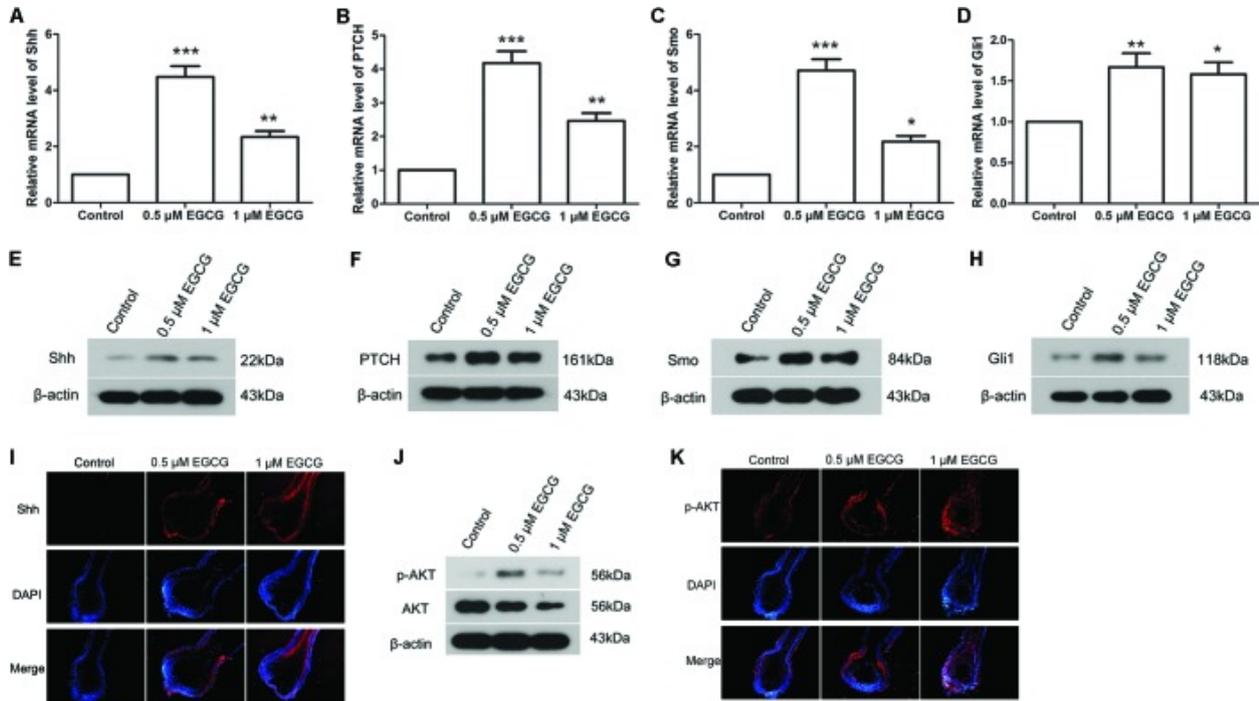


Fig. 2. Epigallocatechin-3-gallate activates the Shh and AKT signaling pathways in hair follicles. After treatment with 0.5 and 1  $\mu$ M EGCG, the mRNA levels of Shh (A), PTCH (B), Smo (C), and Gli1 (D) in hair follicles were detected by qRT-PCR. The relative mRNA levels were calculated using  $2^{-\Delta\Delta C_t}$  method. Western blot was also performed to detect the protein levels of Shh (E), PTCH (F), Smo (G), and Gli1 (H) in hair follicles.  $\beta$ -actin served as the internal reference. (I) Level of Shh in hair follicles was assessed by immunofluorescence. Red fluorescence: Shh; blue fluorescence: DAPI. (J) The phosphorylation level of AKT was assessed by western blot. (K) Level of p-AKT in hair follicles was assessed by immunofluorescence. Red fluorescence: p-AKT; blue fluorescence: DAPI. Each experiment was repeated three times and the results are presented as mean  $\pm$  SD.  $^* p < 0.05$ ,  $^{**} p < 0.01$ ,  $^{***} p < 0.001$  compared with the control group.

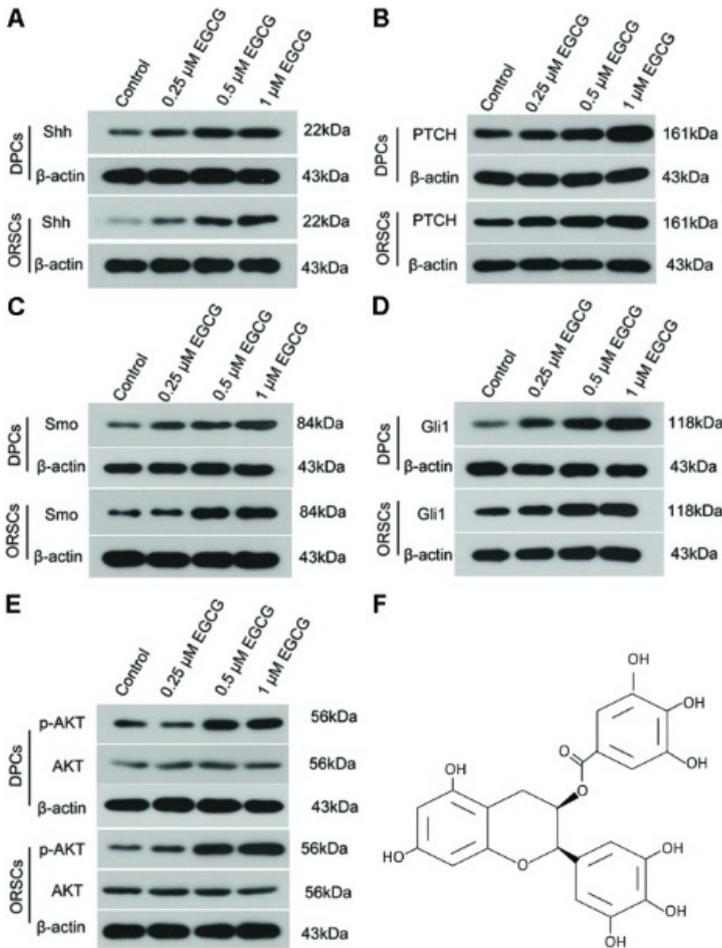
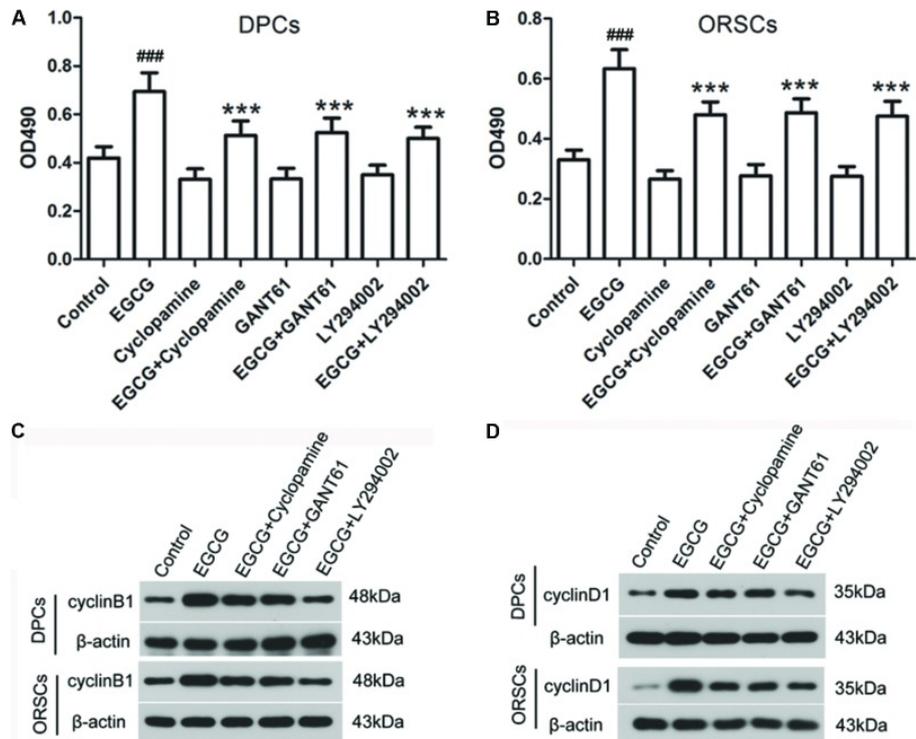


Fig. 3. Epigallocatechin-3-gallate activates the Shh and AKT signaling pathways in DPCs and ORSCs. Upon treatment with EGCG, the protein levels of Shh (A), PTCH (B), Smo (C), and Gli1 (D) in DPCs and ORSCs were detected by western blot with β-actin as the internal reference. (E) Western blot was performed to assess the levels of AKT and p-AKT in each group with β-actin as the internal reference. (F) Chemical structure of EGCG. Each experiment was repeated three times.

Fig. 4. Shh and AKT signaling pathway inhibitors abolish the effect of EGCG on the growth of DPCs and ORSCs. (A,B) After treatment with EGCG and/or cyclopamine, GANT61 or LY294002, the cell viability of DPCs and ORSCs was assessed by MTT assay. (C,D) Protein levels of cyclinB1 and cyclinD1 in DPCs and ORSCs were assessed by western blot with β-actin as the internal reference. All experiments were repeated three times. The results are presented as mean ± SD. ###*p* < 0.001 compared with the control group; \*\*\**p* < 0.001 compared with the EGCG group.



Front Pharmacol. 2018 Jun 26; 9:674.

Doi: 10.3389/fphar.2018.00674. eCollection 2018.

**Review article: Role of satiety hormones in anorexia induction by trichothecene mycotoxins**

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The trichothecenes, produced by *Fusarium*, contaminate animal feed and human food in all stages of production and lead to a large spectrum of adverse effects for animal and human health. An hallmark of trichothecenes toxicity is the onset of emesis followed by anorexia and food intake reduction in different animal species (mink, mice and pig). The modulation of emesis and anorexia can result from a direct action of trichothecenes in the brain or from an indirect action in the gastrointestinal tract. The direct action of trichothecenes involved specific brain areas such as nucleate tractus solitarius in the brainstem and the arcuate nuclei in the hypothalamus. Activation of these areas in the brain leads to the activation of specific neuronal populations containing anorexigenic factors (POMC and CART). The indirect action of trichothecenes in the gastrointestinal tract involved, by enteroendocrine cells, the secretion of several gut hormones such as cholecystokinin (CCK) and peptide YY (PYY) but also glucagon-like peptide 1 (GLP-1), gastric inhibitory peptide (GIP) and 5-hydroxytryptamine (5-HT), which transmitted signals to the brain via the gut-brain axis. This review summarizes current knowledge on the effects of trichothecenes, especially deoxynivalenol, on emesis and anorexia and discusses the mechanisms underlying trichothecenes-induced food reduction.

*Food Chem Toxicol.* 2018 Sep 20. pii: S0278-6915(18)30678-1.

Doi: 10.1016/j.fct.2018.09.034.

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**Mandibular shape in farmed Arctic foxes (*Vulpes lagopus*) exposed to persistent organic pollutants**

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We investigated if dietary exposure to persistent organic pollutants (POPs) affect mandibular asymmetry and periodontal disease in paired male-siblings of Arctic foxes (*Vulpes lagopus*). During ontogeny, one group of siblings was exposed to the complexed POP mixture in naturally contaminated minke whale (*Balaenoptera acutorostata*) blubber (n = 10), while another group was given wet feed based on pig (*Sus scrofa*) fat as a control (n = 11). The  $\Sigma$ POP concentrations were 802 ng/g ww in the whale-based feed compared to 24 ng/g ww in the control diet. We conducted a two-dimensional geometric morphometric (GM) analysis of mandibular shape and asymmetry in the foxes and compared the two groups. The analyses showed that directional asymmetry was higher than fluctuating asymmetry in both groups and that mandibular shape differed significantly between the exposed and control group based on discriminant function analysis ( $T^2 = 58.52$ ,  $p = 0.04$ , 1000 permutations). We also found a non-significantly higher incidence of periodontal disease (two-way ANOVA:  $p = 0.43$ ) and greater severity of sub-canine alveolar bone deterioration similar to periodontitis (two-way ANOVA:  $p = 0.3$ ) in the POP-exposed group. Based on these results, it is possible that dietary exposure to a complexed POP mixture lead to changes in jaw morphology in Arctic foxes. This study suggests that

extrinsic factors, such as dietary exposure to POPs, may affect mandibular shape and health in a way that could be harmful to wild Arctic populations. Therefore, further studies using GM analysis as an alternative to traditional morphometric methods should be conducted for wild Arctic fox populations exposed to environmental contaminants.

*Sci Total Environ.* 2019 Jan 1; 646:1063-1068.  
 Doi: 10.1016/j.scitotenv.2018.07.367.  
 Epub 2018 Jul 26.

### The use of phoxim and bendiocarb for control of fleas in farmed mink (*Mustela vison*)

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#### Background

Fleas (*Ceratophyllus sciurorum*) are common on farmed mink in Denmark. When present, the fleas have a negative impact on the health of the farmed mink and are of nuisance for farm staff. Severe infestations of fleas cause anemia, poor growth and may result in death of mink kits. Changed behavior of the dams is also observed. Further it has been demonstrated that the fleas are vectors of Aleutian disease virus. Flea control is based on use of a few insecticides and resistance has been reported against permethrin. There is thus a need for new flea control products. In this blinded, randomized clinical trial according to GCP standard, phoxim spray and bendiocarb powder for flea control on mink farms were investigated.

#### Results

Both the phoxim spray solution and bendiocarb powder were found to be efficient for the control of *C. sciurorum* fleas on farmed mink. Phoxim treatments reduced the number of fleas by 98.4% and the bendiocarb treatments reduced the number of fleas by 99.0% in the mink nest boxes when compared to counts in controls. No clinical signs were observed post treatment.

### Conclusions

The study demonstrated that phoxim sprayed on the animals and the use of bendiocarb powder in the nest box material were highly efficient for the control of the *C. sciurorum* fleas on farmed mink. Both products were safe to use at the recommended dose rate. Both compounds are recommended to be integrated in a new farm management plan suggested here.

*Acta Vet Scand.* 2018 Sep 22; 60(1):58.  
 Doi: 10.1186/s13028-018-0412-6.

### Preservation of fatty acid signatures in three vertebrate species after six months of storage at various temperatures

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Fatty acid (FA) signatures (FAS) are important tools to assess the foraging ecology of wild animals. The present study was conducted to assess how well the general FAS and the proportions of individual FA are preserved in fat samples stored at different temperatures (-196, -80, -20, +4 and +20°C). Using three species (laboratory rat, American mink and rainbow trout), FAS were determined immediately upon sampling. Thereafter, eight subsamples per storage temperature from the inner part of the sample unaffected by oxygen and light were re-analyzed after 1, 2, 3, 7, 28, 84 and 168 days. Each time the remaining sample was sealed in its vial after replacing air with nitrogen gas. The results were tested with the mixed model and discriminant analyses. Generally, the FAS were well preserved regardless of storage temperature, and only a few major FA showed significant changes even after the 6-month period at room temperature. After an initial first-day change in proportions, presumably due to post-mortem enzymatic activities, the

remaining minor changes could not be clearly attributed to either further autolysis, decomposition or autoxidation. In the discriminant analysis, the species-specific differences dominated and remained distinct even after 6 months. Furthermore, the analysis mostly classified the samples preserved at sub- and above-freezing temperatures separate from each other, and the general deviation from the initial analysis results was present as early as after 1 day. If FAS are to be analyzed in a very precise manner, the analysis should be performed immediately upon sampling. However, FAS remain adequately reliable for long periods of time even without preservation in deep freeze, widening the availability of potential samples for studies on foraging ecology and related disciplines.

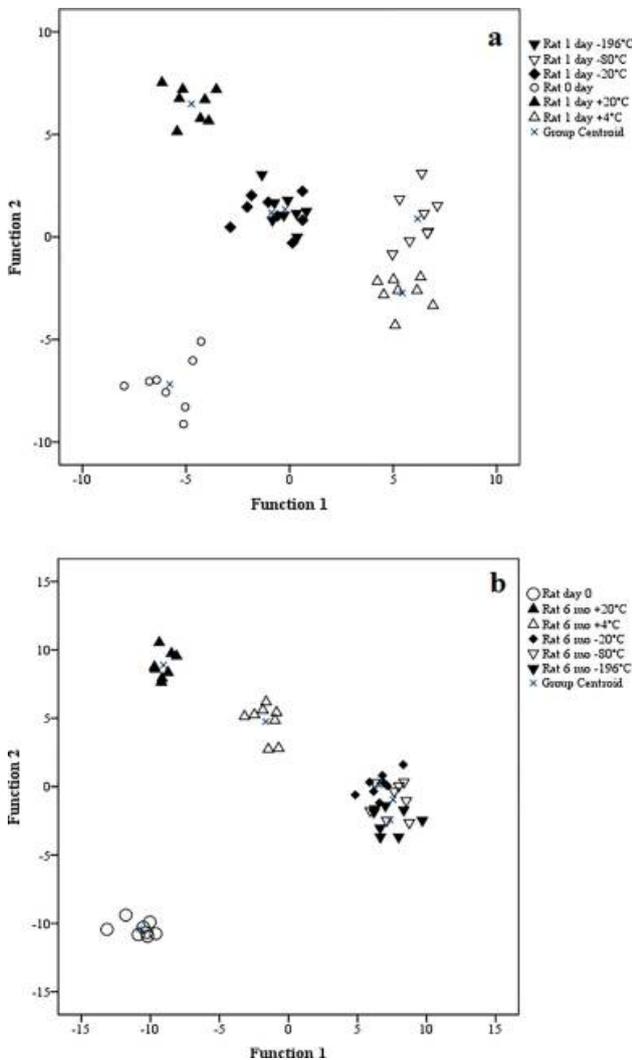


Fig. 1. Discriminant analysis of rat fatty acids at sampling and after 1 day (a) or 6 months (b) at different storage temperatures.

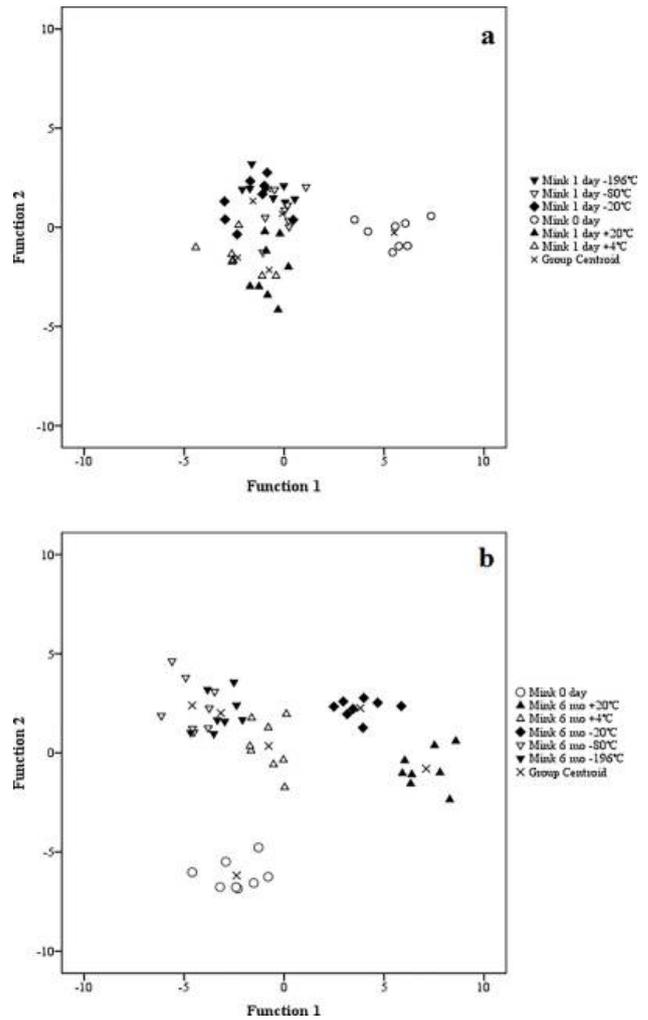


Fig. 2. Discriminant analysis of American mink fatty acids at sampling and after 1 day (a) or 6 months (b) at different storage temperatures.

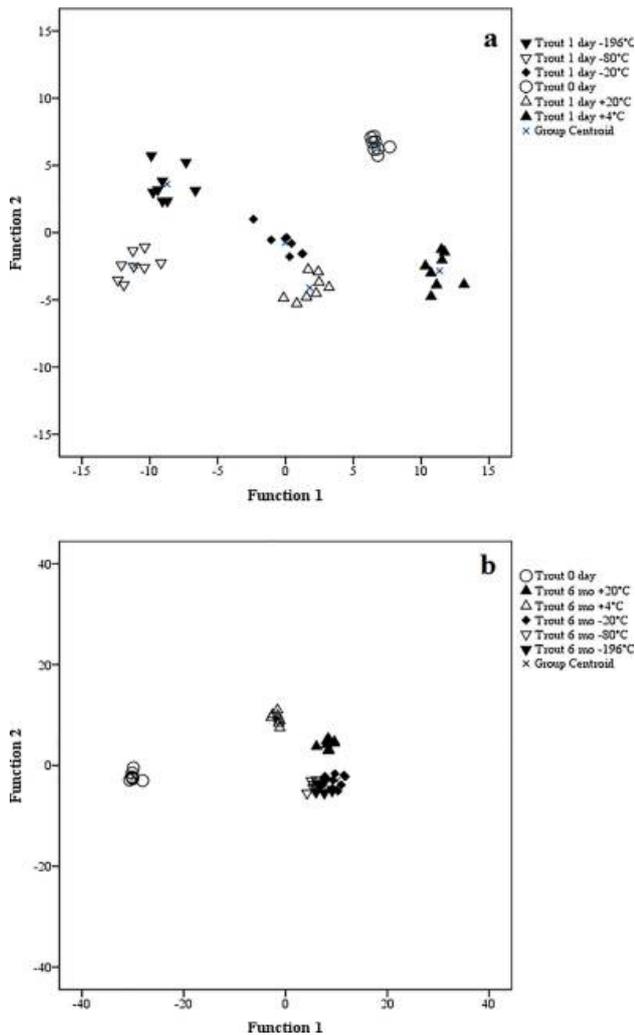


Fig. 3. Discriminant analysis of rainbow trout fatty acids at sampling and after 1 day (a) or 6 months (b) at different storage temperatures.

*PLoS One*. 2018 Sep 17; 13(9):e0204207.

Doi: 10.1371/journal.pone.0204207. eCollection 2018.

### Retinal photoreceptor and ganglion cell types and topographies in the red fox (*Vulpes vulpes*) and Arctic fox (*Vulpes lagopus*)

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The red fox (*Vulpes vulpes*) is the carnivore with the widest distribution in the world. Not much is known about the visual system of these predominantly forest-dwelling animals. The closely related Arctic fox (*Vulpes lagopus*) lives in more open tundra habitats. In search for corresponding adaptations, we examined the photoreceptors and retinal ganglion cells (RGCs), using opsin immunohistochemistry, lucifer yellow injections and Nissl staining. Both species possess a majority of middle-to-longwave-sensitive (M/L) and a minority of shortwave-sensitive (S) cones, indicating dichromatic color vision. Area centralis peak cone densities are 22,600/mm<sup>2</sup> in the red fox and 44,800/mm<sup>2</sup> in the Arctic fox. Both have a centro-peripheral density decrease of M/L cones, and a dorsoventrally increasing density of S cones. Rod densities and rod/cone ratios are higher in the red fox than the Arctic fox. Both species possess the carnivore-typical alpha and beta RGCs. The RGC topography shows a centro-peripheral density gradient with a distinct area centralis (mean peak density 7,900 RGCs/mm<sup>2</sup> in the red fox and 10,000 RGCs/mm<sup>2</sup> in the Arctic fox), a prominent visual streak of higher RGC densities in the Arctic fox, and a moderate visual streak in the red fox. Visual acuity and estimated sound localization ability were nearly identical between both species. In summary, the red fox retina shows adaptations to nocturnal activity in a forest habitat, while the Arctic fox retina is better adapted to higher light levels in the open tundra.

*J Comp Neurol*. 2018 Sep 1; 526(13):2078-2098.

Doi: 10.1002/cne.24493. Epub 2018 Jul 23.

### HEALTH AND DISEASE

#### Acinetobacter in Veterinary Medicine with emphasis on *A. baumannii*

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Acinetobacter spp. are aerobic, rod-shaped Gram-negative bacteria belonging to the Moraxellaceae family of the class Gammaproteobacteria and are considered ubiquitous organisms. Among them, *Acinetobacter baumannii* is the most clinically significant species with an extraordinary ability to accumulate antimicrobial resistance and survive in the hospital environment. Recent reports indicate that *A. baumannii* has also evolved into a veterinary nosocomial pathogen. Although *Acinetobacter* spp. can be identified to species level by the use of the matrix-assisted laser ionization time-of-flight mass spectrometry (MALDI-TOF MS) coupled with an updated database, molecular techniques are still necessary for genotyping and determination of clonal lineages. It seems that the majority of infections due to *A. baumannii* in veterinary medicine are nosocomial. Such isolates have been associated with several type of infections such as canine pyoderma, feline necrotizing fasciitis, urinary tract infections, equine thrombophlebitis and lower respiratory tract infections, foal sepsis, pneumonia in mink and cutaneous lesions in hybrid falcon. Given the potential multidrug resistance of *A. baumannii*, treatment of diseased animals is often supportive and should be based preferably on in vitro antimicrobial susceptibility testing. It should be noted that animal isolates show a high genetic diversity and are in general distinct in their sequence types and resistance patterns from those found in humans. However, it cannot be excluded that animals may occasionally play a role as reservoir for *A. baumannii*. In line, it is of importance to implement infection control measures in veterinary hospitals to avoid nosocomial outbreaks with multidrug-resistant *A. baumannii*.

*J Glob Antimicrob Resist.* 2018 Aug 22. pii: S2213-7165(18)30158-9.

Doi: 10.1016/j.jgar.2018.08.011.

Epub ahead of print.

### Lesser housefly (*Fannia canicularis*) as possible mechanical vector for Aleutian mink disease virus

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Flies are known vectors for a variety of infectious diseases in animals. In fur mink farming, one of the most severe diseases is Aleutian disease, which is caused by the Aleutian mink disease virus (AMDV). The presence of large fly populations is a frequent issue in mink farms; however, no studies assessing their role as AMDV carrier vectors have been conducted to-date. In order to determine the presence of AMDV in aerial flies from an infected mink farm, flies (n=254) and environmental swab samples (n=4) were collected from two farm barns. *Fannia canicularis* (L.) (Diptera: Muscidae) represented more than 99% of the fly population. One hundred and fifty specimens of this species were divided into pools of ten flies and analysed by qPCR, and positive samples were further sequenced. All fly pools and environmental samples tested positive for AMDV, and sequence analysis revealed identical genotypes in both types of samples. This is the first report of AMDV contamination in flies from mink farms, suggesting that *F. canicularis* may act as an AMDV vector. These results may be of interest for epidemiological studies and also for the improvement of control measures against this virus in mink farms.

*Vet Microbiol.* 2018 Jul; 221:90-93.

Doi: 10.1016/j.vetmic.2018.05.024.

Epub 2018 Jun 4.

**Molecular assessment of visitor personal protective equipment contamination with the Aleutian mink disease virus and porcine circovirus-2 in mink and porcine farms**

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Personal protective equipment (PPE) is an element of biosecurity intended to prevent the access or spread of diseases in farms. Nevertheless, to date no extensive reports exist about the effectiveness of different available PPE on farms. Thus, our aim was to estimate the degree of protection of PPE from viral contamination during farm visits. Two farms, infected with Aleutian mink disease virus and porcine circovirus-2 respectively, were visited by six visitors wearing different combinations of PPE: coveralls with hood and bootcovers, both with a certified barrier to infective agents (certified PPE group) and non-certified bootcover and coverall without hood (non-certified PPE group). Seventy-two swab samples from PPE and both hair and street clothes under PPE were taken after the visit and analysed by qPCR. Our results reveal viral exposure during visits, and the external protections of body and shoes were contaminated in all cases (24/24). In addition, protection from viral contamination varied noticeably according to the biosecurity elements used. A higher number of positives were detected in the non-certified PPE group than in the certified PPE group, both in elements under external protections (14/18 vs 3/18) and also in hair (4/6 vs 0/6). In fact, non-certified bootcovers broke during visits, resulting in viral contamination of the internal elements under them; these are consequently not suitable for using with wrinkled surfaces usually found in farm facilities. Thus, certified coveralls should be used in order to prevent contaminations, and workers and personnel of farms should be trained in their proper use. qPCR is a useful tool in the risk management of biosecurity programmes, and our results may serve as a model to evaluate different biosecurity measures.

*PLoS One*. 2018 Aug 27; 13(8): e0203144.

Doi: 10.1371/journal.pone.0203144.

eCollection 2018.

**Nonsuppurative (Aseptic) Meningoencephalomyelitis Associated with Neurovirulent Astrovirus Infections in Humans and Animals**

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Astroviruses are thought to be enteric pathogens. Since 2010, a certain group of astroviruses has increasingly been recognized, using up-to-date random amplification and high-throughput next-generation sequencing (NGS) methods, as potential neurovirulent (Ni) pathogens of severe central nervous system (CNS) infections, causing encephalitis, meningoencephalitis, and meningoencephalomyelitis. To date, neurovirulent astrovirus cases or epidemics have been reported for humans and domesticated mammals, including mink, bovines, ovines, and swine. This comprehensive review summarizes the virology, epidemiology, pathology, diagnosis, therapy, and future perspective related to neurovirulent astroviruses in humans and mammals, based on a total of 30 relevant articles available in PubMed (searched by use of the terms "astrovirus/encephalitis" and "astrovirus/meningitis" on 2 March 2018). A paradigm shift should be considered based on the increasing knowledge of the causality-effect association between neurotropic astroviruses and CNS infection, and attention should be drawn to the role of astroviruses in unknown CNS diseases.

*Clin Microbiol Rev*. 2018 Aug 29; 31(4). pii: e00040-18.

Doi: 10.1128/CMR.00040-18. Print 2018 Oct.

**Detection of Astrovirus, Rotavirus C, and Hepatitis E Viral RNA in Adult and Juvenile Farmed Mink (*Neovison vison*)**

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Mink astrovirus (MiAstV) is known to play a major role in mink pre-weaning diarrhea, and rotavirus and hepatitis E virus (HEV) are both considered potentially zoonotic agents. These viruses are not monitored in commercial mink, and the role of these viral infections in mink health is not well understood. This study assessed the prevalence of mink astrovirus, rotavirus C, mink HEV and swine HEV in 527 pooled healthy adult female mink and mink kit fecal samples from 50 Canadian mink farms in two seasons over 4 years. Viral RNA was extracted and amplified in RT-PCR to detect mink astrovirus and HEV RdRp genes, swine HEV ORF2, and rotavirus C VP6 gene. At least 26% of all positive samples for each virus was sequenced for phylogenetic analysis. Fourteen percent of samples were astrovirus positive, while 3 and 9% of samples were rotavirus C and mink HEV positive, respectively. One adult female sample was found to be positive by PCR for swine HEV. A significantly higher number of kit samples were astrovirus- and HEV-positive compared to adult female samples ( $p = 0.01$  and  $p < 0.0001$ , respectively). Astrovirus was detected in significantly more summer samples from adult females compared to winter samples from adult females ( $p = 0.001$ ). The detected sequences were closely related to previously reported MiAstV, swine rotavirus C, and mink and swine HEV strains. Two astrovirus sequences were distantly related to all other detected sequences as well as previously reported MiAstVs. These results demonstrate low to moderate prevalence of the three viruses in feces from clinically healthy Canadian commercial mink, and suggest that further monitoring of these viruses may provide a better understanding of how these potentially zoonotic agents may play a role in mink enteritis and overall productivity.

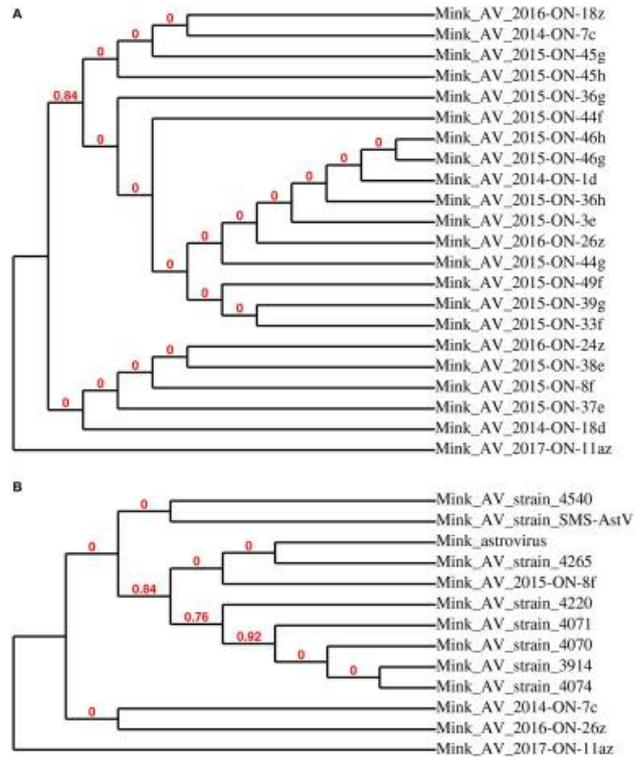


Fig. 1. Phylogenetic relationship of astrovirus RdRp partial sequences detected in mink fecal samples (A). Representative strains AV 2014-ON-7c, 2015-ON-8f, 2016-ON-26z, and 2017-ON-11az from this study were used for phylogenetic analysis with closely related astrovirus strains (B).

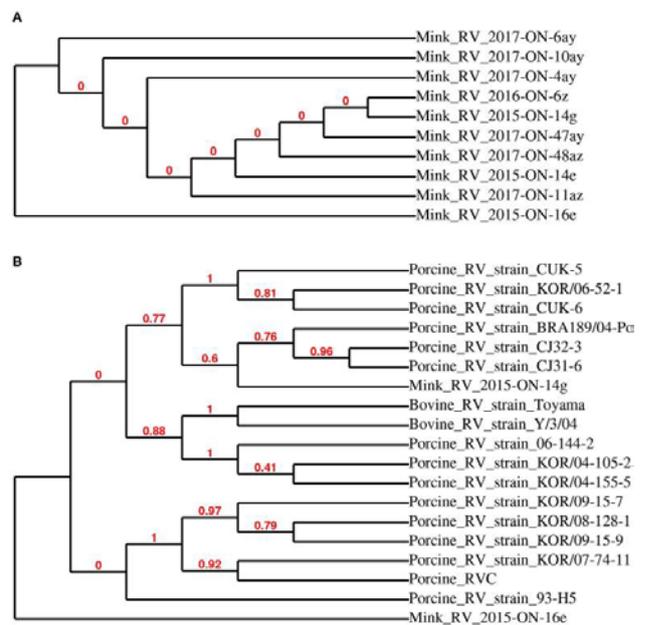


Fig. 2. Phylogenetic relationship of detected rotavirus VP6 partial sequences detected in mink fecal samples (A). Representative strains RV 2015-ON-14g and 2015-ON-16e from this study were used for phylogenetic analysis with closely related rotavirus strains (B).

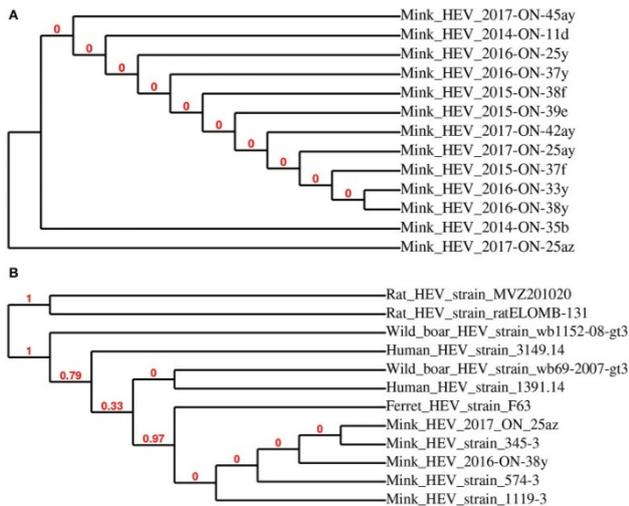


Fig. 3. Phylogenetic relationship of mink hepatitis E virus (HEV) RdRp partial sequences detected in mink fecal samples (A). Representative strains HEV 2016-ON-38y and 2017-ON-25az from this study were used for phylogenetic analysis with closely related HEV strains (B).

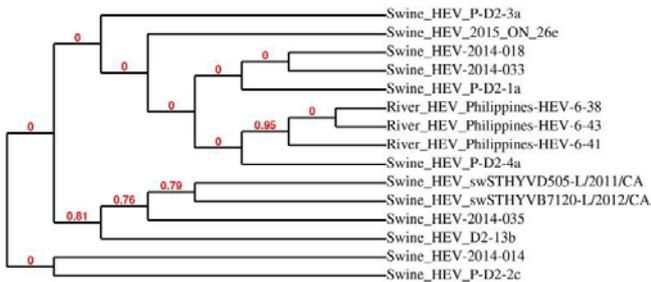


Fig. 4. Phylogenetic analysis of detected swine hepatitis E virus (HEV-2015-ON-26e) open reading frame 2 (ORF2) sequence and closely related viruses. HEV-2015-ON-26e was most closely related to swine HEV ORF2 sequences detected in fecal samples collected from Canadian swine herds.

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Doi: 10.3389/fvets.2018.00132. eCollection 2018.

### First detection and genetic analysis of fox-origin porcine circovirus type 2

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### Molecular and histopathological features of *Cryptosporidium ubiquitum* infection in imported chinchillas *Chinchilla lanigera* in Japan

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Long-tailed chinchillas *Chinchilla lanigera* are a popular rodent species kept both in households, where they are hand-raised as pets, and in zoological facilities. From January 2016 to February 2017, 13 juvenile chinchillas from five facilities in Japan were diagnosed with cryptosporidiosis at the animal hospital. Eight of the cases were fatal. All of the animals were imported from the Czech Republic by the same vendor. Histopathological and multilocus sequence analyses using 18S ribosomal RNA, actin, 70-kDa heat shock protein, and 60-kDa glycoprotein genes confirmed *Cryptosporidium ubiquitum* of subtype XIId as the etiological agent. Multilocus analysis demonstrated the presence of two new sequence types closely related to the *C. ubiquitum* XIId strain isolated from a human in the USA. This study indicated that potentially zoonotic *Cryptosporidium* is widespread and may have caused a high number of deaths among imported juvenile chinchillas.

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Doi: 10.1016/j.parint.2018.09.002.  
Epub ahead of print.

## Mink (*Neovison vison*) kits with pre-weaning diarrhea have elevated serum amyloid A levels and intestinal pathomorphological similarities with New Neonatal Porcine Diarrhea Syndrome

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### Background

Pre-weaning diarrhea (PWD) is a syndrome affecting farm-raised neonatal mink kits. Apart from diarrhea it causes greasy skin exudation, dehydration, and distressed behavior and can ultimately lead to death. No specific causative agents have been identified and the syndrome is regarded as multifactorial. The aim of the present study was to investigate a possible inflammatory state in mink kits with PWD, as indicated by raised serum concentrations of the acute phase protein serum amyloid A (SAA) and by changes in intestinal pathomorphology and intestinal contents of bacteria. Samples collected from 20 diarrheic mink kits with PWD and 20 age-matched non-diarrheic control mink kits from two commercial Danish farms during the pre-weaning period (April-May) in 2016 were analyzed.

### Results

Concentrations of SAA in serum samples from mink kits with PWD were significantly higher (up to 1000-

fold) compared to non-diarrheic control mink kits. Significant features of enterocytic vacuolization, atrophy and fusion of villi in jejunum and mucosal atrophy of the colon of kits with PWD were found. Moreover, attachment of coccoid bacteria to enterocytes was more often found in kits suffering from PWD, while intra-cytoplasmic eosinophil bodies were more frequently observed in control kits. Cellular infiltrations with mononuclear and neutrophil leukocytes were not associated with disease status. Bacteria from the *Staphylococcus intermedius* group, such as *Staphylococcus delphini*, were more frequently cultivated from control mink kits, whereas *Enterococcus* spp. dominated in mink kits with PWD. *Escherichia coli* was cultivated from both control and mink kits with PWD, but with a higher frequency from mink kits with PWD.

### Conclusion

A significant increase in circulating concentrations of SAA was found in PWD affected mink kits from 6 to 23 days old compared to controls. The histopathological changes in PWD mink kits suggest that the type of diarrhea is secretory. Attachment of coccoid bacteria, therefore, might be responsible for an enterotoxic effect causing a loss of balance in movements of ions and water leading to the vacuolization and swelling of the enterocytes. The slight to moderate infiltrations of neutrophils irrespectively of diarrheic status and the attachment of coccoid bacteria to enterocytes are comparable to observations found in piglets suffering from New Neonatal Porcine Diarrhea Syndrome. Mechanisms behind the correlation between increased SAA levels and the observed pathological intestinal features remain obscure.

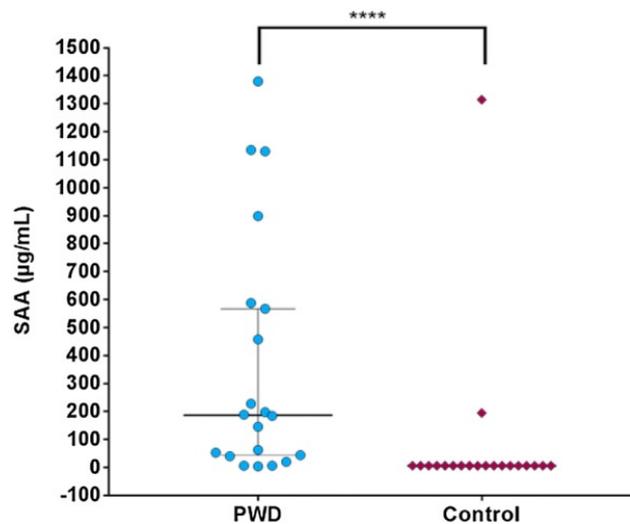


Fig. 1. Serum concentration of serum amyloid A (SAA) protein in PWD and healthy control mink kits as deter-

mined by SAA ELISA. Median of the serum SAA concentrations with 95% confidence interval is depicted (n = 20). The Mann-Whitney test showed a statistically significant higher concentration of serum SAA in PWD kits than in control kits (\*\*\*\*P < 0.0001)

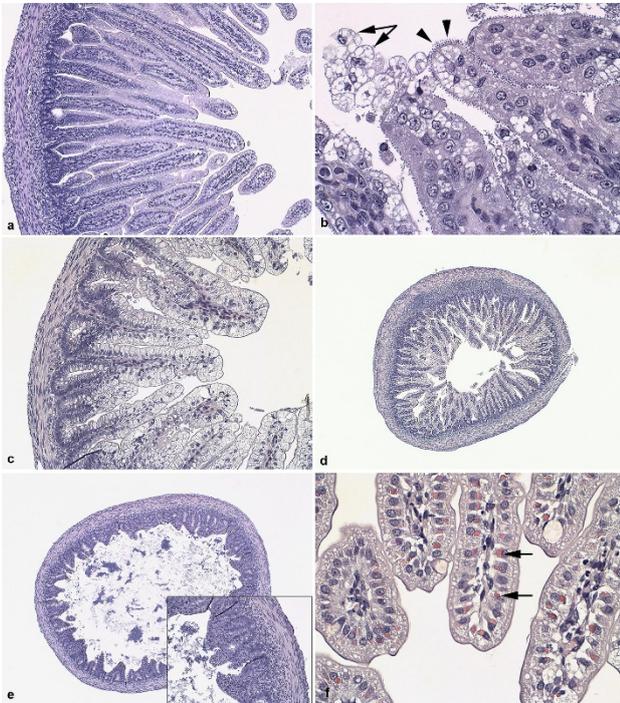


Fig. 2. Photomicrographs of representative examples of intestines from mink kits. **a** Jejunum from healthy mink kit. **b** Vacuolization on the tip of the villi (arrows) and pronounced attachment of coccoid bacteria (arrow heads) to the enterocytes in jejunum of a mink kit with PWD. **c** Severe vacuolization and hypertrophied enterocytes in the colon of a mink kits with PWD. **d** Colon from a healthy mink kit. **e** Atrophy of the mucosa of the colon from a mink kit with PWD. Inset: higher magnification of the mucosal atrophy. **f** Eosinophilic bodies in the enterocytes from jejunum in a healthy mink kit (arrows)

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### **Echinococcus multilocularis in Denmark 2012-2015: high local prevalence in red foxes**

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In Western Europe, the *Echinococcus multilocularis* lifecycle is predominantly sylvatic, typically involving red foxes (*Vulpes vulpes*) as the main definitive hosts with *Microtus* spp. and *Arvicola* spp. as intermediate hosts. During a 4-year surveillance study (2012-2015), Danish red foxes and raccoon dogs (n = 1345) were examined for *E. multilocularis*. Moreover, 134 insectivores and rodents collected in South Jutland during spring and summer 2016 were examined for the presence of metacestodes. The sedimentation and counting technique and molecular typing were used to identify *E. multilocularis* infections in the carnivores, while the rodent livers were examined macro- and microscopically for parasite lesions. Following morphological identification of *E. multilocularis* adult worms, the identity was verified by sequence analysis of the 12S rRNA gene in most cases (n = 13). *Echinococcus multilocularis* infection was demonstrated in 19 red foxes (*Vulpes vulpes*) originating from only two specific areas of South Jutland, namely Højer and Grindsted, and in two raccoon dogs (*Nyctereutes procyonoides*), originating from Højer. In Højer, 28.5% (CI 95% 11.7-45.3) of the examined red foxes were *E. multilocularis* positive per year. Moreover, positive red foxes were identified each year from 2012 to 2015, while *E. multilocularis* positive red foxes were only identified in Grindsted in 2013 (4.0%) and 2014 (6.4%). In contrast, all collected rodents were negative for *E. multilocularis*. We conclude that *E. multilocularis* is locally endemic in South Jutland with a high local prevalence in Højer.

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Doi: 10.1007/s00436-018-5947-y. Epub 2018 Jun 1.

**Morphological and molecular characterisation of *Eimeria* vison-like oocysts (*Apicomplexa:Eimeriidae*) in farmed mink (*Neovison vison*) in Denmark**

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A survey was conducted on 30 Danish mink farms from April to October 2016 to determine the prevalence and species of *Eimeria* in Danish farmed mink. In total, 2.6% of mink faecal samples (108/4140) were positive for *Eimeria* vison-like oocysts by microscopy, with 24.8% (78/315) of mink being positive at least once during the study period. Morphological analysis of sporulated oocysts (n = 20) identified *Eimeria* vison-like oocysts measuring 21.0 × 13.8 µm with a length/width (L/W) ratio of 1.5. Phylogenetic analysis of 18S rRNA sequences (1221 bp) from three positive mink indicated that *Eimeria* vison-like shared the highest genetic similarity to *Eimeria* sp. ex *Apodemus agrarius* from a Striped field mouse (*A. agrarius*) from the Czech Republic (99.6%). Analysis of a shorter region of 18S (531 bp) revealed that the *E. vison*-like genotype sequences grouped in the same clade and shared 97.7% similarity with *E. furonis*. At the cytochrome c oxidase subunit I (COI) locus, mink-derived sequences were not available from GenBank and phylogenetic analysis placed the novel *E. vison*-like in a clade with *E. cf. ictidea* (99.4% similarity) from a black footed ferret (*Mustela nigripes*) from Canada.

*Parasitol Res.* 2018 Sep; 117(9):2933-2939.

Doi: 10.1007/s00436-018-5989-1. Epub 2018 Jul 12.

**The 5' Untranslated Region of the Capsid Protein 2 Gene of Mink Enteritis Virus Is Essential for Its Expression**

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Mink enteritis virus (MEV), as a parvovirus, is among the smallest of the animal DNA viruses. The limited genome leads to multifunctional sequences and complex gene expression regulation. Here, we show that the expression of viral capsid protein 2 (VP2) of MEV requires its 5' untranslated regions (5' UTR) which promote VP2 gene expression at both transcriptional and translational levels. The expression of VP2 was inhibited in several common eukaryotic expression vectors. Our data showed that the 5' UTR of VP2 enhanced capsid gene transcription but not increased stability or promotes nucleocytoplasmic export of VP2 mRNA. Analysis of the functions of 5' UTR fragments showed that the proximal region (nucleotides [nt] 1 to 270; that is, positions +1 to +270 relative to the transcription initiation site, nt 2048 to 2317 of MEV-L) of 5' UTR of VP2 was necessary for VP2 transcription and also promoted the activity of P38 promoter. Unexpectedly, further analysis showed that deletion of the distal region (nt 271 to 653) of the 5' UTR of VP2 almost completely abolished VP2 translation in the presence of P38, whereas the transcription was still induced significantly. Furthermore, using a luciferase reporter bicistronic system, we identified that the 5' UTR had an internal ribosome entry site-like function which could be enhanced by NS1 via the site at nt 382 to 447. Mutation of the 5' UTR in the MEV full-length clones further showed that the 5' UTR was required for VP2 gene expression. Together, our data reveal an undiscovered function of 5' UTR of MEV VP2 in regulating viral gene expression. **IMPORTANCE** MEV, a parvovirus, causes acute enteritis in mink. In the present report, we describe an untranslated sequence-dependent mechanism by which MEV regulates capsid gene expression. Our results highlight the roles of untranslated sequences in regulating the transcriptional activity of P38 promoter and translation of capsid genes. These data also reveal the possibility of an unusual translation mechanism in capsid protein expression and the multiple functions of nonstructural protein. A better understanding of the gene expression

regulation mechanism of this virus will help in the design of new vaccines and targets for antiviral agents against MEV.

*J Virol.* 2018 Aug 29; 92(18). pii: e00787-18.  
Doi: 10.1128/JVI.00787-18. Print 2018 Sep 15.

### **Molecular epidemiology of parasitic protozoa and Ehrlichia canis in wildlife in Madrid (central Spain)**

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Wildlife species are involved in the transmission of diverse pathogens. This study aimed to monitor raccoons (*Procyon lotor*), American minks (*Neovison vison*), and red foxes (*Vulpes vulpes*) as potential reservoirs in central Spain. Specifically, 200 spleen and fecal samples (from 194 raccoons, 3 minks, and 3 foxes) were analyzed molecularly by PCR/qPCR and sequencing for the presence of piroplasmids, Hepatozoon spp., *Toxoplasma gondii*, and *Ehrlichia canis* infections in the Community of Madrid (Spain). Biological samples were obtained in the years 2014, 2015, and 2016. No pathogen DNA was found in fecal samples. In contrast, analysis of raccoon spleen samples revealed that *Toxoplasma* was the most prevalent pathogen (prevalence  $3.6 \pm 2.6\%$ ), followed by *Hepatozoon canis* and *E. canis* (each with a prevalence of  $2.57 \pm 2.2\%$ ). *Hepatozoon canis* was also diagnosed in all three of the analyzed foxes. Analysis of yearly prevalence showed that tick-borne pathogens were less frequent in raccoon in 2015, a dry and warm year compared both to 2014 and 2016. These data suggest that fecal PCR assays are unsuitable for detection of DNA of non-erythrocytic pathogens. Furthermore, they demonstrate that the raccoon (an invasive species often living in proximity to domestic areas) and the red fox are putative reservoirs for pathogenic organisms in the Community of Madrid.

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Doi: 10.1007/s00436-018-5919-2.  
Epub 2018 May 18.

### **A natural zoonotic giardiasis: Infection of a child via Giardia cysts in pet chinchilla droppings**

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Here, we report a case of direct zoonotic transmission of giardiasis between a pet chinchilla and a human. Microscopic and molecular examinations of stool samples from a child and samples of chinchilla droppings revealed cysts/DNA of *Giardia intestinalis*. The transmission from the chinchilla to the child has been confirmed as coprophagous after the 1-year-old toddler ingested pet chinchilla droppings. Molecular analysis of the *gdh* gene from both hosts classified the *G. intestinalis* cysts into the assemblage B genetic group, which has been previously shown to be characteristic of both human and chinchilla giardiasis. Both *Giardia* sub-assemblages BIII and BIV were present in the chinchilla droppings, whereas only the sub-assemblage BIV was isolated from the child's stool sample. To the best of our knowledge, this is the first report of a true zoonotic transmission of giardiasis, supporting the zoonotic potential of assemblage B.

*Parasitol Int.* 2018 Dec; 67(6):759-762.

Doi: 10.1016/j.parint.2018.07.010.

Epub 2018 Jul 24.

### Prevalence of intestinal helminths of red foxes (*Vulpes vulpes*) in central Europe (Poland): a significant zoonotic threat

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#### Background

The red fox (*Vulpes vulpes*) is widely distributed in the world; in central Europe, it is the most numerous wild species of the family Canidae. It can play the role of a definitive host for many intestinal parasites, including zoonotic helminths. Poland, with its geographical location (central Europe), is an interesting area for parasitological investigations of this species. The aim of this study was to evaluate and compare the prevalence of intestinal helminths in red foxes in different regions of Poland.

#### Methods

Intestines of 473 red foxes from four different regions were examined using the sedimentation and counting technique (SCT). In addition, 344 samples of faeces were examined using flotation.

#### Results

Overall, intestinal helminths were found in 98.9% of red foxes. The average prevalence of detected parasites was as follows: Mesocestoides spp. (84.1%); hookworms (67.9%); Alaria alata (61.5%); Toxocara/Toxascaris (49.5%); Taenia spp. (42.5%); Echinococcus multilocularis (25.6%); and Trichuris vulpis (2.3%). The prevalence of the majority of parasite species was similar in each region. Significant differences between regions were observed in the case of E. multilocularis: a low prevalence in the south-western and northern regions (0% and 0.9%, respectively) and a high prevalence in the south-east and northeast (39.3% and 42.7%, respectively). In the case of A. alata, important differences were found between northern (96.5% and 93.7% in northern and northeast regions, respectively) and southern regions (15.2% and 24.7% for south-western and south-east regions, respectively). The percentage of positive samples obtained with coproscopic examination (except for Trichuris) was significantly lower than that obtained with SCT. Analysis of the prevalence estimated in in-

dividual regions with the use of both methods (flotation and SCT) showed a high correlation for all parasite species (except for Mesocestoides spp.). The flotation method also allowed us to detect the eggs of the lung nematode Eucoleus aerophilus (syn. Capillaria aerophila) (76.2% of positive foxes).

#### Conclusions

This study showed a very high percentage of red foxes infected with intestinal helminths in different parts of Poland. Depending on the location, some differences were observed regarding the prevalence of dangerous zoonotic parasites, which should be considered in the assessment of infection risk for humans.

*Parasit Vectors*. 2018 Jul 28; 11(1):436.

Doi: 10.1186/s13071-018-3021-3.

### Seroprevalence of 12 serovars of pathogenic Leptospira in red foxes (*Vulpes vulpes*) in Poland

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#### Background

Leptospira spp. infect humans and a wide range of domestic and wild animals, but certain species such as small rodents and red foxes (*Vulpes vulpes*) play a particular role as reservoirs and transmission of leptospirosis as they easily adapt to many habitats including human environments. To investigate the significance of red foxes in the epidemiology of leptospirosis in Poland, a seroprevalence survey was conducted. During the 2014-2015 hunting season, blood samples of 2134 red foxes originating from the central-eastern part of Poland were collected. Serum

samples were tested by a microscopic agglutination test for the presence of specific antibodies to *Leptospira* serovars Icterohaemorrhagiae, Grippotyphosa, Sejroe, Tarassovi, Pomona, Canicola, Hardjo, Ballum, Australis, Bataviae, Saxkoebing and Poi.

## Results

Antibodies to at least one serovar were detected in 561 sera (26.3%). The highest seroprevalence was found in the Subcarpathia (41.6%) and Warmia-Masuria (40.3%) provinces. Antibodies were mainly directed against serovars Poi (12.4%), Saxkoebing (11.3%), and Sejroe (6.0%).

## Conclusions

Exposure of red foxes to certain *Leptospira* serovars seems to be common in central and eastern Poland. In addition, the high prevalence of antibodies against *Leptospira* spp. in foxes may indicate a potential risk of infection for humans and other species coming into contact with these animals.



Fig. 1. Geographic distribution of red foxes seropositive for pathogenic *Leptospira* in Poland. LD Łódzkie, MP Lesser Poland, MA Masovia, OP Opolskie, PK Subcarpathia, PM Pomerania, SL Silesia, SW Świętokrzyskie, WM Warmia-Masuria, DS Lower Silesia, KP Kuyavian-Pomerania, LB Lubuskie, LU Lubelskie, PD Podlaskie, WP Greater Poland, ZP West Pomerania.

*Acta Vet Scand.* 2018 May 31; 60(1):34.  
Doi: 10.1186/s13028-018-0388-2.

## Associations between pneumonia and residential distance to livestock farms over a five-year period in a large population-based study

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In a recent study of electronic health records (EHR) of general practitioners in a livestock-dense area in The Netherlands in 2009, associations were found between residential distance to poultry farms and the occurrence of community-acquired pneumonia (CAP). In addition, in a recent cross-sectional study in 2494 adults in 2014/2015 an association between CAP and proximity to goat farms was observed. Here, we extended the 2009 EHR analyses across a wider period of time (2009-2013), a wider set of health effects, and a wider set of farm types as potential risk sources. A spatial (transmission) kernel model was used to investigate associations between proximity to farms and CAP diagnosis for the period from 2009 to 2013, obtained from EHR of in total 140,059 GP patients. Also, associations between proximity to farms and upper respiratory infections, inflammatory bowel disease, and (as a control disease) lower back pain were analysed. Farm types included as potential risk sources in these analyses were cattle, (dairy) goats, mink, poultry, sheep, and swine. The previously found association between CAP occurrence and proximity to poultry farms was confirmed across the full 5-year study period. In addition, we found an association between increased risk for pneumonia and proximity to (dairy) goat farms, again consistently across all years from 2009 to 2013. No consistent associations were found for any of the other farm types (cattle, mink, sheep and swine), nor for the other health effects considered. On average, the proximity to poultry farms corresponds to approximately 119 extra patients with CAP each year per 100,000 people in the research area, which accounts for approximately 7.2% extra cases. The population attributable risk percentage of CAP cases in the research area attributable to proximity to goat farms is approximately 5.4% over the years 2009-2013. The most probable explanation for the association of CAP with proximity to poultry farms is

thought to be that particulate matter and its components are making people more susceptible to respiratory infections. The causes of the association with proximity to goat farms is still unclear. Although the 2007-2010 Q-fever epidemic in the area probably contributed Q-fever related pneumonia cases to the observed additional cases in 2009 and 2010, it cannot explain the association found in later years 2011-2013.

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eCollection 2018.

### **In Vivo Safety Studies With SPBN GASGAS in the Frame of Oral Vaccination of Foxes and Raccoon Dogs Against Rabies**

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In order to obtain Marketing Authorization for an oral rabies vaccine in the European Union, not only safety studies in the target species, red fox and raccoon dog, are required. Since baits are distributed unsupervised in the environment, specific safety studies in selected non-target species are compulsory. Furthermore, oral rabies vaccines are based on live, replication-competent viruses and thus distinct safety studies in the target species for such type of vaccines are also mandatory. Here, the results of these safety studies in target and selected non-target species for a 3rd generation oral rabies virus vaccine construct, SPBN GASGAS (Rabitec), are presented. The studies included the following species; red fox, raccoon dog, domestic dog, domestic cat, domestic pig, wild rodents. The following safety topics were investigated; overdose, repeated dose, dissemination, shedding, horizontal and vertical transmission. It was shown that SPBN GASGAS did not cause disease or any other adverse reaction in vaccinated animals and naïve contact animals. The vaccine did not disseminate within the host beyond the site of entry. No horizontal transmission was observed in wild rodents. In the target species, there was evidence that in a few cases horizontal transmission of vaccine virus could have occurred under these experimental conditions; most likely immediately after vaccine administration. The vaccine

construct SPBN GASGAS meets therefore the latest revised minimal safety requirements as laid down in the European Pharmacopoeia.

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### **Systemic Staphylococcus pseudintermedius infection in an arctic fox (*Vulpes lagopus*) with severe multifocal suppurative meningoencephalitis and nephritis**

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A 2-year-old female arctic fox (*Vulpes lagopus*) developed anorexia, dehydration, and emaciation during the quarantine period for importation from Norway, and died 17 days later. At necropsy, a fistula was observed on the left gluteal region, and the left eye, left brain, and kidneys were discolored. Histologically, severe diffuse suppurative meningoencephalitis and renal abscesses were detected. Numerous Gram-positive cocci were detected in these lesions. Multidrug-susceptible *Staphylococcus pseudintermedius* were isolated from the lesions. These results suggest that *S. pseudintermedius* can cause severe multifocal suppurative meningoencephalitis and nephritis in foxes. This is the first report of multidrug-susceptible *S. pseudintermedius* meningoencephalitis and nephritis in a fox.

*J Vet Med Sci*. 2018 Aug 10; 80(8): 1219-1222.

Doi: 10.1292/jvms.18-0061. Epub 2018 Jun 11.

### Severe parasitism by *Versteria mustelae* (Gmelin, 1790) in the critically endangered European mink *Mustela lutreola* (Linnaeus, 1761) in Spain

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The riparian European mink (*Mustela lutreola*), currently surviving in only three unconnected sites in Europe, is now listed as a critically endangered species in the IUCN Red List of Threatened Species. Habitat loss and degradation, anthropogenic mortality, interaction with the feral American mink (*Neovison vison*), and infectious diseases are among the main causes of its decline. In the Spanish Foral Community of Navarra, where the highest density of *M. lutreola* in its western population has been detected, different studies and conservation measures are ongoing, including health studies on European mink, and invasive American mink control. We report here a case of severe parasitism with progressive physiological exhaustion in an aged free-ranging European mink female, which was accidentally captured and subsequently died in a live-trap targeting American mink. Checking of the small intestine revealed the presence of 17 entangled *Versteria mustelae* worms. To our knowledge, this is the first description of hyperinfestation by tapeworms in this species.

*Parasitol Res.* 2018 Oct; 117(10):3347-3350.

Doi: 10.1007/s00436-018-6043-z. Epub 2018 Sep 4.

### What does the fox say? Monitoring antimicrobial resistance in the environment using wild red foxes as an indicator

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The objective of this study was to estimate and compare the occurrence of AMR in wild red foxes in relation to human population densities. Samples from wild red foxes (n = 528) included in the Norwegian monitoring programme on antimicrobial resistance in bacteria from food, feed and animals were included. All samples were divided into three different groups based on population density in the municipality where the foxes were hunted. Of the 528 samples included, 108 (20.5%), 328 (62.1%) and 92 (17.4%) originated from areas with low, medium and high population density, respectively. A single faecal swab was collected from each fox. All samples were plated out on a selective medium for Enterobacteriaceae for culturing followed by inclusion and susceptibility testing of one randomly selected *Escherichia coli* to assess the overall occurrence of AMR in the Gram-negative bacterial population. Furthermore, the samples were subjected to selective screening for detection of *E. coli* displaying resistance towards extended-spectrum cephalosporins and fluoroquinolones. In addition, a subset of samples (n = 387) were subjected to selective culturing to detect *E. coli* resistant to carbapenems and colistin, and enterococci resistant to vancomycin. Of these, 98 (25.3%), 200 (51.7%) and 89 (23.0%) originated from areas with low, medium and high population density, respectively. Overall, the occurrence of AMR in indicator *E. coli* from wild red foxes originating from areas with different human population densities in Norway was low to moderate (8.8%). The total occurrence of AMR was significantly higher;  $\chi^2$  (1, N = 336) = 6.53, p = 0.01 in areas with high population density compared to areas with medium population density. Similarly, the occurrence of fluoroquinolone resistant *E. coli* isolated using selective detection methods was low in areas with low population density and more common in areas with medium or high population density. In conclusion, we found indications that occurrence of AMR in wild red foxes in

Norway is associated with human population density. Foxes living in urban areas are more likely to be exposed to AMR bacteria and resistance drivers from food waste, garbage, sewage, waste water and consumption of contaminated prey compared to foxes living in remote areas. The homerange of red fox has been shown to be limited thereby the red fox constitutes a good sentinel for monitoring antimicrobial resistance in the environment. Continuous monitoring on the occurrence of AMR in different wild species, ecological niches and geographical areas can facilitate an increased understanding of the environmental burden of AMR in the environment. Such information is needed to further assess the impact for humans, and enables implementation of possible control measures for AMR in humans, animals and the environment in a true "One Health" approach.

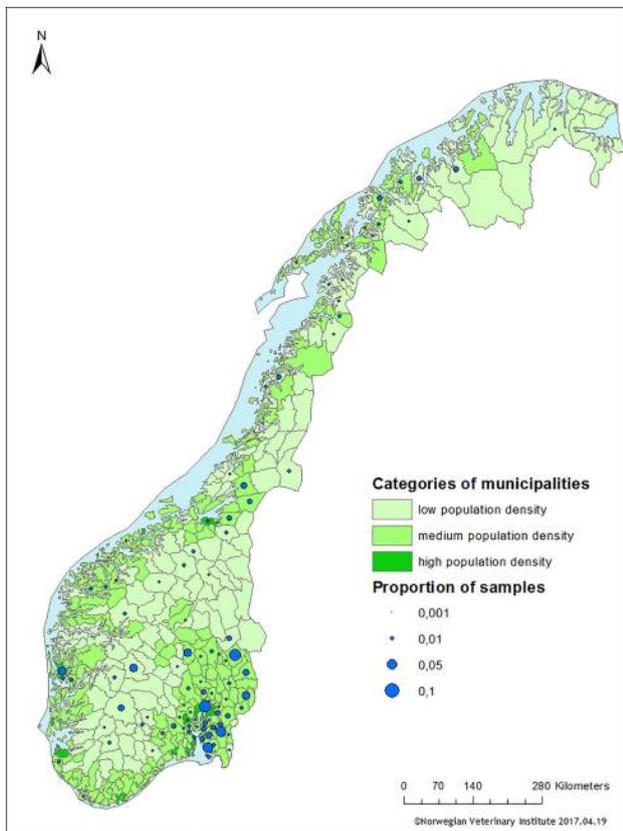


Fig. 1. Distribution of sampled wild red foxes per municipality. Each municipality was categorised according to the human population density (in green) and the proportion of samples within each municipality is displayed as blue circles.

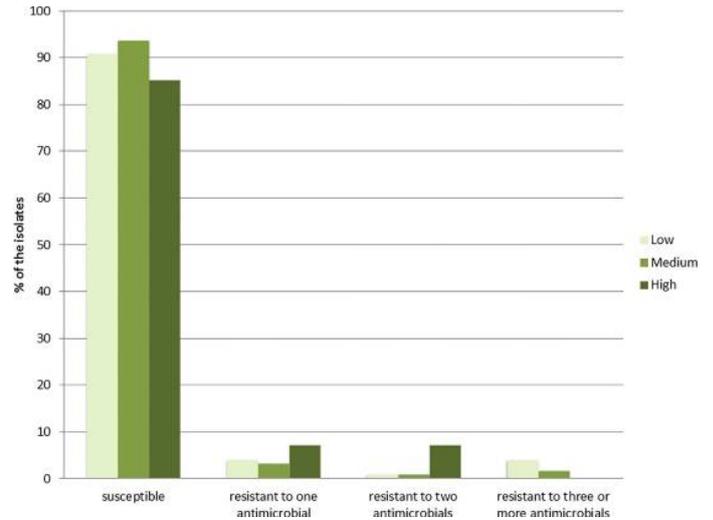


Fig. 2. Occurrence of antimicrobial resistance among *Escherichia coli* (N = 434) isolated from wild red foxes in Norway in 2016. The isolates are categorized according to human population density in the area where the foxes were hunted, i.e. in low population density (n = 98), medium population density (n = 268) and high population density (n = 68).

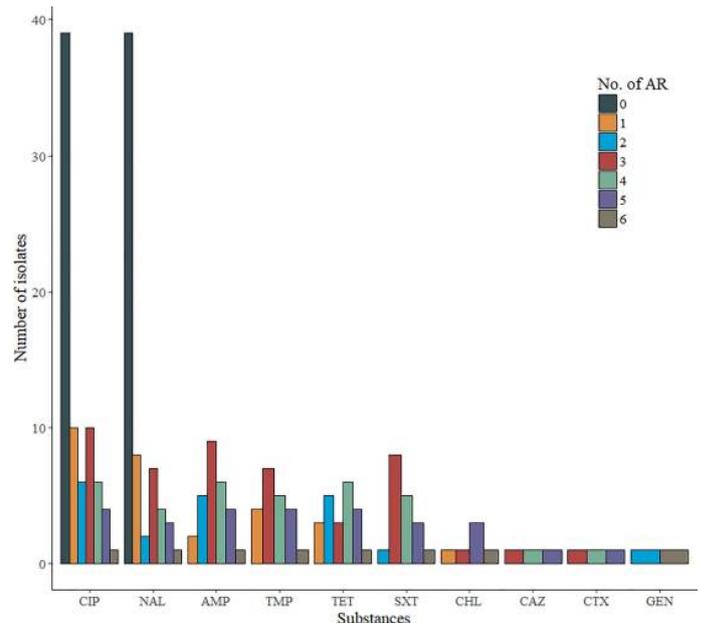


Fig. 3. Resistance profiles of the fluoroquinolone resistant *Escherichia coli* isolates (n = 76) isolated by selective screening from wild red foxes in 2016 in Norway. No. of AR = Number of additional resistances to other antimicrobial classes than quinolones (including nalidixic and/ or ciprofloxacin), CIP = ciprofloxacin, NAL = nalidixic acid, AMP = ampicillin, TET = tetracycline, TMP = trimethoprim, SXT = sulfamethoxazole, CHL = chloramphenicol, CTX = cefotaxime, CAZ = ceftazidime, GEN = gentamicin.

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## **Fleas and Ticks of Red Foxes as Vectors of Canine Bacterial and Parasitic Pathogens, in Slovakia, Central Europe**

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The present survey aimed to investigate flea and tick fauna parasitizing Slovak red fox populations with special emphasis on canine pathogens they transmit. A total of 407 fleas and 105 ticks were collected from 90 red foxes from two geographically distant regions. Seven flea species (*Chaetopsylla globiceps*, *Pulex irritans*, *Archaeopsylla erinacei*, *Chaetopsylla rothschildi*, *Chaetopsylla trichosa*, *Ctenocephalides canis*, and *Ctenophthalmus assimilis*) and three species of hard ticks (*Ixodes ricinus*, *Ixodes hexagonus*, *Haemaphysalis concinna*) were recorded on sampled animals. Consequently, the DNA of five different pathogen taxa was confirmed in collected arthropod vectors: *Bartonella* spp. (in *P. irritans*, *Ch. globiceps*, and *Ct. assimilis*), *Rickettsia* spp. (in *A. erinacei*, *I. ricinus*, *I. hexagonus*, and *H. concinna*), *Anaplasma phagocytophilum* (in *I. ricinus*), *Theileria* sp. (in *Ch. globiceps* and *H. concinna*), and *Hepatozoon canis* (in *I. ricinus* and *I. hexagonus*). *Mycoplasma* spp., *Dipylidium caninum*, and *Acanthocheilonema reconditum* were not found in fleas or ticks in this study.

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**Actual Mink Research 2018**  
**Meeting at Research Centre Foulum**  
**Faculty of Science and Technology**  
**Aarhus University, Denmark**  
**20 September 2018**





### **Iron supplementation to mink kits at day three after birth**

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Iron (Fe) plays an important role in growth and hemoglobin formation. After birth and until mink kits begin to ingest solid feed, the hemoglobin level diminishes indicating that mink kits in this period are likely to suffer from iron deficiency (anemia). Therefore, the purpose of this experiment was to investigate the effect of injecting mink kits with iron day 3 after birth on hemoglobin and hematocrit levels and content of iron in the liver as well as growth. The experiment showed that injection with 2 mg iron day 3 after birth resulted in a significant increase in hemoglobin and hematocrit day 18 but not day 39 and 58 after birth. Injection with iron also caused an increase in the iron content of the liver day 18 but had no effect on growth day 18, 39 and 58. The injected iron dose corresponded to the dose per gram of body weight used as a standard for suckling pigs day 3 after birth to prevent anemia. However, mink kits have a relatively higher growth rate compared with suckling pigs. Consequently, the used iron dose per gram of growth was considerably less for the mink kits compared with suckling pigs. This probably explains the lack of effect of iron on growth and hemoglobin and hematocrit day 39 and 58. However, further experiments with higher iron doses are required to confirm this. If future experiments with a higher iron dose show similar positive effects on growth and survival of mink kits, as the right iron dose has been shown to have in suckling pigs, it will contribute to increased productivity and thereby improve the competitiveness of the mink breeders.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 6-13. Author's abstract.*

### **The excretion of vitamin B in the urine of female mink during the period January to May**

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The vitamin B complex plays an important role in a wide range of metabolic pathways and the need for adding the vitamin B complex to the feed may change during the year. The aim of the present project was to evaluate the excretion of B vitamins in the urine of female mink during the period January to May where they experience very different feeding regimens and where pregnancy also may influence the need for vitamin B. Four groups of female mink were included in the experiment, they were fed diets containing either 0, 33, 66 or 100 % of the recommended amount of the vitamin B complex. Urine samples were collected from the female mink on January 3, February 28, March 6, March 30, and April 19. Furthermore, urine samples were collected from their kits on June 2. The samples were analyzed using non-targeted metabolomics. The results showed that the amount of riboflavin, niacin, pantothenic acid and pyridoxine excreted in the urine increased linearly with increasing amount of vitamin B in the feed. Hence, the amount of vitamin B added to the feed can be lowered during January to May. However, the urine samples collected from the kits showed that their excretion of vitamin B generally was lower than the excretion observed from the females in April and the excretion was very low in the groups fed diets containing less than the recommended amount of vitamin B. In conclusion, the results suggests that the amount of vitamin B added to the diets of female mink may be lowered during the period January to May whereas lactating mink and mink kits should be fed the recommended amount of vitamin B.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 14-19. Author's abstract.*

### **Realistic selection for feed efficient mink**

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Feed costs are the largest day-to-day costs of the mink industry. Therefore, selection for feed efficient mink is beneficial in mink production, as it can reduce the amount of feed required. Mink is commonly

houses in pairs (typically a male and a female per cage) during the growing-furring period and feed intake is therefore measured as the sum of two animals feed intake making it a group record. The aims with the project were to develop a simple model for group recorded feed intake, which allows for records from cages of mixed sex (most common) and cages of same sex and still give variance estimates for each sex separately. This was done by using a univariate animal BLUP-model:

$$FI = \text{YEAR} + \text{GSEX} + \text{ID1} + \text{ID2} + \text{LIT1} + \text{LIT2} \\ + \text{GYL} + \beta \times \text{GSBW} + e$$

where FI was feed intake, YEAR and GSEX was the fixed effects of birth year and sex combination in the group (male-male, male-female, female-female), ID1 and ID2 was the random animal effects of the two animals in the group, LIT 1 and LIT 2 were the random effects of the litter of birth of the two animals, GYL was the random effect of the combination of selection lines in the group,  $\beta$  was the regression coefficient of a fixed regression on the sum of start body weight (GSBW) of the two animals and  $e$  was the random residual. The random animal effects were estimated using the pedigree. Results showed approx. twice as large variance parameters in males than females. The genetic correlation between males and females were high, but differed from unity. The residual variance were not estimated sex specific and the heritability could therefore not be calculated. The model needs to be developed further such that the residual variance can be estimated sex specific. Furthermore the model needs to be implemented in a multi-trait setting with end body weight and litter size.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 25-29. Author's abstract.*

## Use of auction data in the genetic evaluation of mink

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Our aim was to investigate the possibilities of using auction data in the genetic evaluation of mink.

We used data from individually pelted mink raised at Foulum research farm, Aarhus University, Denmark, to investigate heritability for mink pelt size and quality based on the auction label for mink graded by Copenhagen Fur. We also estimated heritability for auction price. We compared our heritabilities to similar heritabilities for size and quality assessed at the live pelt grading in November, where size was measured as weight in gram. We also compared to pelt size and quality evaluated on dried pelts prepared for auction. We found a heritability for all traits, but the auction labels with relative few and rough categories resulted in lower heritabilities than the ones estimated from the more clear defined categories for traits assessed from the live pelt grading and the dried pelts. For size, the heritability was 0.46 from the live grading (weight), 0.44 based on dried pelts (cm) and 0.39 from auction size label (6-cm classes). For live quality, the heritability was 0.31, for dried pelts the heritability was 0.30. For auction quality label the heritability was only 0.11. Heritability for the "pseudo-trait" prize was 0.16. As we expected, we found a high positive genetic correlation between pelt size and price; live weight vs price: 0.78; dried pelt size vs price: 0.88; auction size label vs price: 0.88. After correction for pelt size, the genetic correlation between quality and price was also positive; live grading quality vs price: 0.56; dried pelt quality vs price: 0.40; auction quality label vs price: 0.28. If auction data from previous generations should be included in the genetic evaluation of present generation of mink, it will be preferred to use the measures for quality and size that form the basis for the auction label, instead of using the final labels.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 30-36. Author's abstract.*

## MRSA in mink

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During 2015-2017, a number of projects regarding MRSA in mink were performed at the National Veterinary Institute, Technical University of Denmark. Results from this study support the hypothesis of mink feed as a likely route of MRSA transmission to mink. MRSA may persist for more than 26 days after exposure to MRSA in-feed. Regarding human exposure, airborne transmission seems unlikely. However, due to the anatomical location of MRSA on mink (paws and pharynx), mink farmers should be aware of the risk of MRSA infection through bites and scratches while handling of mink.

The four studies included:

1. Screening of mink send for diagnostic investigations during 2015. A total of 34% (20/58) farms were found positive of MRSA.
2. A field study on five Danish mink farms during May-August 2017 to identify the animal prevalence and environmental reservoir of MRSA on-farm. MRSA-positive mink were found in 4/5 farms, and the prevalence ranged from 20-34% in kits and 0-25% in breeding mink. The bacterium was widely distributed in the environment, as it was found in feed, gloves, cages and nest boxes. In contrast to pig farms, MRSA could not be isolated from air.
3. A microbiological study of mixed mink feed and raw ingredients in three feed producers during 2016-2017. MRSA was found in raw ingredients of pork and poultry origin from two of the three feed producers.
4. Feed spike on 28 naïve brown mink in closed test-facilities in 2017. MRSA could be isolated from all mink 24 hours after MRSA was added to the feed. Mink were followed 26 days after the feed spike was ceased. At this point in time, MRSA could still be re-isolated from 11/28 mink.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 37-40. Author's abstract.*

### **Mink have become less fearful than previous generations and coat colour influences temperament**

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*<sup>1</sup>Department of Animal Science, Aarhus University, Denmark.*

The aim of the study was to investigate which factors had an effect on the temperament test performed during WelFur certifications. There were two hypotheses: that mink with Silver coat colour would be more explorative than mink with Brown coat colour (i.e. the effect of colour) and that there would be an overall reduction in fear since the chosen farms last were tested (in 1988, i.e. the effect of time). Data was collected using the temperament test performed on three colour variants: Brown, Silver and Pearl. Depending on the mink's reaction to the temperaments test, it would be classified as either "Aggressive", "Fearful", "Curious" or "Indecisive". Alongside my own data, data from a previous temperament test, performed in 1988, was utilized to investigate the effect of time on temperament. From my own results, odds ratios (the ratios between two significantly different odds) for the effect of coat colour were calculated.

Mink with Silver or Pearl coat colours turned out to be more fearful and less curious than mink with Brown coat colour. The effects of colour seen in this study were attributed to the pleiotropic effect of coat colour on either endocrine production or sensory organs. Although, the odds ratios for these effects were relatively low, so it was concluded that other factors (e.g. management) would affect temperament more than colour. Also, the average frequency of fearful temperament across all farms fell from 30.6% to 20.8% over the 30-year period. This decrease in fear over time was concluded to be due to the indirect selection performed by farmers during the yearly production, the high heritability of the behaviours studied and the consistent environment the mink were kept in.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 41-45. Author's abstract.*

### **Early transfer favour dam acceptance of a foster kit and induce higher kit growth in mink**

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Only few systematic studies focus on cross fostering of mink offspring and levelling of litters early in life. Methods: Brown mink was used, and 408 transponder marked kits were transferred from large litters (9-14 kits) to foster mothers (N=408, 4-7 kit per litter) on either Day 2 (N = 201) or Day 6 (N=207) relative to the day of birth (Day 0). Besides timing of transfer, the foster mother age class (young: 1.st parity, old: 2<sup>nd</sup>-3<sup>rd</sup> parity) was a factor in the study. The transferred kits were 50% male and 50% female. Results: Mink mothers retrieved the novel kit quicker to the nest at Day 2 than at Day 6. The offspring body-weight (Day 56) was higher after transfer at Day 2 than at Day 6. Young and old dams displayed an equal acute acceptance of the foster kit, but the growth of kits was significantly better in litters nursed by the experienced, older dams. Foster kits had a higher risk of dying (15.8 % vs. 11.4 % for the biological kits in the receiving litter) until Day 56. The mortality of foster kits was not different from that in a group of intact control litters with 8-14 kits. There was a strong influence of offspring sex on the amount of damages observed around Day 49. Out of the 2516 examined kits in receiver litters, 6.8% of the males and 46.1% of the females, had some sort of a damage (primarily crusts/wounds on the side of the neck), scored in different intensities from 1-5. Conclusion: It is recommended to transfer mink kits early (within the first days) after birth and to choose old rather than young dams as receiver of foster kits.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 59-68. Author's abstract.*

**Positive research effects of extra water for kits and ad-libitum feeding of mink dams after delivery are also seen in practice, but research results differ from practice**

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Research has shown a better development of mammary gland tissue in mink dams if they are fed ad libitum after birth, rather than more restricted. It has also been shown that additional water for mink kits close to the next box helps the kits to drink at an earlier age, grow faster, have less wounds and therefore fewer have to be moved from the litter. In order to test the effects under varied management conditions we

tested an additional water nipple and ad libitum feeding on four private farms, compared to no additional water nipple and a feeding strategy where food allowance was not increased until some time after birth. As on research farms we found a positive effect of extra water for kits and ad-libitum feeding for the dams in terms of less kit mortality while the number of injuries increased. The effects of both extra water for kits and ad-libitum feeding for the dams differed with dame age.

*Meeting at Research Centre Foulum, Faculty of Science and Technology, Aarhus University, Denmark. DCA report no. 129, September 2018 (in Danish) p. 69-75. Author's abstract.*

# **TEMPERAMENT IN MINK**

## **The effect of coat colour, housing system and time**



**MSc thesis by Louise Maiken Beagan Ring**

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Aarhus University**

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The aim of this study was to investigate three hypotheses: that silver mink would be more explorative than browns (effect of colour), that there would be more aggression and fear in group-housed mink (effect of housing), and that there would be an overall reduction in fear since the chosen farms last were tested (1988/1996 – i.e., effect of time). Seven farms were selected based on previous participation in temperament tests (either in 1988 or 1996), coat colours produced and types of housing system. Data was collected using the stick test, and approximately 100 mink were tested per colour type and housing system in weeks 39-42 in 2017. Also, data from a similar study in 2012 was included. This study had been performed at the research facility Foulum and had focused on the effect of housing system on brown mink.

All three hypotheses were to a certain degree supported by my results: brown mink had higher odds of being explorative than silver and pearl mink, there were higher odds of aggression in group housing (although in an interaction with colour), and fearful behaviour fell from 48% to 23% between the 1988/96 study and this study. Results on colour were attributed to the pleiotropic effect of coat colour on endocrine production or sensory organs. However, the odds ratios found in this study were relatively low, which indicated that other factors such as management would likely affect temperament more than coat colour. The increased aggression seen in group housing was possibly due to groups consisting of mainly females (territorial aggression). Finally, the decrease in fear seen in this study was likely due to the indirect selection performed by farmers, the high heritability of the behaviours studied, and the consistent environment the mink were kept in.

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