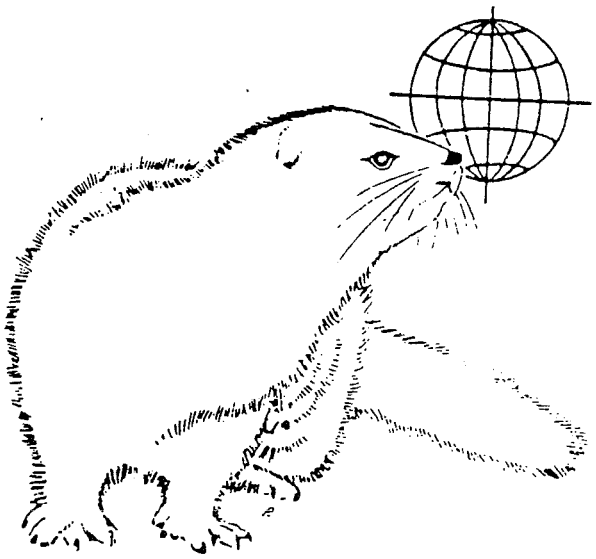

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Titles of other publications - not abstracted

Chinchillas (Review). *H.L. Hoefer. Veterinary Clinics of North America, Small Animal Practice, Vol. 24, No. 1, pp. 103-111, 1994. Code 1-14-O.*

Chinchilla; a marvel of animal naturel. *P.L. Caballero. Anales de la Sociedad Rural Argentina, Vol. 124, 1/3, pp. 22-28, 1991. In SPAN. Code 1-14-O.*

4. Genetics

5. Reproduction

6. Nutrition

Titles of other publications - not abstracted

Iniencephaly and other neural tube defects in a litter of ferrets (*Mustela putorius furo*). *B.H. Williams, E.J. Popek, R.A. Hart, R.K. Harris. Vet. Pathol. 31: 260-262, 1994. Code 9-O.*

Evaluation of fox-chasing encosures as sites of potential introduction and establishment of *Echinococcus multilocularis*. *Gregory W. Lee, Kimberly A. Lee, William R. Davidson. Journal of Wildlife Diseases, Vol. 29, No. 3, pp. 498-501, 1993. Code 9-F.*

Distemper vaccination in ferrets. *W.S.K. Chalmers, M.R. Geary. Veterinary Record, Vol. 130, No. 6, pp. 127, 1992. Code 9-O.*

Megaesophagus in a domestic ferret. *C.A. Harms, G.A. Andrews. Laboratory Animal Science, Vol. 43, No. 5, pp. 506-508, 1993. Code 9-O.*

Mycobacterium bovis infections in wild ferrets. *G.W. de Lisle, K. Crews, J. de Zwart, R. Jackson, G.J.E. Knowles, K.D. Paterson, R.W. MacKenzie, K.A. Waldrup, R. Walker. New Zealand Veterinary Journal, Vol. 41, No. 3, pp. 148-149, 1993. Code 9-O.*

Aleutian disease in laboratory ferrets. *S.E. Wolfensohn, M.H. Lloyd. Veterinary Record Vol. 134, No. 4, pp, 100, 1995. Code 9-O.*

Viral enteritis in mink. *N.S. Bukina. Krolikovodstvo i Zverovodstvo, No. 1, pp. 23, 1993. Code 9-M.*

Notes
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At the end of July 1995, one of the world's few, and certainly the largest, Fur Animal Research Departments run with governmental funds no longer exists. On 31 July we must say goodbye to the Department for Small Farm Animal at the Danish Institute of Animal Science, Denmark, which will together with the other animal species specific departments be restructured into discipline oriented departments.

For a person who has since 1958 been employed at - and from 1965 and until July of 1993 was responsible for - the Fur Animal Department at the Institute it is hard to believe that this is a good decision, although it is done in the name of efficiency and internationalization.

Well knowing that very much and very advanced research - also on fur animals - is done by discipline departments at a large number of universities and research institutes worldwide, I really feel that the Danish fur breeders, and also the whole international fur animal production family, have lost not only a partner but a department covering important research disciplines, in which everybody from top to bottom identified themselves and their scientific problems with the problems of the fur animal producers.

Personally I want to thank all my colleagues at the former Fur Animal Department for your enthusiasm and loyalty to your research and to the fur breeders whom you were always aware of serving. I wish you all the best in the future and hope that you will go on feeling some loyalty to fur animals. Also my thanks to the Danish Fur Breeders Association that from the start in 1947 sponsored a new fur animal research farm, and in 1963 they again supported a research farm, and through its annual donations to the department made it possible to build up the world's largest research unit on fur animals.

Sorry for all this "private" talk in the leading international fur animal scientific journal. All these facts have, however, given rise to a variety of reflections over the last months, and why not use SCIENTIFUR for that as well.

As the editor of SCIENTIFUR I see no reason for negative thinking. We have more material than ever, both as original reports and as abstracts. I would therefore like to take this opportunity to thank all our contributors and the growing, although slowly, stock of subscribers for the confidence you show. It is the vitamin we need to maintain our optimism.

It has now been confirmed by the President of IFASA, prof. Einar J. Einarsson, that we maintain the subscription prices of SCIENTIFUR at the same level in 1996 as in the years before, i.e. NOK 600 per volume for ordinary subscribers and NOK 500 for personal members of IFASA. This is possible thanks to the substantial support of the Council of European Fur Breeders' Association (CEFBA) to the production of SCIENTIFUR.

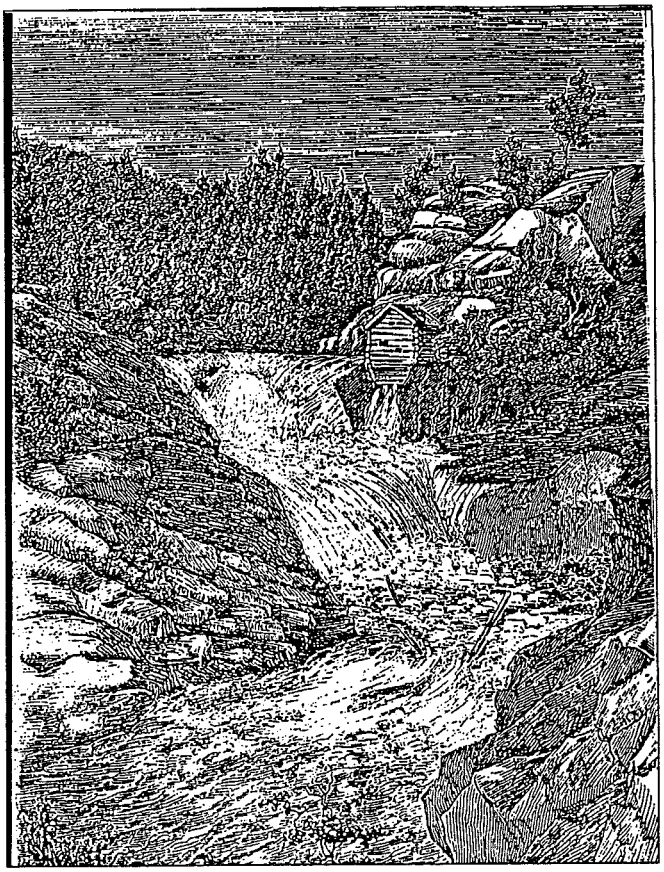
The SCIENTIFUR ELECTRONIC INDEX is becoming still more popular and for those who have already purchased the index, it is hard to imagine how they could possibly manage without it. An updated SCIENTIFUR ELECTRONIC INDEX covering Vol. 1-19 will be available around February 1996. Also for the index, the price will be the

same as before, i.e. Updating of existing indexes NOK 200.-, New index NOK 350.- for IFASA members and NOK 500.- for others. All prices are excl. postage.

Finally, we would like to ask you to pay attention to the other services frequently advertized in SCIENTIFUR such as books and previous volumes of SCIENTIFUR, which are necessary to derive full benefit from the electronic index.

We wish you a pleasant summer.

Your editor
and his enthuastic assistants



Original Report

The effects of cross-mating on the development of behaviour during the primary socialization period in silver foxes (*Vulpes vulpes*)

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Summary

Previous studies have demonstrated that the duration of the primary socialization period in foxes selected for domestic behaviour is longer than in unselected foxes showing enhanced aggressiveness towards humans (*Belyaev et al., 1985*). The present results with reciprocal matings between domestic and aggressive foxes show evidence for maternal effects on the formation of behavioral patterns relevant to the limitation of the socialization period and behaviour towards humans in adult hybrids. Thus, the primary socialization period was of the same duration in domestic hybrids from two mating types (either the mother was domestic and the father was aggressive, or *vice versa*) as in offspring from both domestic parents. This means that the period was not restricted to 60 days. However, in domestic pups from an aggressive mother and a domestic father, locomotor activity was significantly decreased like in the aggressive pups from aggressive parents. In the aggressive pups from aggressive mothers and domestic fathers, the sensitive socialization period was the same in pups from aggressive parents, i.e. it was restricted to 40 days. However, aggressive pups from domestic mothers and aggressive fathers showed significantly smaller motor activity only at the age of 50 days, thereby showing prolongation of the sensitive period. The maternal effect on behaviour formation contributes to the setting of time limits for the sensitive socialization period in foxes.

Introduction

Domestication has profound and many-faceted effects on the behaviour of animals. Particular emphasis has been on the modified response of domestic animals towards man. It has been observed that aggressive and fearful responses shown by wild animals have been gradually replaced by emotionally positive responses towards man under the effect of domestication (*Belyaev, 1962, 1974; Hediger, 1968; Hale, 1969*).

Many species have periods during which they are sensitive to particular environmental influences. What the animal learns during these sensitive periods usually affects it for the rest of its life. It is well known that in many mammals early experience during the period of primary socialization affects subsequent social adjustment (*Scott, 1962*). It has been demonstrated in numerous experiments with animals, including husbandry animals, that early handling, gentling and nursing in a rich environment significantly affects behaviour in adulthood. In

regard to foxes, handling during different periods of postnatal development decreases fear responses to humans and novel stimuli and reduces stress sensitivity in adult farm-bred silver foxes (*Pedersen, Jeppesen, 1990; Pedersen, 1992*). In her studies, Vasilyeva (1991) has also established higher values for domestic traits in adult foxes which had been handled early and selected for tame behaviour. In female foxes, competition capacity, which is correlated with reproductive function in later life, is closely related with the sensitive period of primary socialization (*Bakken, 1992*). The duration of this period changes under the effect of domestication. Thus, it is limited to 6-7 weeks of life in the wolf (*Woolpy, Ginsburg, 1967*), and 10-12 weeks in the dog (*Freedman et al., 1961; Scott, 1962*).

We have previously demonstrated that the sensitive period of primary socialization is restricted to 40-45 days of life in farm-breed silver foxes (*Vulpes vulpes*) while in pups from the population of domesticated foxes selected for tameability, the sensitive period is prolonged to over 60-65 days of life (*Belyaev et al., 1985*). Thus, domestic foxes do not show the defensive responses, which set a limit on this sensitive period, at the time when their counterparts not selected for domestication, are overtly defensive.

Also, the results of reciprocal crosses of domesticated to aggressive foxes show evidence for early maternal effects modifying the physiological threshold of the defensive responses (*Trut, 1980a*).

The role of maternal influences on physiological mechanisms limiting the sensitive period of socialization, such as the time of eye opening and the time of appearance of the defensive response to novelty, in hybrid offspring from cross matings between domestic and aggressive foxes in analyzed in this paper.

Materials and methods

The subjects were silver foxes, the first (F_1) generation from reciprocal crosses between domestic and aggressive foxes. For the experimental crosses, foxes were taken from two populations, one selected for tameability (domestic behaviour), and the other selected for enhanced aggressiveness (A). All the foxes were bred at the Novosibirsk Experimental Farm of this Institute. Selection for domestic behaviour (D foxes) has been carried out for 30 generations and a population of foxes very similar in behaviour to dogs was established (*Belyaev, 1979; Belyaev and Trut, 1982*). Selection for outstanding aggressiveness (A foxes) towards humans dates from 1970 (*Trut, 1980a, b*).

Our description of the expression of aggressive-fearful behavioral patterns conformed to that of Fox (1971). The observed pups were obtained by cross-matings: 92 (DA) from 20 litters produced by a domestic mother x an aggressive father (DxA) and 73 (AD) from 16 litters produced by an aggressive mother x a domestic father (AxD). The groups representing the parental generation showed very low variation in the traits.

After weaning at the age of 45 days, the pups were kept in litters to the age of 60 days. Thereafter each pup was housed in a separate cage. Each was marked individually at the age of 10 days. The timing of eye functioning from the appearance of a narrow slit to full opening of the eyes was registered individually.

We established behavioral patterns by observing exploration, fear response in a novel situation from days 20-60 of age after each 10 days (*Belyaev et al., 1985*). Exploratory behaviour was scored by placing a pup alone in a new cage for 10 min. Scored were the latency to move (sec), response to sound presented 3 times for 10 s during the test, locomotion type (central or peripheral).

The determination of the fear response to novel stimuli in pups was based on a sharp decrease in locomotor activity and exploration; the appearance of the "freezing" response in novel surroundings. Aggressive responses had various manifestations, including vocalizations, assumption of threatening postures and jump attacks.

The final estimate of behaviour towards humans was obtained at the age of 6-7 months; all foxes were tested at the same time of the day in the home cage. Domestic and aggressive-fearful behaviour towards humans were estimated individually by registration based on a subjective four-point score scale (for more detailed description, see *Trut, 1980b; Plyusnina et al., 1991*). As a result, the

behavioral phenotype was determined on a subjective scale as either domestic (from +0.5 to +4) or aggressive (from 0 to -4).

The data were subjected to analysis of variance, and intra class correlations were calculated with differences between litters taken into account. The two-way ANOVAs (cross-mating, i.e. DxA or AxD type x behavioral offspring phenotype) for the variables of run number and total locomotion time were performed. The data were tested for significance by the Student's t-test. The χ^2-test was applied to the treatment data for adult foxes showing domestic or aggressive-fearful behaviour towards humans (*Snedecor, Cochran, 1967; Rokitski, 1978*).

Results

The results of reciprocal crosses of domestic to aggressive foxes (fig. 1) were the same as previously reported (*Trut, 1980a*).

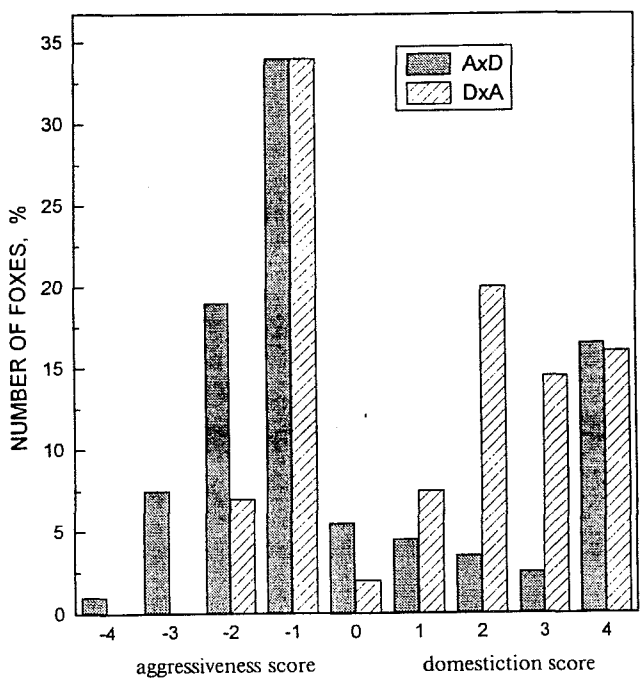

Fig. 1. Distribution according to behaviour of offspring from reciprocal crosses between domestic (D) and aggressive (A) foxes.

The F_1 offspring showed a wide variation in behavioral responses towards humans. The number of domestic individuals (60%) was significantly higher in crosses from domestic females to aggressive males than *vice versa*: the percentage of aggressive pups (64%) was higher in offspring from aggressive mothers and domestic fathers ($P<0.01$, χ^2-test). The domestic DA did not differ significantly in the degree of tameness from the domestic AD hybrids ($+2.3\pm0.14$ and $+2.5\pm0.29$, $P>0.05$, respectively). The aggressive DA showed significantly lower aggressiveness towards humans compared to the aggressive AD hybrids (-0.6 ± 0.07 and -1.2 ± 0.12, $P<0.001$, respectively). An interpretation of these results has been given elsewhere (*Trut, 1980a*).

The significant difference in the eye opening time was the largest for the domestic offspring from the two cross matings, and the aggressive offspring from the two crosses tended to differ in this parameter (table 1).

Sex differences for opening of the eyes were not observed. The eyes were fully opened in all the pups by day 20 (table 1).

Variance analysis demonstrated significant correlation coefficients between sibs (r_w) both for the time of the beginning of eye opening and full eye opening. It should be noted that r_w for the time of full eye opening between sibs from domestic mothers is almost twice that r_w between these sibs for the time of the beginning of eye opening ($r_w=0.41$, $P<0.001$ and $r_w=0.20$, $P<0.02$, respectively). The r_w between sibs from aggressive mothers does not differ in the time of the beginning and full eye opening ($r_w=0.48$, $P<0.001$ and $r_w=0.52$, $P<0.001$, respectively).

When placed in a new cage at the age of 20 days, all the hybrid pups behaved similarly; their orientation was the usual infantile and they did not differ significantly in parameters of locomotor activity (figs. 2-3). When encountering new surroundings at the age of 30 days, their behaviour was mostly exploratory and associated with increased locomotion. However, run number and total locomotion time were significantly higher in domestic than aggressive offspring from the two cross-matings (figs. 2-3).

Table 1 The time of eye opening in pups from reciprocal matings of domestic (D) and aggressive (A) foxes

♀D x ♂A				♀A x ♂D			
domestic		aggressive		domestic		aggressive	
N	x ± S.E.	N	x ± S.E.	N	x ± S.E.	N	x ± S.E.
First appearance of eye narrow slit (days)							
51	15.6±0.18	41	15.4±0.21*	23	16.4±0.4	49	15.9±0.24
Full eye opening (days)							
51	17.8±0.15*	41	17.7±0.20*	23	18.7±0.28	49	18.1±0.20

* $P < 0.01$ compared to domestic offspring from AxD, Student's t-test

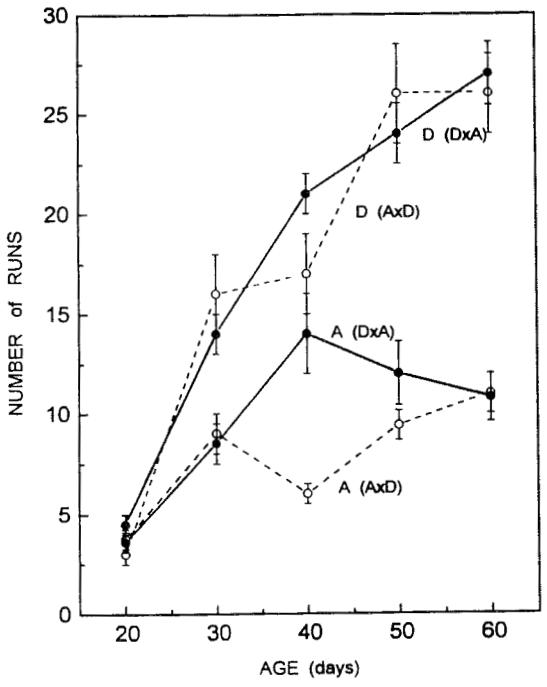

Fig. 2. The time course of changes in number of runs in domestic (D) and aggressive (A) pups from reciprocal crosses of domestic and aggressive foxes, solid line: DxA, dashed line: AxD.

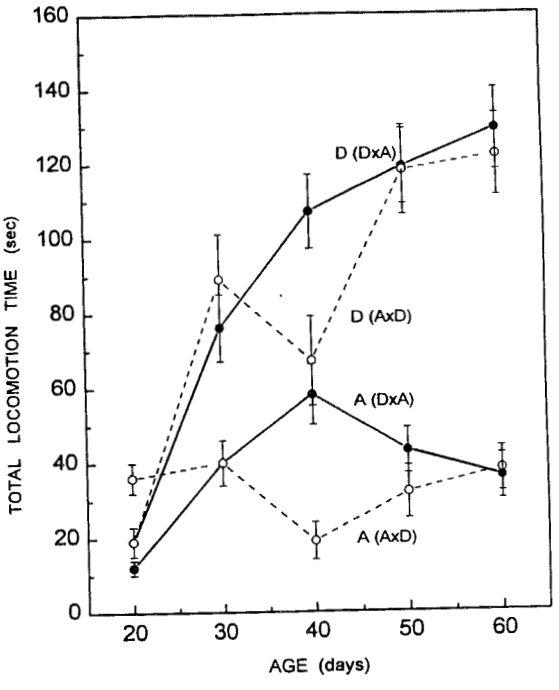

Fig. 3. The time course of changes in total locomotion time in domestic and aggressive pups from reciprocal crosses of domestic and aggressive foxes; the designations are the same as in fig. 2.

When tested at an older age (40, 50, and 60 days), all domestic pups, regardless of the cross-mating type, showed the characteristic exploratory response to the novel environment and sound stimuli; the locomotion parameters increased. However, the domestic AD exhibited attenuated exploratory behaviour at the age of 40 days compared to the domestic DA pups. As a result, total locomotion time for the former was significantly lower than for the latter pups. The aggressive AD offspring started to display defensive behaviour in a novel environment; the pups rapidly ran to avoid the center of the cage, and their locomotion was concentrated to its periphery. As a result, their total locomotion time was significantly lower at the age of 30 and 40 days (fig. 3). The pattern observed for the aggressive DA offspring was different; at the age of 40 days, run number and total time of locomotion of these offspring were significantly higher than in that from AxD mating. It is noteworthy that aggressive pups from the two cross-matings did not differ significantly at the ages of 50 and 60 days. When placed in a new cage, their brief exploration alternated with aggression: they assumed aggressive postures, growled, and jumped to assault the sound source.

The domestic and the aggressive pups from matings of both types did not differ significantly in latency to move from day 30. They all responded by fear at the beginning of the test in the new cage.

Two-way ANOVAs demonstrate a statistically significant effect of both crosses DxA or AxD and behavioral offspring phenotype (i.e. domestic or aggressive) for the variables run number ($F=12.4$, $P=0.001$ and $F=30.7$, $P<0.001$, respectively) and total locomotion time ($F=16.3$, $P<0.001$ and $F=26.2$, $P<0.001$, respectively) only at the age of 40 days. Next, the effect of offspring behavioral phenotype is significant only for these variables at all ages except for 20 days ($P<0.001$ for both variables). Finally, there is no significant cross-mating type x offspring phenotype interaction at all ages.

Discussion

As well known, the age of eye opening, along with the development of other sensory systems and locomotor activity, are of importance at the early steps of the establishment of social relationships (*Scott, 1962*). We have previously shown that domestication is associated with earlier full eye opening in foxes (*Belyaev et al., 1985*). The present results suggest that early maternal environment may affect the eye opening time. The effect, however, is dependent on offspring behavioral phenotype. It should be emphasized that we have tested heterozygous F_1 offspring from reciprocal crosses between domestic and aggressive foxes. The present data, in continuation of the previous data (*Trut, 1980a*), demonstrate, contrary to expectations, that the F_1 offspring showed extreme variation in behaviour ranging from aggressive to domestic. The heterozygous F_1 offspring from domestic mothers showed domestic behaviour by opening their eyes significantly earlier than offspring also showing domestic behaviour, yet born to aggressive females; a tendency towards a somewhat later eye opening was observed in aggressive offspring from aggressive mothers compared to the aggressive from domestic foxes. We have previously provided evidence indicating that the wide variation in the behaviour of F_1 offspring is unrelated to the segregation of genes largely contributing to the trait. This variation can be explained by multiple minor effects of a modifier gene on the physiological threshold of behavioral responses and also modification of this threshold by the maternal environment (*Trut, 1980a*). The fact that the influence of the early maternal environment of the females is different in offspring of different behavioral phenotypes suggests that the minor effects of modifier genes contribute to the variations in the behaviour of the F_1 offspring. The influence of the maternal environment on eye opening time depends to some extent on these modifier genes.

The contribution of behavioral phenotype to the development of the response to novelty is manifested in pups from reciprocal crosses from day 30. Furthermore, significant maternal effects on behavioral responses in novel surroundings are observed only at the age of 40 days (see figs. 2 and 3). We regard this age of 40 days as critical for the formation of the defence response in foxes. Our previous results are relevant (*Belyaev et al., 1985*). Thus, the behaviour of aggressive foxes in an unknown situation is exploratory up to 35 days of life; the next developmental period tested was 40 days, and all estimates of exploratory behaviour are sharply decreased in aggressive foxes. This was

evidence of the enhancement of the defence response to novelty from 34-40 days of life.

An important consequence of maternal effects is that differences in locomotion parameters between reciprocal hybrids of the same behavioral phenotype are observed only at the age of 40 days. Indeed, domestic offspring from AxD matings showed significantly lower total locomotion time than their counterparts from DxA matings precisely at this age. Heterozygous offspring with aggressive responses from the two mating types likewise differed in both parameters of locomotion. Its values were higher in aggressive pups from DxA matings at the age of 50 days. These results demonstrate a prolongation of the socialization period in aggressive pups from DxA mating. Evidence for the effects of prolongation is the significant reduction in aggressiveness score of these aggressive pups in adulthood. In this line is also our previous evidence according to which the weaker the defensive-aggressive response to humans in adult foxes, the longer should have been their primary socialization period (*Belyaev et al., 1985*). It thus appears that we are dealing here with a case of interaction between maternal and offspring genotypes and a contribution of this interaction to the formation of behaviour during development.

Maternal effects have previously been demonstrated for many behavioral and physiological traits in animals at different phylogenetic levels (*Ponomarenko et al., 1975*). To our knowledge, studies with wild and domestic Norway rats concerning maternal effect on behaviour formation include two comparable responses towards humans (*Richter, 1954; Barnett, 1960*) and another describing open field behaviour (*Price, Loomis, 1973*). The responses towards humans were inconsistent: the hybrids were either behaviorally domestic (*Richter, 1954*) or, the reverse, wild (*Barnett, 1960*). As for open field behaviour, all the reciprocal hybrids were intermediate with respect to parental behaviour (*Price, Loomis, 1973*). Relevant are the effects of cross-fostering; when tested in adulthood, cross-fostered rats behave in open field more like their native mothers (*Galef, 1970; Hughes, 1975*). These results were taken to mean that, whatever the postnatal maternal effects may be, they are minimal compared to the genetic effects in adults. However, when tested at an early age, cross-fostered rats exhibited foster mother effect on defecation rates and total activity (*Hughes, 1975*). The maternal effects on the response to novelty brought out in aggressive foxes from DxA mating at the age of 40 days was observed, when their behaviour towards man was tested in adulthood.

In discussion of the nature of these effects, genomic imprinting cannot be excluded. It should be recalled that genomic imprinting implies the influence of the parent on gene expression in offspring (*Baranov, 1988*). With respect to our results, this would mean that the heterozygous offspring, which receive "the domestication genes" from the mother, differ in many behavioral parameters from the offspring which probably received them from the father. Other genetic phenomena may be involved in determination of maternal effects. However, their consideration is beyond the scope of this study.

Acknowledgements

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Original Report

Platform use by juvenile and adult silver foxes (*Vulpes vulpes*)

Hannu Korhonen, Paavo Niemelä

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Abstract

The European Convention has issued the recommendation that each weaned fox shall have some kind of platform or nest box in its cage. In the present study, use of a U-type platform (wooden, slightly U-shaped bottom; area 3090 cm^2) was studied in juvenile and adult silver foxes (*Vulpes vulpes*) from August to December. Video recordings revealed that use was high in July-August when juveniles were on platforms on average 1108 $\pm$ 44 min/24 h (mean $\pm$ SE). Thereafter, usage dramatically declined ($p < 0.001$), reaching a low level in November (255 $\pm$ 72 min/24 h). Daily scan sampling observations gave parallel results. Platform use was sex-related, being typically higher in females than males. According to the scan sampling observations, platform use in adults (4.7% of total observations on platforms) was significantly lower ($p < 0.001$) than that of juveniles (47.2%). Circadian variations in platform usage were highest in July-September, but later became very slight because of low usage on the whole. In late autumn, daily platform use declined with decreasing daily temperature and *vice versa*. This relationship was most significant in October. The main function of platforms was not that of an observation site alone, as over 80% of total platform time was spent sleeping. The platforms remained rather clean, without any visible damages to the fur or wellbeing of the animals. In conclusion, the present studied platform type was found to be suitable for juvenile silver foxes.

Introduction

It has recently been emphasized that traditional housing conditions where foxes are kept in bare wire-mesh cages are obviously insufficient in terms of animal welfare not only because the animals are exposed to seasonal changes in environmental conditions, but also because bare, unenriched cages do not provide enough activating stimuli. The Standing Committee of the European Convention on the Protection of Animals Kept for Farming Purposes has therefore issued the requirement that each weaned fox shall have a whole-year shelter, equipped with either a resting platform or nest box (*European Convention, 1991*). Compliance with the recommendations is problematic, however. Firstly the recommendations are too general in form in spite of the fact that the results of many shelter experiments lead us to suppose that platform use is very variable, and depends at least on the construction model, age of the fox, individual, season and country (*Valtonen & Moss, 1983; Hoffmeyer, 1986; Sønderup, 1986; Harri et al., 1991; Mononen et al., 1993; Pedersen & Jeppesen, 1993; Korhonen & Niemelä, 1994*). Secondly, platforms and nest boxes do not have exclusively positive welfare

effects in terms of lower stress or fear (*Jeppesen & Pedersen, 1991, 1992*), but at least in some construction models can cause poorer fur quality and ventral wearing, or even be a potential source of health problems like urinary tract infections (*Harri et al., 1991, 1992; Korhonen & Niemelä, 1991a, 1995*). There is also recent evidence (*Pedersen & Jeppesen, 1993; Korhonen & Niemelä, 1994*) that platform usage may differ between two different fox species, the blue fox (*Alopex lagopus*) and the silver fox (*Vulpes vulpes*), whereby the amount of use appears to be lower in the former one. The current European recommendations appear, however, to treat both species equally. Additional platform experiments are therefore needed to clarify whether there is a need for further reconsideration of the recommendations, including more species-specific guidelines.

The purpose of the present study was (1) to measure the extent of platform use by silver foxes (*Vulpes vulpes*) as evaluated by both video and scan sampling methods. The present studied platform type was a large wooden platform with a slightly U-shaped bottom which has previously been found to be superior in the case of blue foxes (*Korhonen & Niemelä, 1994*). Therefore, it was assumed that this type would also be the most suitable for silver foxes. Earlier silver fox experiments have been mainly carried out with either small flat-bottomed platforms (*Mononen et al. 1991, 1993*) or constructions where the flat platform formed part of the entrance to one of the three nest boxes within the cage (*Pedersen & Jeppesen, 1993*).

A second aim (2) was to find out to what purpose platforms are used. This point requires further clarification because, according to previous studies, the main function of the platform is an enrichment (*Mononen & Harri, 1991; Mononen et al., 1993; Korhonen & Niemelä, 1994*).

As an additional function, (3) we studied to what extent platforms are used for defecation or as a biting object. Finally, we sought (4) to compare the differences in platform use between adult and juvenile silver foxes without any previous platform experience. Recent studies on blue foxes (*Korhonen & Niemelä, 1993a, 1994*) support the assumption that previous platform experience would be crucial for later platform use.

Materials and methods

Animals and general management

The present study was carried out at the Fur Farming Research Station of Kannus, in western Finland during 1993. Adults (20 males, 30 females; born in May 1992) and juveniles (31 males, 29 females; born in May 1993) were used in the experiments. The animals had no previous platform experience. They were housed in two-row sheds, with two (juveniles, a male and female together) or one (adult) occupying one wire-mesh cage measuring 107 cm wide x 110 cm long x 70 cm high. Each animal was ear-tagged for identification. Fresh-mixed fox feed manufactured by the local feed kitchen was supplied twice a day (at 9.00 a.m. and 1.00 p.m.) from July to mid-September in the case of juveniles, and thereafter once a day (1.00 p.m.) with a feed machine. Adults were fed once a day (1.00 p.m.) throughout. Feed portions (from 400 to 800 g/animal/day) were based on the conventional feeding standards of the Finnish Fur Breeders' Association. All animals remained healthy throughout the experiments. Ambient air temperatures were read at 8 a.m., 12 a.m., and 3 p.m. from a conventional thermometer which was placed inside the studied shed.

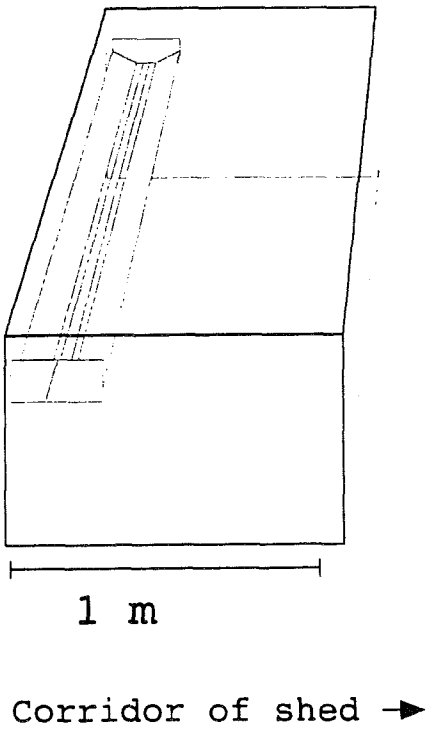

Fig. 1. Schematic picture of the platform type studied. For details see Materials and Methods

Platforms

Schematic picture of the platform type studied is given in Fig. 1. The experimental platforms (103 cm long x 30 cm wide; area 3090 cm^2) used were made of wooden material (5" board, thickness 22 mm) and had walls (10 cm high) at the ends only. Their bottom slightly resembled the shape of the letter U (maximum depth 2 cm). The platforms were placed in the shorter side of the cage crosswise in the shed. Their distance from the cage roof was 23 cm. The platforms were cleaned once a week to remove faeces and urine. At the same time, the number of dirty and bitten platforms was recorded.

Monitoring platform use

Platform use was monitored by two means: (1) continuous 24-hour measurements by video camera equipment (CCD video camera 720, Bische UB-480 tape recorder, Koyo monitor, Bische 12-300 infrared light: 500 W). Altogether 10 males and 10 females were randomly selected for the video recordings. They were taped during three periods of the study, i.e. in July-August, September and November.

(2) Daily use by scanning observations which were carried out three times a day (8 a.m., 12 p.m. and 3 p.m.) during workdays (July-December in juveniles, August-December in adults). The only exception was Friday when observations were done only at 8 a.m. and 12 p.m. because farm work ended at 2 p.m. The total number of observations per fox amounted to 56 each month. The month was considered to be the basic unit within which platform use comparisons were made.

Platform use by this method is thus based on counts expressed as percentages of the total monthly scan sampling. For the scan sampling observation the experimenter walked quietly and slowly past the row of cages and manually recorded the location of the fox (on the platform or not) (*Pedersen & Jeppesen, 1993; Korhonen & Niemelä, 1993a*).

When standing in front of cage no. 2 the location of the fox in cage no. 3 was recorded and so forth. If the fox fled from the experimenter, the location of the fox before it fled was recorded.

Statistics

Data were analyzed using the SAS package (*SAS, 1988*). The differences in platform use between sexes and age groups were analyzed with the Mann-Whitney U-test.

Regression analyses were used to calculate relationships between platform use and ambient air temperature. Video tape data were analysed using multivariate analysis of variance (MANOVA).

Although the juvenile foxes were ear-tagged, identification directly from the video tapes was impossible. Therefore, as it was not possible to separate platform use between the sexes within the same cage, the data were analyzed per cage. Scan sampling material, however, was possible to present also by sexes. Data were normalized using arcsine transformation.

Results

Platform use by scan sampling

Adults used the platforms significantly less ($p < 0.001$) than juveniles during each study month (Table 1). In the case of juveniles, there were no differences in platform use between sexes in July and August. After that, however, usage was lower in males than females. Adult females used the platforms more than males (Table 1).

Platform use by juvenile foxes was highest in July-August but declined significantly thereafter ($p < 0.001$) up to the onset of the pelting time. In adults, platform use was highest in September, after which a clear decreasing trend was observed (Table 1).

Platform use by video recordings

The general results obtained for platform use were by video recording observations (Table 1). Thus, according to video recording, platform use in juveniles was also highest in July-August, but decreased thereafter. In November, usage was already significantly ($p < 0.001$) lower than in July-August. In addition, the video data revealed that actual time spent on the platforms was very high in July-August because the foxes spent about 77% of their daily 24 h on them. In November, the time spent on platforms dropped radically to around 18% per

24 h (Table 2). During the first recording period, usage was normally distributed as indicated by the medians in Table 2.

However, in November platform use was no longer normal; there were still some individuals that used the platforms to a large extent but, on the other hand, most of the foxes preferred the cage floor.

Platforms were mostly used for sleeping and the least for jumping (Table 2). The actual time spent on the platforms dropped dramatically from July-August to November. The proportion of time spent sleeping on platforms from the total time budget remained almost the same between the two first periods (88.9% vs. 89.1%), but decreased slightly thereafter (to 81.6%). The proportion of lying on platforms stayed rather constant over the two first periods (9.6% vs. 9.0%) but increased thereafter (to 14.9%). The proportion of jumping behaviour showed an increasing trend over the study periods (1.5% vs. 2.0% vs. 3.9%, respectively).

Circadian distribution of platform use showed a rather similar profile between the first two observation periods (Fig. 2). In the morning hours of the working day (8:30-11:00 a.m.) use typically decreased, but increased again quite soon thereafter.

Table 1 Platform use (% of observations on mean ± SE) in both sexes of juvenile and adult silver foxes. Juveniles were housed in pairs and adults singly. Data are based on scan sampling observations. Statistical comparisons are given between sexes of the same age. *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$ (Mann-Whitney U-test)

	JUVENILES		ADULTS	
	Males	Females	Males	Females
July	88.9 ± 0.8	90.3 ± 0.8	-	-
August	66.7 ± 1.1	67.3 ± 1.1	3.6 ± 1.2	7.5 ± 0.9*
September	44.2 ± 1.1	51.6 ± 1.2***	6.8 ± 0.9	10.9 ± 0.6**
October	30.6 ± 1.1	39.1 ± 1.2***	3.1 ± 0.6	7.2 ± 0.5**
November	23.4 ± 1.0	26.2 ± 1.0*	0.5 ± 0.3	4.1 ± 0.4***
December	15.8 ± 1.3	22.8 ± 1.6***	0.7 ± 0.5	2.9 ± 0.5*

Table 2 Distribution of platform use (min ± SE/24 h) in juvenile silver foxes. Medians are given below means. Data are based on video recordings of 10 males and 10 females which were summarized due to difficulties in differentiating sexes in the video tapes. Lying indicates a short duration (1-10 min) on the platform

Period	Sleeping	Lying	Jumping	Total use
26.7.-2.8.	985 ± 44	106 ± 6	17 ± 1	1108 ± 44
	1004	94	17	1114
15.9.-22.9	820 ± 68	83 ± 10	18 ± 2	920 ± 72
	841	81	17	970
18.11.-23.11.	208 ± 63	38 ± 10	10 ± 2	255 ± 72
	51	10	7	121

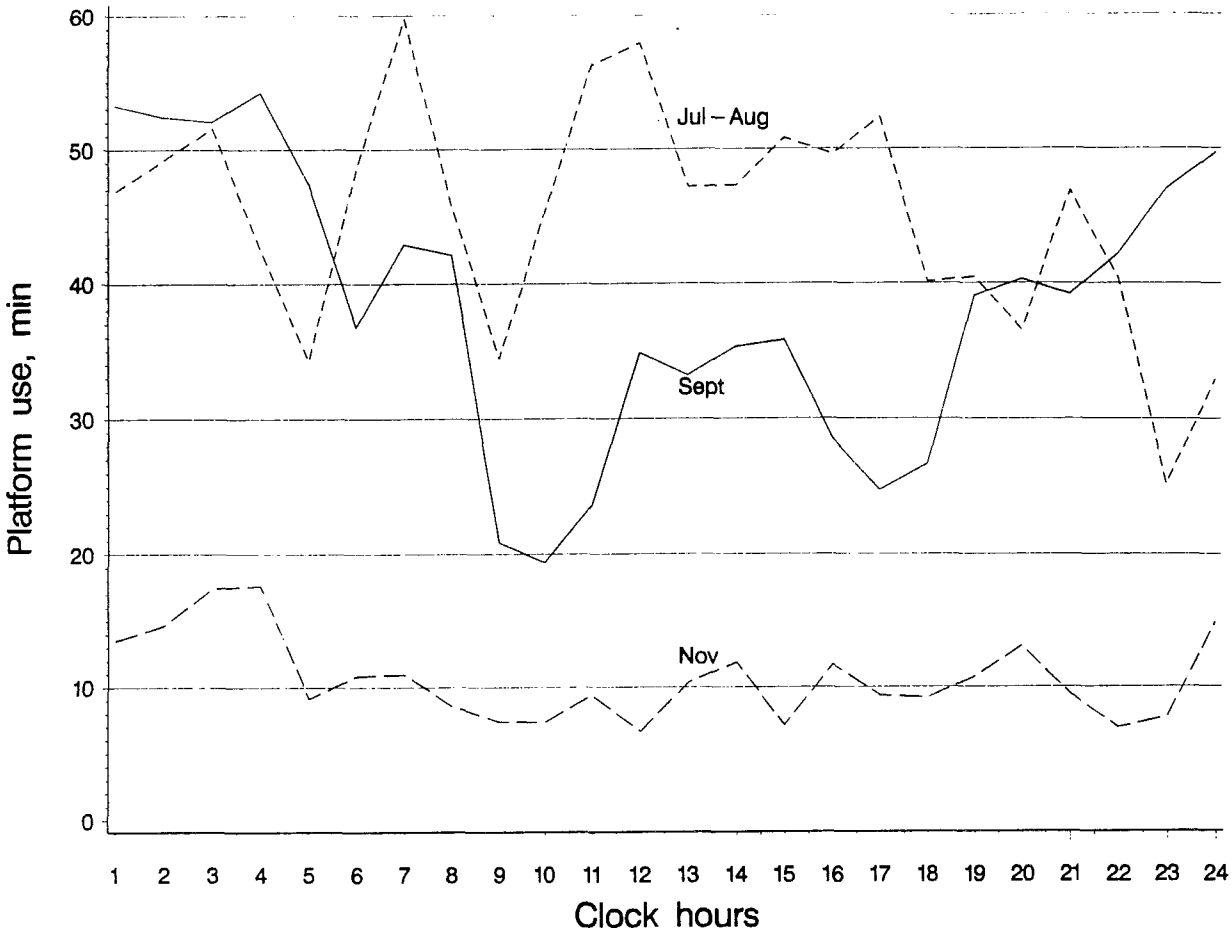

Figure 2 Circadian distribution of platform use (min.) in juveniles during three study periods. Data are based on video recordings of 10 male and 10 female silver foxes.

There also appeared to be some decrease in platform use after the end of the working day, at about 4 p.m. During the second period (September), use was lower during working hours than outside them. In November, the circadian use of platforms as a whole was rather low, with minor hourly variations.

Use in relation to temperature

During July-September, when the monthly mean temperatures ranged from +18.9 to +6.5°C, respectively, no dependence between platform use and ambient air temperature was found. However, in October (monthly mean +2.7°C), there was a significant relationship between platform use (Y) and ambient air temperature (X), both in juveniles ($Y=1.39X + 30.8$; $p<0.001$; $F=27.03$; $R^2=0.59$) and adults ($Y=0.89X + 3.82$; $p<0.001$; $F=28.45$; $R^2=0.60$). In November (monthly mean -2.7°C) a statistically significant relation was also found in juveniles as follows: $Y=0.43X + 24.1$; $p<0.037$; $F=4.74$; $R^2=0.14$), but not in adults. Towards pelting time the relation became weaker because platform usage in general declined dramatically. This was especially seen in adults with their minimal platform use.

Dirtiness and damage of platforms

The platforms remained rather clean throughout the study (Table 3). In juveniles, dirtiness increased somewhat towards winter, but not to a large extent. The reverse situation was found in adults, i.e. the amount of dirty platforms even decreased with time from August onwards. Because our experimental animals were not pelted (they were left for breeding), we do not have any accurate results on their fur quality parameters. However, a general visual overview of live animals' fur did not reveal any noticeable damage or dirtiness.

Platform biting increased throughout the study months, and in October only a minimal part of the platforms remained totally unbitten (Table 3). Nevertheless, the degree of biting was generally low and none of the studied platforms required replacement up to the end of the experiment.

Table 3 Degree of platform dirtiness (% of animals) and amount of bitten platforms (cumulative %). *p < 0.05; ***p < 0.001 (Mann-Whitney U-test)

	Juveniles	Adults
Clean platforms (%):		
July	100.0	-
August	98.9	91.7*
September	88.7	93.3
October	95.8	99.1
December	83.3	96.4*
Unbitten platforms (%):		
July	100.0	-
August	84.4	91.7
September	41.3	22.3***
October	24.2	19.0

Discussion

A previous platform experiment at the Fur Farming Research Station of Kannus showed that silver foxes used platforms on average 70 ± 10 min/24 h (*Mononen et al., 1993*). This is a significantly lower amount than that (761 ± 52 min/24 h) found in the present study. There are probably several explanations for the large difference in the results of these studies. Firstly, technical reasons can affect use. In the former experiment (*Mononen et al. 1993*) the platform size was very small, i.e. 54 cm long x 33 cm wide (area 1782 cm^2), while that in the present study was substantially bigger (3090 cm^2). Results of previous experiments with blue foxes have already indicated that platform use appears to be dependent on platform size (*Korhonen & Niemelä, 1993a, 1994*). Platform ceiling is another influencing factor. Distance of the platform from the cage roof in the silver fox experiment of Mononen et al. (1993) was 20 cm but in the present study it was 23 cm. The previous experiments in which the effects of ceiling have been compared have revealed that ceiling significantly influences use so that use is increasing with decreasing platform ceiling and *vice versa*. A ceiling smaller than 23 cm appears to be crucial in platform use (*Harri et al., 1988; Korhonen & Niemelä, 1994*). The time when the platforms were provided can also explain some part of the differences in platform use. In the present study, the platforms were provided to juveniles already after weaning in early July, whereas in the experiment of Mononen et al. (1993) the foxes received platforms in the later part of August. In blue foxes, it has been quite clearly shown that the later the platforms were given after weaning, the less they were used (*Korhonen & Niemelä, 1993a*). Likewise in the present study, a very significant difference in platform use was found between animals that had access to platforms soon after weaning (juveniles) compared with those that did not have their first platform experience before adulthood (adults).

Platform use of the presently studied foxes decreased markedly from summer towards winter. Parallel results have also been found in other silver fox (*Mononen et al. 1991, 1993*) and blue fox (*Korhonen & Niemelä, 1993a, 1994*) experiments. In addition, a year-around study in blue foxes (*Korhonen & Niemelä, 1994*) has revealed that platform use varies seasonally very significantly, being lowest during the cold season of the year (winter) and highest during the warmest season (summer). It is not exactly known by what mechanism season affects use, but one explanation could be the fact that with decreasing temperature the platform bottom becomes colder and thus more uncomfortable to lie on. Animal's heat loss from a frozen surface is also higher than that from an empty cage floor (*Korhonen, 1987*). The recent results provided additional evidence that the relation of platform use to ambient air temperature is the crucial explanation behind changes in seasonal platform use. Clarification of such a theory would require an experiment where platform usage could be compared between two groups housed at two different temperatures simultaneously.

The recommendations of the European Convention (1991) imply that the needs for resting, observing and hiding should be satisfied by a platform. The presently studied platform type seems to fulfil these needs because both long (sleeping) and short (lying, jumping) time activities were encountered. Sleeping can be considered to be an expression of resting use. We are also assuming that when a fox was on the platform a short period of time (< 10 min), it was for observing. The question as to what extent platforms can serve as a hiding place is more difficult to answer, however. If a fox jumps onto a plat-

form for a short time or if it remains there for a longer time, can we interpret both or even one of these two activities as an expression of hiding behaviour? Answering this would require more specific knowledge about what was the crucial impetus for the use in question.

Understanding the main function of platforms on the basis of the present results is problematic. Those of our foxes that were video recorded used 82-89% of their platform time for sleeping. As a result, the amount of short-term use remained quite low. The relations between these short and long-term functions remained rather constant from summer to winter, although a slight decreasing tendency in sleeping use towards colder periods was evident. These results agree well with the previous results obtained on blue foxes studied in farm cages (*Korhonen et al., 1994*). Thus, it is obvious that the main function of the platform is not that of an observation site alone, although such speculations are available (*Mononen & Harri, 1991)*, because foxes commonly used the platforms for sleeping. As the use in general declined very sharply towards winter, the platform does not serve as a shelter against cold weather either (*Mononen et al., 1993*). On the contrary, it is tempting to ask if platform could be a shelter against warm weather because foxes spent so much of their summer time on the platforms. At least the platform is located higher than the cage floor and is, therefore, more in shade. In the present study, however, no relationship between daily temperature and platform use was found in the warmest summer (July-September). Clarification of this hypothesis requires more experiments. According to Mononen & Harri (1991), the platform may also serve only as an environmental enrichment which reduces the barrenness of the cage environment, and thus the animals are satisfied in the awareness that the platform is available for their occasional use. However, it is difficult to completely concur with this explanation because of such intensive use with large seasonal variations. It is also possible that the platform has different functions in different seasons. Further studies, particularly year-round ones, will be needed in order to clarify the above points. Evidence that platforms can serve as biting objects for farmed silver foxes was found. Obviously, like many dogs, farmed foxes appear to look for something to gnaw on and since the farm cage is otherwise barren, the wooden platform automatically fulfils that need. However,a piece of extra wood for gnawing placed on the cage floor might prevent platform biting and thus keep platforms in better condition. The extent to which platform biting is a signal of some stress or frustration is difficult to estimate. In some farm animals, however, it is known that at a certain level of excitement, and where adequate stimulus is lacking, for instance, eating behaviour can proceed on a substratum which in itself is unsuitable for food intake, but which has the advantage of being bitable and chewable (*Sambraus, 1985*). This explanation might also be applicable to the present observations because during late autumn the feed availability to our farmed foxes, as is the conventional practice, was restricted to avoid excessive obesity. Moreover, we observed that platform biting in blue foxes (*Korhonen & Niemelä, 1993b*) markedly increased when feed portions were restricted during winter.

The present results showed that both juvenile and adult silver foxes used platforms as a defecation site only to a minor extent. Thus, it is obvious that platform dirtiness will not cause marked problems in commercial silver fox farms, assuming that platforms with a ceiling height of 23 cm are cleaned regularly. The frequency of once-a-week cleaning appears to be sufficient according to the present experience. In platform experiments with blue foxes (*Korhonen & Niemelä, 1993a, 1995*), animals have been observed to be fairly eager to defecate on a platform resembling the present constructions. In a shelter experiment with a combined platform-nest box housing system, Pedersen & Jeppesen (1992) also found that blue foxes defecated more in the shelters compared to silver foxes, both in the numbers of animals defecating and in the amount of faeces deposited. Thus, it is possible that the patterns and/or functions of defecation between the genera *Alopex* and *Vulpes* are somewhat different. Blue foxes may seek solid floors, such as platforms, for fixing their faeces which can then better act as scent-signals. Another explanation could be the fact that the body size of silver foxes (i.e. height and length) is larger than that of blue foxes, thus preventing the animals from achieving the appropriate defecation posture if the platform ceiling is about 23 cm from the cage roof. In blue foxes, it has been observed that animals defecated

on platforms when the ceiling was very low (30 cm), but defecation decreased when the ceiling was 23 cm high, and decreased even more when it was only 18 cm high (*Harri et al., 1988; Korhonen & Niemelä, 1994*). Further experiments in silver foxes with different platform ceiling heights would be required to justify this explanation. Use of the studied U-type platform by our silver foxes was significantly higher compared to that found in blue foxes with a similar platform model (*Korhonen & Niemelä, 1994*) when the comparison is made using video recording data (761 min/24 h vs. 244 min/24 h). In the experiments of Mononen et al. (1992, 1993; the former reference also includes material from Harri et al., 1991) platform use, based on automatic sampling by thermocouples placed on the platform bottom, was slightly higher in blue than silver foxes (98 min/24 h vs. 70 min/24 h). However, the platform size employed in those experiments was much larger in the former species. If the comparison of the present results is based on the visual scan sampling material, the platform use of our silver foxes was somewhat lower (47.2% vs. 56.1%) than that of blue foxes (*Korhonen & Niemelä, 1994*). Hoffmeyer (1986), who used only scan sampling, also observed slightly higher platform use in blue foxes compared to silver foxes (94% vs. 83%). The number of animals studied per species in the study of Hoffmeyer (1986) was very small, however, i.e. 18 blue foxes and 6 silver foxes. Using scan sampling, Pedersen & Jeppesen (1993) found that silver foxes used platforms more than blue foxes (estimated from the figures: 10% vs. 4%) in cages where both a platform and three nest boxes were available. Blue foxes preferred the nest boxes more. It should be kept in mind, however, that one crucial problem when comparing different studies is that the experimental arrangements typically vary considerably, e.g. in terms of platform type and placement in the cage, ceiling, cage size, season, farm, animal number and sex combination, age and previous platform experience, all of which can cause variation between different experiments (*Korhonen & Niemelä, 1994*). While previous data have been collected either by visual or automatic scan samplings, or by video recordings, it is not surprising that differences exist. For instance, the fact is that visual scan sampling and video recording methods partly measure different characteristics of platform use. Thus, video data indicate the exact amount of time a fox stays on a platform, but visual scan sampling data shows in what percentage of sampling observations the fox in question was on a platform. The latter method tends to provide more variable results because the presence of the observer obviously affects the animals' willingness to jump on or off the platform differently in different animal material. On the other hand, the difficulty with video recordings is that the animal number must be limited due to technical and analytical reasons. With visual scan samplings experimental group size is not a limiting factor. In the case of the automatic sampling by thermocouples (*Mononen et al., 1992, 1993*) one crucial problem is the fact that the short-term use of a platform cannot be measured reliably, because the temperature does not change before the animal has totally settled down on the platform. Thus, such a method easily underestimates the use. In view of the above, we propose that more data should be gathered before any final conclusions concerning possible differences in platform use between silver and blue foxes are made. In conclusion, it may be stated that the presently studied wooden platform type with the slightly U-shaped bottom, is suitable for juvenile silver foxes because (1) the animals use them frequently and (2) they do not cause observable problems in terms of platform dirtiness. As concerns adult silver foxes without previous platform experience, however, platforms cannot be considered to be necessary because of the very low amount of their use. Finally, the present study supports the premise that the present recommendations of the European Convention require further consideration.

Animal welfare implications

The recommendations of the European Convention (1991) imply that needs for rest, observation and seclusion can be satisfied by a platform. The presently studied platform type fulfilled these needs fairly well in juvenile silver foxes in view of (1) its high amount of general use, and (2) it functioned appropriately as a place for observation and rest. In addition, this platform type did not cause any visible damage to the fur or wellbeing of the animals.

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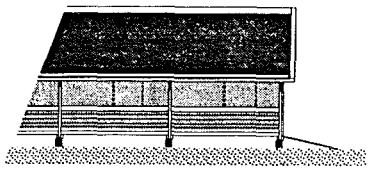
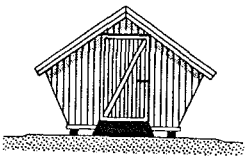
Environmental and economical relations affecting the design of buildings in fur animal production



Kjetil Aarstrand



New doctor in the family. We congratulate Dr. Kjetil Aarstrand with the fine result of investigating new fields in fur animal production.

The thesis is based on the following reports.

(Part 1). Effect of climate on fur animal production. A study of the literature.

The object of this literature study was to find dimensions for determining the effect of low temperatures and wind speed on energy requirement and fur quality. Data are presented from 32 references within the period 1950 - 1988. Because of the uncertain data material, it is only possible to suggest the levels of the different quantities:

Mink:
- Energy requirement of mink for main-tenance: 527.5 - 816.4 kJ/kg$^{0.75}$·day.
- Lower critical temperature (T_{lc}) of mink: 21-22°C.
- Increase in energy requirement at temperatures below T_{lc}: 9.6 - 15.5 kJ/kg·°C·day.

Fox:
- Energy requirement of fox for maintenance: 272.1 - 389.4 kJ/kg body weight·day.
- In early references T_{lc} of blue fox was estimated to -30°C or lower. Korhonen et al. (1985) claim that T_{lc} of blue fox is approximately -6°C.

Increasing wind speed seems to have a negative effect on the energy economy of mink. The insulation ability of the winter fur is reduced by 15-20% when wind speed increases from 1 to 5 m/s.

Norsk Landbruksforskning 1989, 3: 49-59. In NORW, 3 tables, 1 fig., 33 refs.

(Part 2). Effects of light on Fur Animal Production. Literature Review. *Scientifur, Vol. 15, No. 2, pp 130, 1991.*

(Part 3). Effect of different storage methods on qualities and chemical composition of manure from fur bearing animals. *Scientifur, Vol. 17, No. 1, pp 34, 1993.*

(Part 4). Short term effects of different water contents in feed on blue fox (*Alopex lagopus*) and silver fox (*Vulpes vulpes*). *Scientifur, Vol. 17, no. 1, pp 50, 1993.*

(Part 5). Building economics of fur animal production. A cost analysis of different types of buildings by the use of models.

This cost analysis uses three pre-defined building models for fur animal production. Capacity was set to 100 breeding vixens of blue fox. The three building-construction models compared were:

1) Open building with two rows of cages (O2R)
2) Enclosed building with four rows of cages (L4R)
3) Enclosed building with six rows of cages (L6R)

The price basis was the Norwegian HolteProsjekt building costs, and refer to September 1991.

Calculations for the three models showed that O2R had the lowest total building costs. Building costs for L4R was 15% higher and for L6R, 3% higher.

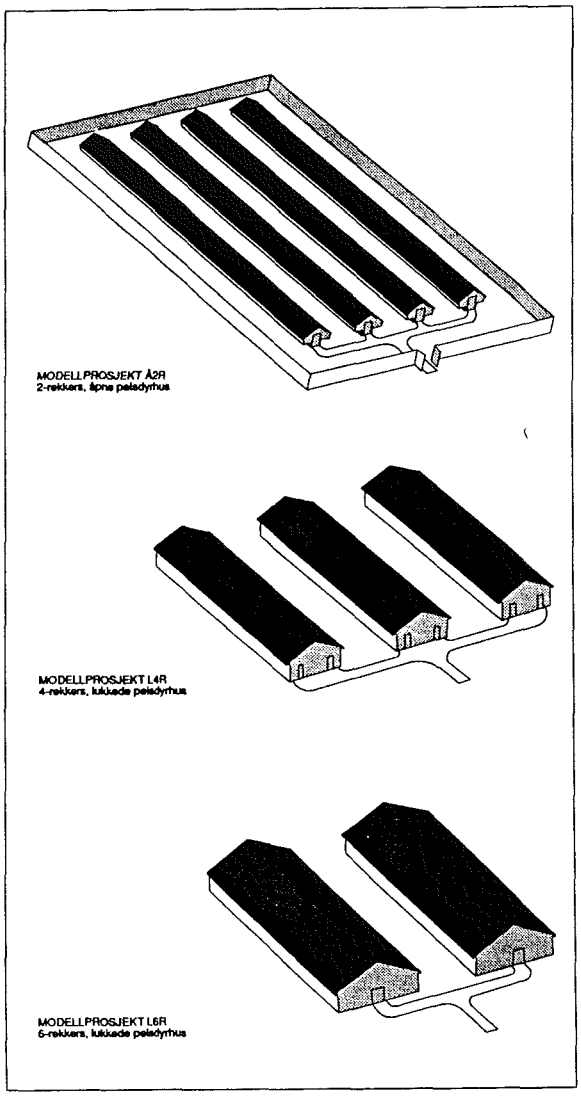


Fig. 2. Model projects used in calculating building costs.

Roof costs form 31-33% of the total building costs for all three models. Roof costs include eavestroughs. For the model O2R, eavestrough costs form 10% of the total building costs. Cages and indoor construction costs formed 22-25% of the total costs.

The total costs are distributed as follows: material costs (M) 62-65%, labour costs (A) 19-20% and other expenses (O) 17-18%. These figures do not include: constructional site costs, planning costs and financial costs.

In relation to the total building costs, material costs represents approximately 52% for all three models.

Terrain and ground conditions on the site as well as the amount of the farmers own labour input will affect the total costs. An evaluation indicates a higher cost reduction potential for O2R, especially with a stepwise development. With a short time development, the differences in cost reduction potential between the three models will be smaller.

From an evaluation of the possibilities of alternative use of the building, the enclosed building types, and especially L6R, seem to have definite advantages. This might affect the long-term economy of the farm, and should be taken into consideration, particularly on farms where fur animal production is combined with agriculture, forestry or other supplementary sources of income.

ITF-trykk 52/1992. Agricultural University of Norway, Department of Agricultural Engineering. In NORW. 35 pp, 4 tables, 9 figs., 18 refs.

Estimate of size using weight or grading in cage or trap

Ulla Lund Nielsen, Anette Svendsen

The sizes of 347 wild type male mink were estimated by respectively weighing in a grading trap, visual assessment in a grading trap and visual assessment in a cage. The sizes were thereafter compared to the actual measured pelt lengths.

An analysis of the correlation between kit size and pelt length revealed that the best results were obtained by weighing (corr. = 0.91). Both the visual estimates resulted in acceptable correlations to pelt length (0.69 and 0.73). Weighing can therefore be recommended to predict pelt length while visual estimates can be used if weighing is not possible.

In DANH. 2 tables, 1 fig., 2 refs. Technical Year Book 1993/94, pp 7-12, 1995.

Stretching of heated pelts on boards

Ulla Lund Nielsen

A total of 120 pelts were placed in one of 2 groups - an experimental group or a control group. Pelts from both groups were stretched on boards in the usual manner immediately following drumming. After drumming, the experimental pelts were rolled in a wrung towel and placed in an incubator at 60°C for 30 minutes. This resulted in the pelts having a temperature of approximately 35°C. Pelts from both groups were dried in the usual manner (3 days at 18°C and 52% R.H.).

As a result of the heat treatment, it was possible to stretch the pelts more. Thus, the heat-treated pelts were significantly longer than those in the control group after boarding. However, if the increase is calculated as a per cent of the length before boarding, there was no significant difference between the groups. This was also the case with the pelt measurements from March 29, 1993.

The conclusion is, therefore, that it is not worthwhile to heat the pelts. This is partly because the effect is minimal and partly because of extra costs due to increased energy consumption and the risk of heat damage to the pelts.

In DANH. 1 table. Technical Year Book 1993/94, pp 206-207, 1995.

Reference intervals for insulin concentrations and insulin: glucose ratios in the serum of ferrets

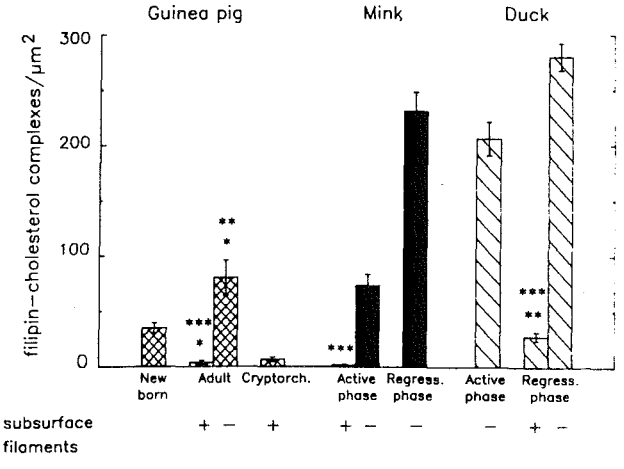
F.A. Mann, S.L. Stockham, M.B. Freeman, C. Wagner-Mann, C.L. Besch-Williford, R.F. Nachreiner

Serum samples from 443 adult ferrets (*Mustela putorius furo*) of both sexes from three different sources were assayed for glucose using a glucose oxidase dry slide method and immunoreactive insulin (IRI) using a commercial radioimmunoassay kit. Ferrets from a separate research project (n=10) had significantly higher IRI and insulin:glucose (I/G) ratio than ferrets housed in a research animal supply facility (n=30) ($P \leq 0.05$). The ferrets housed in the research animal supply facility were used to establish reference intervals for serum insulin concentration and I/G ratio. The resultant reference intervals were recorded in conventional units (glucose, 100-163 mg/dl; IRI, 4.6-43.3 μU/ml; IG ratio, 3.6-34.1 μU/mg) and Système International units (glucose, 5.6-9.0 mmol/L; IRI, 33-311 pmol/L; I/G ratio, 4.6-44.2 pmol/mmol). Further work with the commercial IRI assay kit used in this study is necessary to determine the diagnostic serum IRI concentrations and I/G ratios in ferrets with insulin-secreting pancreatic tumors.

Journal of Small Exotic Animal Medicine 2 82), 79-83, 1993. 2 tables, 3 figs., 25 refs. Authors' abstract.

Filipin vs Enzymatic Localization of Cholesterol in Guinea Pig, Mink, and Mallard Duck Testicular Cells

R.-Marc Pelletier, María Leiza Vitale



To test the validity of filipin cytochemistry for localization of cholesterol in testicular cells, we compared the results obtained by this technique with those obtained by a two-step enzymatic method involving cholesterol esterase and cholesterol oxidase. In all the animal models tested (guinea pig, mink, and mallard duck) the disappearance of subsurface filaments along Sertoli cell junctional membranes was accompanied by a significant increase in the number of filipin-cholesterol complexes/μm^2 in these membranes. Enzyme histochemistry allowed localization of free cholesterol in the limiting mem-

brane of multivesicular bodies, in membranes within lysosomes, in mitochondrial membranes, and in junctional membranes, with or without subsurface filaments. The method also permitted selective visualization of cholesterol esters in lipid droplets. We conclude that filipin mapping of cholesterol induces false-negative cytochemical results. The enzymatic method is superior to filipin because it allows localization of free cholesterol in junctional membranes and of cholesterol esters in lipid droplets. This compartmentalization of the compounds may represent the basis of a system that helps to maintain constant free cholesterol levels in the testis.

The Journal of Histochemistry and Cytochemistry, Vol. 42, No. 12, pp. 1539-1554, 1994. 1 table, 15 figs., 91 refs. Authors' summary.

Prolactin binding sites in the adrenal glands of mink (*Mustela vison*)

Jack Rose, Charles Wert

1. The purpose of this study was to determine if the mink adrenal gland might be a target organ for prolactin by establishing whether or not binding sites for the hormone exist in adrenal cell membranes.
2. Adrenal glands were collected from adult female mink in November 1991, homogenized and subjected to differential centrifugation into three particulate fractions: 1500, 15,000, and 50,000 g. All binding determinations were made using ^{125}I-oPRL and 200-300 μg protein from the 50,000 g particulate fraction. Optimal binding occurred within 8 hr at 25°C.
3. Scatchard analysis of saturation data revealed a single set of high affinity (K_d=9.27 x 10^{-11} ° 1.63 M), low capacity (B_{max}=34.22±5.37 fmol/mg) binding sites.
4. Binding sites appeared to be hormone specific as only oPRL (73% displacement) and oLH (8% displacement) inhibited binding of ^{125}I-oPRL to adrenal membranes. No inhibition of ^{125}I-oPRL binding to adrenal membranes occurred in the presence of a 500-fold excess of bTSH, oGH or oFSH.
5. Prolactin binding sites were readily detected in adrenal and kidney tissue, but were low in liver and almost non-detectable in spleen or lung tissue.
6. Our data suggest that the mink adrenal gland is a target organ for prolactin and that an interaction between the pituitary and adrenal glands may exist that is important for the regulation of such physiological processes as fur growth cycles.

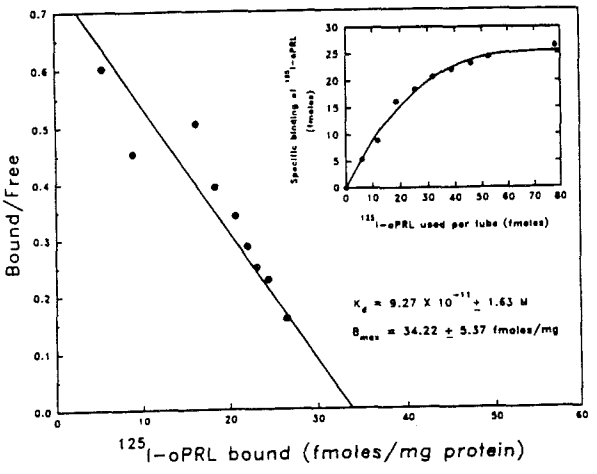


Fig. 2. Scatchard and saturation (inset) analysis of the specific binding of ^{125}I-oPRL to mink adrenal membranes. A constant quantity (200-300 μg) of microsomal protein was incubated in triplicate with increasing concentrations of ^{125}I-oPRL (6-78 fmol, 24-318 x 10^3 cpm) with or without a 500-fold excess of competing nonradioactive hormone for each concentration of labelled hormone. Each point represents the mean of three trials.

Comp. Biochem. Physiol., Vol. 104B, No. 4, pp. 759-763, 1993. 3 figs., 41 refs. Authors' abstract.

The role of prolactin in the reactivation of hair follicles in relation to moulting in cashmere goats

P. Dicks, A.J.F. Russel, G.A. Lincoln

The effects of the suppression or elevation of plasma prolactin concentrations in spring on the timing of the reactivation of the hair follicles and the timing of the spring moult were investigated in cashmere goats.

Thirty eight adult female goats, housed under conditions of natural photoperiod at 55°55'N from mid-December until May, were allocated to four groups starting on 5 January: ten served as untreated controls, eight received 2 mg ovine prolactin subcutaneously every 12 h for 7 weeks (PRL), twelve received 35 mg bromocriptine intramuscularly every 14 days for 17 weeks (BCR) and eight received injections of both ovine prolactin and bromocriptine at the above dose rates for 7 weeks (PRL+BCR).

In the PRL group there was an earlier reactivation of the secondary hair follicles (PRL vs control, proportion of secondary follicles in anagen, weeks 1-5, $P<0.01$) associated with an earlier moult of secondary fibres (cashmere) but no significant difference in the activity of the primary hair follicles. In the BCR group there was a delay in the reactivation of both the secondary and primary hair follicles (BCR vs control, proportion of secondary and primary hair follicles in anagen, weeks 5-13, $P<0.01$) and a delay in the moult. In the PRL+BCR group there was an early reactivation and moult similar to the PRL group. Voluntary food intake (VFI) and liveweight were also measured.

Only in the BCR group was there a decrease in VFI compared with the controls but with no effect on liveweight. It was concluded that the seasonal increase in prolactin secretion which normally occurs in spring is causally involved in the reactivation of primary and secondary hair follicles and moulting in cashmere goats.

Journal of Endocrinology 143, pp. 441-448, 1994. 1 table, 4 figs., 25 refs. Authors' abstract.

Effect of isoflurane on hematologic variables in ferrets

R.P. Marini, L.R. Jackson, M.I. Esteves, K.A. Andrutis, C.M. Goslant, J.G. Fox

Effects of isoflurane on the CBC in ferrets were studied. There was rapid decrease in all hematologic variables after induction of anesthesia. Percentage reductions in indices of the erythron (hematocrit, RBC count, hemoglobin concentration) exceeded those of plasma protein concentration and WBC count at the first postinduction time point. There was little additional decrease in these variables for the duration of anesthesia. The values had partially recovered to preanesthetic baseline at 45 minutes after anesthesia. Although these alterations appear to be well tolerated in healthy ferrets, care should be exercised when subjecting anemic, geriatric, or debilitated ferrets to isoflurane-induced anesthesia.

Am J Vet Res, Vol. 55, No. 10, pp. 1479-1483, 1994. 5 figs., 33 refs. Authors' summary.

Cloning of $p27^{Kip1}$, a cyclin-dependent kinase inhibitor and a potential mediator of extracellular antimitogenic signals

Kornelia Polyak, Mong-Hong Lee, Hediye Erdjument-Bromage, Andrew Koff, James M. Roberts, Paoul Tempst, Joan Massagué

We cloned $p27^{Kip1}$, a cyclin-dependent kinase inhibitor implicated in G1 phase arrest by TGFß and cell-cell contact.

$p27^{Kip1}$ associates with cyclin E-Cdk2 complexes in vivo and in vitro, prevents their activation, and inhibits previously activated complexes, and $p27^{Kip1}$ overexpression obstructs cell entry into S phase. $p27^{Kip1}$ potently inhibits Rb phosphorylation by cyclin E-Cdk2, cyclin A-Cdk2, and cyclin D2-Cdk4. $p27^{Kip1}$ is highly conserved and broadly expressed in human tissues, and its mRNA levels are similar in proliferating and quiescent cells. $p27^{Kip1}$ has a region of sequence similarity to $p21^{Cip1/WAF1}$, the Cdk inhibitor whose transcription is stimulated by p3. A $p27^{Kip1}$ peptide corresponding to this region retains Cdk inhibitory activity.

We suggest that cell contact, TGFß, and p53 all restrain cell proliferation through related Cdk inhibitors.

Cell, Vol. 78, pp. 59-66, 1994. 1 table, 5 figs., 39 refs. Authors' summary.

Centric-fusion translocation and whole-arm heterochromatin in the karyotype of the blue fox (*Alopex lagopus* L.): synaptonemal complex analysis

M. Switonski, I. Gustavsson

Conventional observations of mitotic chromosomes from two male blue foxes, revealing a centric-fusion translocation and whole-arm heterochromatin, were verified by synaptonemal complex analysis.

This analysis revealed that the centric fusion had been preceded by a conspicuous loss of chromosome material in the two one-armed chromosomes involved, but the chromosomal origin of the centric-fusion kinetochore could not be established. The nontranslocated chromosomes of the trivalent, which in all cells but one were in cis configuration, had reached by early pachytene a stage in which almost complete homologous pairing and nonhomologous association or pairing of the free ends of the chromosomes could be observed.

In later stages, complete pairing of the nontranslocated chromosomes with the corresponding arms of the centric-fusion translocation was seen occasionally.

One to six autosomal bivalents demonstrated unpaired heterochromatic arms in early pachytene, and the heterochromatic chromosome arms were sometimes unpaired even in late pachytene. Some of them showed a distinct size heteromorphism in late zygotene and early pachytene.

In most late-pachytene cells, however, the heteromorphic chromosomes were completely length-adjusted. Only a small fraction of the cells showed pairing interference between nonhomologous chromosomes.

Cytogenetics and Cell Genetics, Vol. 57, pp. 1-8, 1991. 2 tables, 4 figs., 43 refs. Authors' abstract.

A new mink mutation

I.B. Thikhomirov

The mutation, which resulted in mink with a coat similar to that of an otter, occurred at a fur farm in 1991. Guard hairs of the mutant animals have a light band, located as in silver foxes, but varying from almost white to light brown. The mutation was designated "Talitsa" after the place where it occurred.

Matings were carried out between 424 females and 84 males of the following groups: (1) dark brown males with dark brown females; (2) dark brown males with Talitsa females; (3) Talitsa males with dark brown females; (4) Talitsa males with Talitsa females.

For the 4 groups, respectively, pregnancy rate was 91.7, 91.3, 92.2, and 92.1%, and litter size 6.68,

6.08, 6.67, and 5.98. It was possible to differentiate Talitsa and dark brown kits from 15 to 20 days of age onward. Some Talitsa kits were born in mating groups 2-4. For 558, 620, and 545 progeny in these groups, the ratio of Talitsa to dark brown kits was 1.11:1, 0.97:1, and 3.16:1, respectively. White spotting occurred in some Talitsa mink. It was concluded that Talitsa is a dominant trait with approximately full penetrance.

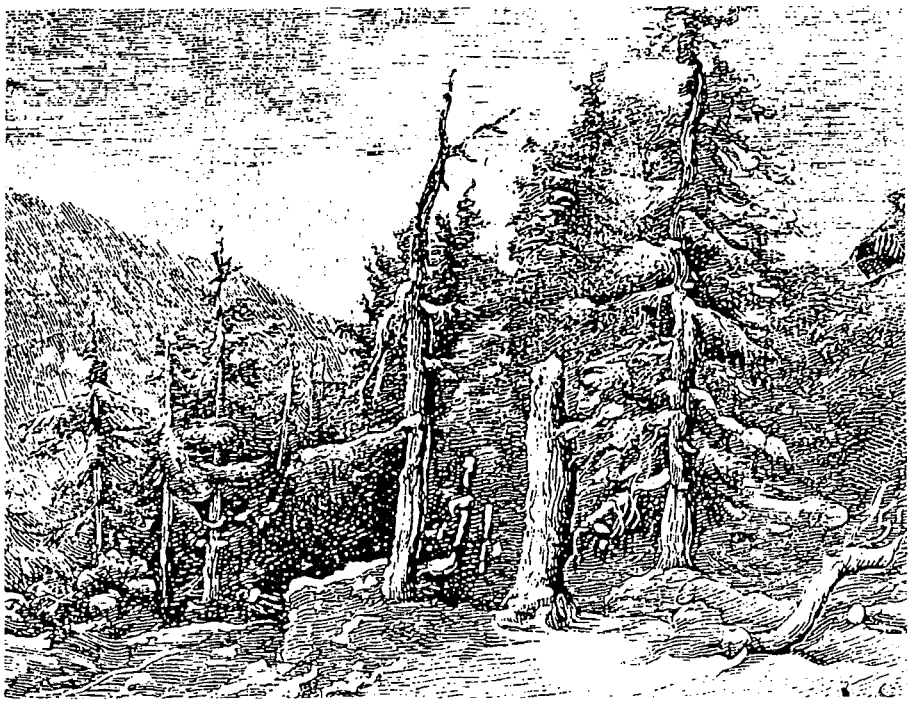
Krolikovodstvo i Zverovodstvo, No. 4, pp. 4 + 6, 1993. In RUSS. 2 tables. CAB-abstract.

Constitutional traits of coloured foxes

N.N. Shumilina, K.G. Dekanosidze, V.G. Lobanov

For 240 foxes of the golden platinum, platinum, red and silver varieties, data are tabulated on body weight, body measurements and body indices, and weights of the uterus, ovaries, liver, spleen, lungs, and heart. At slaughter for pelt production, body weight averaged 5.9, 5.6, 6.5, and 6.3 kg in the 4 colour varieties resp. in males, and 4.8, 4.9, 5.35, and 5.2 kg in females, uterus weight averaged 983, 1043, 1147, and 1240 g, and weight of ovaries 367, 359, 430, and 440 g.

Krolikovodstvo i Zverovodstvo, No. 5, pp. 8, 1991. In RUSS. 3 tables, CAB-abstract.



Original Report

The effect of Receptal (busereline) on some reproductory indices in female polar foxes (Part I)

Kazimierz Sciesinski, Beata Pieta

Animal Breeding Department, Warsaw Agricultural University - SGW, ul. Przejazd 4, 05-840 Brwinow, Poland

Summary

The study investigated the effect of Receptal on the reproduction effectiveness in polar foxes including per cent pregnant females and the mean litter size. Receptal belongs to the GnRH type hormones and its international name is busereline. The investigations were carried out on 70 female polar foxes. The administration of Receptal at a dose of 0.5 ml/animal (0.002 mg busereline) to female polar foxes resulted in an increase in per cent pregnant females and mean litter size. The female polar foxes which had received Receptal and had been fertilized in a natural way gave birth to healthy litters capable of further breeding.

Introduction

One of the most important factors determining economical results in animal production is reproduction. The profitability of breeding polar foxes, multifetal and monoestral animals, depends on the number of born and reared cubs. Foxes have a defined physiological ability for reproduction amounting to 1-23 young in polar and 1-13 young in common foxes. That number is reduced by various factors at particular stages of development from the time of ovulation until weaning. The factors are either hereditary (e.g. deformations of the reproductive organs, endocrine secretion disorders which can result in polycystic ovaries, disturbances in ovulation and the course of sexual cycle phases) or environmental (e.g. feeding errors, organic or functional changes in the reproductory organs caused by diseases or climatic conditions).

In order to limit the unfavourable effects of the above mentioned factors on the results of reproduction, apart from the optimization of management conditions, different compounds such as vitamins, microelements and hormones are also used. One of them, which improves the effects of animal reproduction, is the hormonal preparation Receptal. Its effectiveness has already been proved in investigations on cows, horses and rabbits.

The present work is a trial to evaluate the influence of Receptal on the reproductory effectiveness in polar foxes expressed, among other things, by the number of pregnant females and the mean litter size.

Polar foxes, like common foxes, mature sexually in the first year of life, i.e. at the age of about 10 months. Both species are monoestral, i.e. there is one sexual cycle a year with typical phases (*Wolinski, Slawon, 1964*). The preoestral period lasts 30-

40 days. In polar foxes it begins in the middle of January, while in common foxes it starts a month earlier. At that time the females become restless or depressed, often urinate and assume a characteristic posture with the tail drawn sidewards.

Before the proper oestrus begins, some changes are observed in the internal and external reproductive organs. Those changes appear 7-14 days prior to the proper oestrus and in common foxes 4-7 days.

Receptal (by Hoechst) is an injectable solution containing a hormone which is very active biologically and peptide-like structurally. It is a chemical analogue of gonadoliberien - a releasing hormone (RH), luteinizing hormone (LH) and folliculotropin, a follicle-stimulating hormone (FSH). Such a substance which can release LH and FSH is described by the following abbreviations: LH/FSH - RH, LH-RH and GnRH (gonadotropin releasing hormone). The international chemical name is busereline.

Contrary to natural GnRH consisting of 10 amino acids (decapeptid) the active substance in the Receptol preparation includes only 9 amino acids. The structural variation causes the difference in the effectiveness of the hormonal action between the synthetic and natural hormones. It consists in a better bending of monopeptides with proper hormonal receptors in the pituitary gland. Unlike decapeptides, monopeptides are also more resistant to the action of decomposing enzymes which results in the prolongation of the synthetic GnRH action as compared to the natural GnRH.

As a result, a higher hormonal activity of the substance contained in the preparation is connected with the increased secretion of the luteinizing hormone (LH) and folliculotropin (FSH) by the pituitary gland after the administration of the preparation. In turn, increased hormonal secretion increases hormonal activity of the target sexual glands.

Experiments performed on laboratory animals confirmed the higher biological action of the GnRH analogue as compared to the natural GnRH (gonadoliberine).

In rats, as a result of the administration of the preparation, the concentration of LH was 18 times higher and the concentration of FSH 15.7 times higher as compared to the concentration of those gonadotropins resulting from the action of the natural GnRH. At the same time the qualitative ratio FSH:LH does not differ from the physiological standard (*Sandow, 1979; Information about Receptal preparation by Hoechst*).

The aim of the investigations carried out on female polar foxes was the improvement of the reproduction indices by:

- the induction of ovulation and synchronizing ovulation with the time of fertilization in order to improve the effectiveness of mating.

- the induction of superovulation which allows a simultaneous fertilization of a greater number of the egg cells thus resulting in a larger litter.

Material and methods

The experiments were carried out on a farm of common and polar foxes of the Agricultural Co-operative Duchnice near Ozarow in the years 1988-1991. Observations were performed on female polar foxes aged 1 to 4 years. The first experiment included 40 females. During the oestrus period, twenty female polar foxes (blue and white) from that group were administered one intramuscular injection of 0.5 ml Receptal preparation (0.002 mg pure busereline/animal) in order to induce ovulation. The preparation used is a solution of the synthetic GnRH-LH (FSH) gonadotropin releasing hormone and 1 ml of the preparation contains 0.004 mg pure busereline which corresponds to the natural hormone releasing LH and FSH.

After Receptal administration the females were fertilized in a natural way. Twenty female polar foxes from the first experiment comprised the control group. The second experiment was also carried out on 40 female polar foxes which in the previous year did not have large litters. Twenty females from that group were administered Receptal in the dose as the above during the oestrus

period. The remaining 10 females comprised the control group. The females which were administered the preparation as well as the control ones were fertilized in a natural way.

The results of the investigations were analysed statistically considering the significance of differences between the means as to the number of cubs born in particular experimental groups as compared to the control ones. To compare the mean values those calculations were based on the C-Cohran and t-Student tests at the significance levels of L - 0.05 and L - 0.01.

The authors want to express their gratitude to Dr. Moller-Holtkamp, the representative of Hoechst for the donation of the preparation.

Results and discussion

Table 1 presents the results of the first experiment regarding the effect of Receptal on the number of cubs in polar fox litters. Table 2 presents the results of the effect of Receptal on per cent pregnant females and the mean litter sizes in polar foxes. The results of the investigations of the Receptal effect on the number of cubs in litters of the female polar foxes which had small litters in the previous season are presented in Table 3 and 4.

Table 1. The effect of Receptal (in the dose of 0.5 ml/animal - 0.002 mg busereline) on the number of cubs born in the litters of female polar foxes

No.	Female		No. of cubs born
	Control group		
1	P	103	6
2	Z	1402	10
3	C	1610	-
4	W	1680	-
5	W	516	11
6	W	1718	-
7	C	1618	8
8	A	722	9
9	A	282	12
10	P	12	-
11	A	1512	10
12	A	940	9
13	A	278	-
14	A	1306	5
15	A	622	14
16	A	1328	14
17	A	962	10
18	D	358	11
19	A	1220	5
20	Z	772	8
	Group of females which received the preparation		
1	A	88	-
2	W	482	-
3	W	776	14
7	A	442	13
5	A	962	10
6	A	1328	14
7	B	484	15
8	B	728	-
9	C	1112	-
10	P	105	-
11	B	2126	2
12	W	515	11
13	A	278	-
14	A	940	9
15	A	1060	8
16	A	1520	10
17	B	1522	12
18	D	8	-
19			
20			

Table 2 The effect of Receptal (in dose of 0.5 mg/animal, i.e. 0.002 mg busereline/animal) on the per cent of pregnant females and the mean litter size in polar foxes

Group	% pregnant females after fertilization	Mean number of cubs in a litter
1. Group with GnRH (20 female polar foxes)	60	11.3
2. Control group without GnRH (20 female polar foxes)	75	9.4

Table 3 The effect of Receptal (in the dose of 0.5 ml/animal, i.e. 0.002 mg busereline) on the number of cubs in the litters of those female polar foxes which produced small litters in the previous season

No.	Female	No. of cubs born in the previous season	No. of cubs born in the investigated season
Control group			
1	A 1232	4	-
2	A 136	4	-
3	A 820	3	2
4	A 900	4	-
5	U 24	4	3
6	Z 1516	4	4
7	Z 1488	4	-
8	A 1248	3	-
9	A 1216	4	2
10	U 30	3	4
Group of females which received Receptal			
1	U 820	5	-
2	U 1620	4	-
3	T 1104	3	11
7	Z 114	5	10
5	Z 1672	4	10
6	W 452	3	8
7	P 113	7	7
8	Z 304	1	15
9	Z 16	11	-
10	W 1004	8	-
11	A 742	10	11
12	A 830	9	10
13	A 902	6	8
14	A 904	10	-
15	U 810	4	16
16	W 902	4	6
17	W 1446	4	9
18	W 1618	4	10
19	W 1622	4	-
20	B 836	4	-

In experiment I fertilization and pregnancy were obtained in 12 females (comprising 60% of mated animals), the litters amounted to 1 to 15 cubs which were capable of further breeding, i.e. 11.3 cubs on average (see Table 1 and 2).

In the control group, which did not receive Receptal, fertilization and pregnancy were obtained in 15 female polar foxes (thus comprising 75% of mated animals) and the litters amounted to 1 to 14 cubs capable of further breeding, i.e. 9.4 animals on average (see Table 1 and 2).

Significant differences were observed in the mean number of cubs in the litters of females which received Receptal.

In experiment II, performed on female polar foxes which received the preparation, fertilization and pregnancy were obtained in 15 females (comprising 75% of mated animals), the litters amounted to 1 to 16 cubs which were cable of further breeding, i.e. 10 cubs on average (see Table 3 and 4).

In the control group of female polar foxes fertilization and pregnancy were obtained in 5 females (thus comprising 50% of mated animals) and the litters amounted to 2-4 cubs capable of further breeding, i.e. 5 animals on average (see Table 3 and 4).

Highly significant differences were observed in the mean number of cubs in the litters of females which received Receptal.

In experiment I, as results from the data presented in Table 1 and 2, after the administration of Receptal, there was a significant increase of the mean litter size in female polar foxes. However, the per cent of pregnant female polar foxes did not increase.

In experiment II (Table 3 and 4), the mean litter size and per cent pregnant females increased after the administration of Receptal.

While comparing the results of the investigations of per cent pregnant females, one can notice that they are not explicit since in experiment I no improvement was observed.

The investigations were carried out on a productive farm and included a relatively small number of animals. Thus, it would be advisable to repeat the experiments on a larger number of foxes.

The investigations performed on rabbits showed an increase in per cent pregnant females and a slight increase in litter size (acc. to Information by Hoechst).

In mares, after the administration of Receptal, one could observe that the per cent of pregnancies nearly doubled as compared to the control group (Information by Hoechst, *Beaupoil, 1979*).

Table 4 The effect of Receptal (in the dose of 0.5 ml/animal, i.e. 0.002 mg busereline/animal) on per cent pregnant females and the mean litter size in polar foxes

Group	% pregnant females after fertilization	Mean number of cubs per litter
1. Group with GnRH (20 female polar foxes)	75	10
2. Control group without GnRH (10 female polar foxes)	50	5

In cows, after the Receptal administration, the per cent of pregnancies increased from 9.2 to 13.1 (*Leidl, Bostedt, 1979*).

Two of the basic indicators of reproduction, namely per cent pregnant females and the number of cubs per litter, were evaluated in the present investigations. The main effect of the administration of synthetic hormone GnRH, consisting of gonadotropin release was appraised only indirectly. It was impossible to evaluate the gonadotropin level using biochemical methods. It is indicated to continue the investigations of the use of compounds improving the reproduction indices on the fox farms since it is important for the profitability of production.

Conclusions

1. In polar foxes per cent pregnant females and mean litter size increase after the administration of Receptal at a dose of 0.5 ml/animal, i.e. 0.002 mg busereline.

2. Female polar foxes which had received Receptal and were fertilized in a natural way give birth to healthy progeny capable of further breeding.

3. Considering the obtained results it would be advisable to continue the investigations on a larger number of female polar foxes.

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Original Report

The use of Receptal (busereline) in the course of reproduction in female silver foxes (Part 2)

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Summary

The investigation was carried out on 55 female silver foxes which were administered Receptal. After the administration of Receptal in female silver foxes the per cent of pregnant females and the mean litter size increased. Receptal causes the appearance of oestrus in female silver foxes.

Introduction

The aim of the present investigation was to determine in what way Receptal (busereline) affects the percentage of pregnant females, the mean litter size and the occurrence of heat in female silver foxes.

Receptal has been used to induce ovulation and improve the percentage of pregnant cows, mares and female rabbits and also in cases of reproductory disturbances in cows and mares. As in laboratory animals, the investigations performed in cows revealed a higher hormonal activity of busereline than the natural GnRH.

In cows the highest concentration of gonadotropins in the blood was observed 135 min. after administration of that busereline.

As a result of gonadotropin secretion following the Receptal administration one can observe the maturation of ovarian follicles, ovulation and the development of yellow bodies, i.e. there occurs an oestrus cycle with a physiological course.

The effectiveness of Receptal was investigated in cows with reproductive disturbances such as ovarian follicular cysts, inhibition of the oestrus cycle, delayed ovulation and obliteration of ovarian follicles.

After busereline administration in cows with ovarian follicular cysts there is a quick
rise of FSH and LH levels in the plasma which reach their highest value within 1 to 2 hours. It leads to the growth and maturation of ovarian follicles and ovulation.

The administration of Receptal at the time of insemination or mating or 6 hours before mating is effective in the treatment of delayed ovulation and ovarian follicle obliteration. Ovulation occurs within 24 h after the drug administration.

Receptal was also used in order to improve the percentage of conceptions in cows. The administration of the synthetic hormone GnRH

(gonadotropin releasing) during oestrus allows the synchronization of ovulation with the moment of copulation. Ovulation occurs within 24 h of administration of the preparation (*Humke, R., Moller-Holtkamp, 1982*). The results of all the investigations explicitly point to the fact that the administration of Receptal causes a significant improvement of the reproductory indices in cows. The best effects are obtained in cases of hormonal disturbances in the period preceding the appearance of oestrus (Information by Hoechst).

In horses, as in other species, hormonal activity of busereline is higher as compared to the natural GnRH. The investigations performed in mares confirmed the action of Receptal in the stimulation of ovulation. That fact has been used in order to improve the per cent of conceptions in mares and in cases of reproductory disturbances such as ovarian follicular cysts and oestrus cycle inhibition.

In the experiments on rabbits the busereline action inducing ovulation was applied in order to improve the effectiveness of fertilization. Artificial stimulation of ovulation is a necessary condition for the success of insemination of female rabbits (Information about the drug by Hoechst).

Inducing ovulation by the GnRH contained in Receptal is possible only with high estrogen levels. Physiologically, the highest levels occur the first days after parturition and after weaning. Receptal can be administered already 24 h after parturition. The ovulation-stimulating action of Receptal can also be used at coitus in female rabbits which as such causes ovulation.

The results from the experiments show that the per cent pregnant female rabbits clearly increases after Receptal administration (Information about Receptal by Hoechst). The aim of the present investigations was to increase the percentage of pregnant females and the litter size as well as the stimulation of oestrus in silver foxes.

Material and methods

The experiments were performed on a farm of polar foxes of the Agricultural Cooperative Duchnice near Ozarow in the years 1988-1992. The observations were carried out from January to May every year.

The first experiment included female silver foxes which had produced small litters in the previous seasons.

The experiments were performed on 20 females out of which 10 were the control and 10 the experimental group. The females from the experimental group received an intramuscular injection of 0.5 ml Receptal (0.002 mg busereline). Receptal is a solution of synthetic GnRH - LH (FSH) (gonadotropin releasing hormone).

After Receptal administration and during the oestrus cycle the females were naturally fertilized.

The second experiment was performed on female silver foxes aged about one year in which ovulation did not occur at the anticipated time (January - March).

Twenty-five females received injections of 0.5 ml Receptal in order to induce oestrus. Ten female silver foxes comprised the control group.

The results of the investigations were analysed statistically to test the significance of differences between the means of the number of cubs born in particular experimental groups as compared to the control ones. To compare the mean values the calculations were based on the C-Cohran and t-Student tests at the significance levels of L - 0.05 and L - 0.01.

The preparation was delivered by a representative of Hoechst, Dr. Moller-Holtkamp, to whom we want to express our gratitude.

Results and discussion

Table 1 presents the data obtained in the investigation on the effect of Receptal at a dose of 0.5 ml/animal (0.002 mg busereline) to female silver foxes which in the previous year had produced small litters.

Table 2 presents the results of the investigations of the effect of Receptal on the per cent of pregnant females and the mean litter size in silver foxes.

Table 1 The effect of Receptal on the number of cubs in the litters of female silver foxes producing small litters

No.	Female farm number		No. of cubs born in the previous season	Number of cubs born	
				Without Receptal	After Receptal administration
Control group					
1	A	186	4	3	
2	P	146	4	4	
3	A	24	4	4	
4	A	398	4	4	
5	A	166	4	3	
6	B	4	3	-	
7	B	252	4	5	
8	B	426	4	4	
9	B	436	3	3	
10	T	22	3	6	
Experimental group					
1	Z	126	2		7
2	Z	130	1		7
3	P	248	3		6
7	Z	232	4		6
5	T	30	4		4
6	Z	206	4		6
7	Z	92	4		7
8	A	192	3		6
9	B	78	2		8
10	B	184	4		6

In experiment I in the group of female silver foxes fertilization and pregnancy were obtained in 10 females (comprising 100% of the mated animals), and the litters amounted to 1 to 8 cubs which were capable of further breeding, i.e. 6.3 cubs on average (see Table 1 and 2).

In the control group fertilization and pregnancy were obtained in 9 female silver foxes (thus comprising 90% of mated animals) and the litters amounted to 1 to 6 cubs capable of further breeding, i.e. 4 animals on average (see Table 1 and 2).

Highly significant differences were observed in the mean number of cubs in the group of females which received Receptal.

Per cent pregnancies and mean litter size increased in female silver foxes after the administration of Receptal. The investigations carried out on rabbits showed an increase in per cent pregnant females and a slight increase in litter size (Information about Receptal by Hoechst). As shown in data in the literature the mean percentage of conceptions in cows also increases (*Leidl, Bosted, 1979*).

Table 3 presents the results of the investigations of the effects of Receptal on the appearance of oestrus in female silver foxes.

In the second experiment the signs of oestrus appeared in 6 animals (24% females; Table 3).

In that experiment (Table 3) Receptal was administered to female silver foxes at the end of the reproductory cycle.
Prior to Receptal administration no signs of oestrus were observed.

After a single administration of Receptal at a dose of 0.5 ml/animal, oestrus was observed in 24% females.

Table 2 The effect of Receptal (0.5 mg/animal, i.e. 0.002 mg busereline/animal) on per cent pregnant females and mean litter size in silver foxes

Group		% pregnant females after fertilization	Mean number of cubs per litter
1.	Group with GnRH (10 female silver foxes)	100	6.3
2.	Control group without GnRH (10 female silver foxes)	90	4.0

Table 3 The effect of Receptal (0.5 ml/animal, i.e. 0.002 mg busereline/animal) on the appearance of oestrus in female silver foxes

Group	No. of females	No. of females in which oestrus did not appear within 11 days after the administration of Receptal
Experimental group	25	6
Control group	10	-

Because of the time of the drug administration (the end of the reproductory cycle) some of the females could already be past the oestrus period which had not been noticed because of the lack of its external symptoms (quiet oestrus). The farm does not use any other methods of oestrus detection. It is impossible to induce oestrus in those animals which had already had it. The lack of oestrus in the experimental females could also result from the diseases of the reproductive organs and in such cases the action of the preparation is not effective.

In the above experiments the final clinical effects (number of cubs per litter, per cent pregnant females and appearance or lack of oestrus) were estimated and the main effect of the administration of the synthetic hormone (GnRH) consisting in the release of gonadotropins was evaluated only indirectly. It was not possible to assay the gonadotropin level using biochemical methods.

The investigations of the effects of Receptal on the appearance of oestrus should be treated as introductory. They were performed on a productive and not an experimental farm. Thus, it is advisable to repeat the above experiments on a larger number of females. The experiments with Receptal to induce oestrus were also performed on mares and cows. In mares, after the drug administration, oestrus appeared in 1-10 days in 32% of the mares, 10-20 days in 25.8% and in over 20 days in 3.7% of the mares.
All together oestrus appeared in 61.5% of the mares (acc. to Information about the drug by Hoechst).

In cows, after the administration of Receptal, oestrus appeared in 12 days in 33.3% of the cows, in 13-24 days in 34.0%, in 25-36 days in 15.4%, in 37-48 days in 8.3% and in over 48 days in 7.1% of the cows. All together the oestrus appeared in 98% of the cows (*Humke, R., Zuber, H., 1977*).

It is advisable to continue the investigations of the use of compounds to improve the reproductory index on productive fox farms as it is of primary importance for the profitability of production.

It would also be advisable to use Receptal to synchronize oestrus and ovulation for artificial insemination in polar and silver foxes.

Conclusions

1. The per cent pregnant females and litter size increased in silver foxes after the use of Receptal at a dose of 0.5 ml/female, i.e. 0.002 mg busereline.
2. Receptal causes the appearance of oestrus in female silver foxes.
3. Receptal can be used for induction and synchronization of ovulation in female silver foxes because after its administration one can observe the improvement of the reproductory index.

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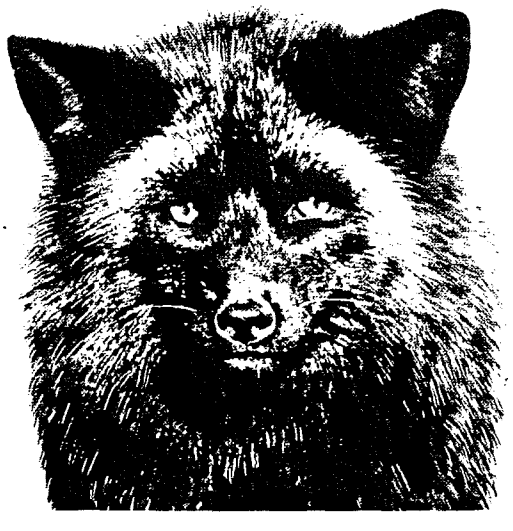

Ovarian follicular development in mink (*Mustela vison*)

D.A. Douglas, R.A. Pierson, B.D. Murphy

Ovarian follicular dynamics were studied during the breeding season, before and after ovulation in mink. Nulliparous female mink were stimulated to ovulate with an injection of 4 μm GnRH. Ovaries from three animals were collected on days 0, 2, 3, 4, 5, 6 and 7 after hormone treatment. A second dose of GnRH was administered on day 8 and ovaries were collected from three animals on day 9. Corpora lutea and follicles were identified in histological sections and follicles were classified by stage of development, healthy versus atretic, and by diameter. Preovulatory follicles (diameter 0.7-1.0 mm) were present in the ovaries of all animals on day 0 and these responded to GnRH treatment by ovulating. A synchronized wave of follicular development occurred following ovulation. Changes in follicle populations indicated that follicles are recruited from the small antral follicle class (0.2-0.4 mm) into the 0.4-0.6 mm class, with the first defined changes occurring between days 2 and 4. From the recruited group, a smaller cohort of follicles is selected to become the dominant follicles between days 4 and 6, and these acquire the ability to respond to a stimulus which induces ovulation at diameters of >0.7 mm. The ovaries of unmated mink also contained substantial numbers of large, degenerating, luteinized, unruptured follicles. These degenerating, luteinized follicles are considered to represent the demise of large follicles that failed to receive an ovulatory stimulus.

Journal of Reproduction and Fertility, 11, pp. 583-590, 1994. 1 table, 5 figs., 9 refs. Authors' summary.

Roles of melatonin and prolactin in testicular crudescense in mink (*Mustela vison*)

C.B. DiGregorio, A. Conzález Reyna, B.D. Murphy

Peripubertal male mink (*Mustela vison*) were treated with prolactin, melatonin or antibodies against melatonin to determine the effects of altered circulating concentrations of prolactin and melatonin throughout one season of testicular development. Treatment began on 1 October and continued until 4 March. Administration of 0.5 g ovine prolactin day^{-1} by minipump increased the circulating concentration of prolactin for the duration of the study and increased serum concentrations of LH. This treatment had no effect on the testosterone concentration or on testis size. Neither chronic treatment with melatonin throughout the period of crudescence nor passive immunization against melatonin for 79 days affected the circulating concentrations of prolactin, LH, testosterone or testis size. These results show clearly that, unlike in other seasonally breeding species, prolactin does not play a significant role in testis growth in mink. Administration of melatonin to male mink in October did not affect testis growth, presumably because the melatonin signal that cues photoperiodic events had already been received. Administration of antibodies against melatonin did not affect any of the features measured, suggesting that melatonin may have neural but not peripheral effects. Further support for this view can be found in the absence of an influence of melatonin on testis growth or on the plasma concentration of testosterone.

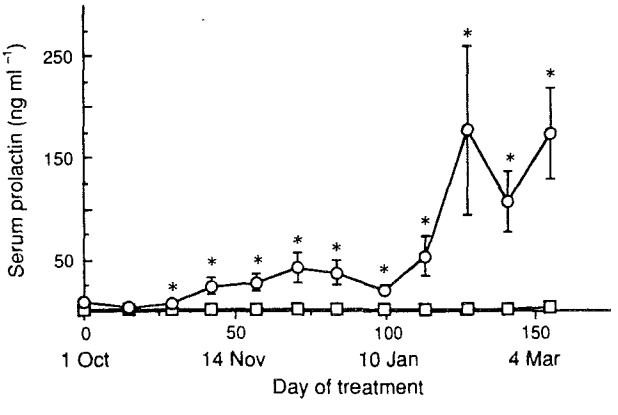

Fig. 2. Mean (±SEM) serum concentrations of prolactin between 1 October and 4 March in male mink that were either untreated (□) or infused with 0.5 mg ovine prolactin (○) day^{-1} for 110 days beginning on day 32 (1 November). Asterisks indicate significant differences between means on the day the sample was taken ($P<0.05$).

Journal of Reproduction and Fertility, 102, pp.1-5, 1994. 1 table, 3 figs., 32 refs. Authors' summary.

Original Report

Histopathological and histochemical studies of the internal organs of polar fox (*Alopex lagopus*) fed a diet supplemented with powdered fat "ERAFET"

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Summary

The objective of the study was to determine the effects of a fat concentrate "ERAFET" on the pathomorphology of liver, kidneys, pancreas, heart muscle, spleen, stomach and small intestines, and to observe lipid fat, and activity of succinate dehydrogenase and acid phosphatase in liver cells.

Histopathological and histochemical studies were performed on 30 randomly selected polar foxes, divided into three equal groups. Group I was fed standard feed mixture, group II received a standard diet supplemented with 4% and group III with an 8% powdered fat "ERAFET".

The animals on the standard diet were characterized by many degenerative changes and disturbances in blood circulation in the liver, kidneys and heart muscle.

Histochemical examinations revealed an increase of acid phosphatase activity and a decrease of succinate dehydrogenase activity, as well as the presence of many vacuoles with lipids in the liver cells. A 4% fat supplement decreased the number of damaged liver and kidney cells.

A diet containing an 8% fat supplement did not cause any morphologic changes in the internal organs and re-established physiologic activity of SDH and acid phosphatase.

Introduction

Use of fat supplements to increase the energetic value of fox diets is justified from a biological as well as an economic point of view. Carnivorous animals are evolutionarily adapted to consume fats originating from birds and mammals, viz. the animals which constitute their basic diet in the natural conditions. They contain a variety of chemical substances, some of which - such as e.g. unsaturated fatty acids: arachidic, linoleic and linolenic - must be supplied with the food as they are not synthesized in the animal organism. Fat content in the diet is important for a variety of reasons: it constitutes the source of condensed meta-

bolic energy 81 g = 38.9 KJ), determines assimilation of vitamins A, D, E, and K, and plays a protective function towards proteins. Fats containing multiunsaturated fatty acids increase animal requirements for vit. E and selenium. Their deficiency may lead to degenerative changes in the internal organs, especially in the liver (*Rouvinen and Kiiskinen, 1989; Rouvinen, 1991*). Ender and Helgebostad (1975) observed that vit. E and selenium deficiency in mink resulted in growth disturbances, anaemia, hair depigmentation, yellow fat, muscle degeneration and frequent mortalities. According to Lorek et al. (1993, 1994), Bieguszewski et al. (1986), Tauson et al. (1991) and Kinsella (1987) increased percentage of fat in the diet seems to be justified from a scientific point of view. The type of supplementary fat to be used is still the subject of studies. Rouvinen et al. (1989) showed that fish and rapeseed oils were quite suitable in feeding fox and mink. The objective of the study was to determine the effect of the commercial fat concentrate "ERAFET" on the pathomorphology of selected internal organs and the activity of some enzymes in polar fox liver.

Material and methods

The commercial dietary fat "ERAFET" (produced by "Kencpasz Ltd.") is in the form of a cream to light yellow powder stabilized without antioxidants or food preservatives. The concentrate contains 80% crude fat including 0.5% decanoic acid, 2.2% myristic acid, 0.9% hiragonic, 12.2% palmitic, 1.5% linoleic, 27.65 oleic, 26.2% stearic, and 0.9% arachidonic acid, and its energetic value is 30 MJ/kg.

Studies were carried out on 120 randomly selected polar foxes. The animals were divided into 3 groups having the same number of males and females. Since weaning until slaughter the animals were given diets supplemented with dietary fat "ERAFET". Group I received the standard feed mixture, group II the mixture with 4% fat supplement, and group III with 8% fat supplement. At the end of the experiment the animals were killed and dissected. Samples for histopathological and histochemical examinations were collected from 10 randomly selected animals in each group.

Table 1 Macroscopic changes observed in 120 dissected foxes

Organs	Macroscopic changes	Number of animals with damaged organs		
		Group I	Group II	GroupIII
Liver	parenchymatous degeneration	40	35	15
	congestion/ blood stasis	40	38	20
Kidneys	parenchymatous degeneration	35	20	15
	congestion	37	20	20
Heart muscle	parenchymatous degeneration	17	8	8
	congestion	19	10	9
Stomach	gastritis	32	24	20
Duodenum	duodenitis	40	26	20
Jejunum	jejunitis	2	4	6

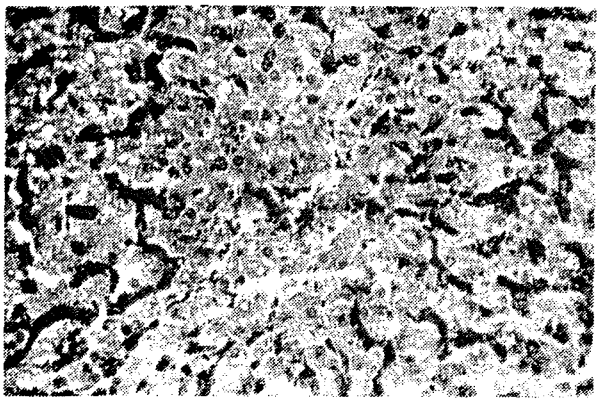

Fig. 1. Microscopic picture of fox liver in group I, parenchymatous and vacuolar degeneration is noticeable as well as blood stasis in the interlobule vessels. HE staining, magn. 240 x.

Histopathological studies were performed on the segments of liver, kidneys, spleen, heart muscle, stomach, duodenum, jejunum and pancreas. The segments were fixed in 10% neutralized formalin, immersed in paraffin blocks, cut into microtome scaps and stained with haematoxylin and eosine (HE) and PAS, according to the method of McManus.

Sections of liver were collected for histochemical studies. They were frozen in liquid nitrogen. Neutral lipids were determined in the caryostatic scarps according to the method of Lillie-Ahsburn, with the use of Sudan IV, activity of succinic acid dehydrogenase (SDH) with the method of Nachlas, and activity of acid phosphatase with the precipitation method of Gomori.

Results and discussion

Dissection studies revealed that a 4% supplement of the dietary fat "ERAFET" limited morphologic damages in the liver, kidneys, heart muscle, stomach and duodenum of the animals, and an 8% addition of the dietary fat was even more satisfactory because the number of animals with morphologic damages in the internal organs was much lower, and so was the extent of damages. The relevant results are presented in Table 1.

Histopathological examination of the liver revealed parenchymatous and vacuolar degeneration (Fig. 1), haemostasis, swelling of the stellate cells and glycogen disappearance, most pronounced in foxes from group I, less so in groups II and III.

Histochemical analyses revealed increased activity of acid phosphatase in the liver of the animals in group I (Fig. 2) and decreased activity of succinate dehydrogenase. Activity of the two enzymes was less disturbed in foxes from group II, while in group II it was physiologic (Fig. 3).

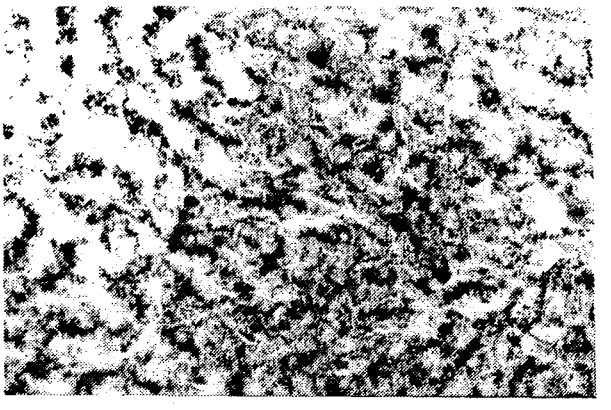

Fig. 2. Increase of acid phosphatase activity as reflected by granular and liquefied reaction in liver cells of a fox from group I. Precipitation reaction with the method of Gomori. Magn. 240 x.

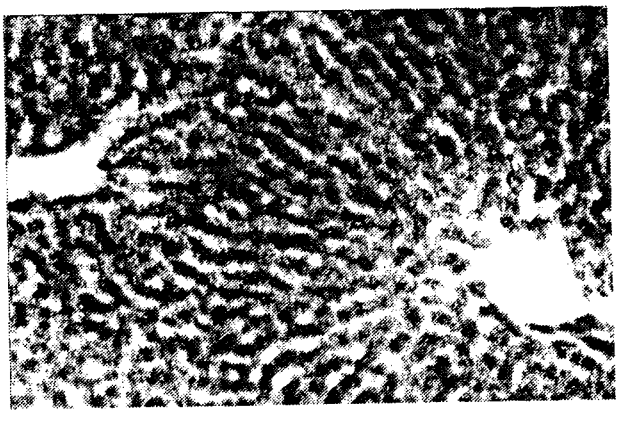

Fig. 3. High activity of succinic acid dehydrogenase in liver cells of a fox from group III. Reaction according to Nachlas. Magn. 240 x.

The results of histopathological and histochemical studies on fox liver are presented in Table 2. Strong parenchymatous degeneration of the epithelial cells of kidney tubules was observed in foxes from group I, together with PAS positive hyaline cast in the tubules and necrosis of many epithelial cells of the collecting tubules. Basal membranes in

the glomerules were thick. In the animals from group II these changes were less intensive while in group III no necrosis of the tubule epithelial cells was observed and there were no hyaline cast in the tubules (Table 3).

Parenchymatous degeneration of heart muscle fibres was observed in foxes from group I. Congestion, enlarged splenic follicles and numerous haemosiderine granules were observed in the spleen.

Histopathological examination of pancreas revealed parenchyma congestion. Extrasecretive pancreatic cells were enlarged and contained numerous acidophilic granules. Small foci of colliquative necrosis in pancreatic cells were observed only in two foxes (Table 4). Symptoms of acute mucitis were observed in the stomach and duodenum. They were less pronounced in the jejunum than elsewhere. Changes observed in the digestive tract are presented in Table 5.

Table 2 List of histopathological and histochemical changes in the liver of foxes from groups I, II, and III

Animal group	Histopathological changes in liver						Presence of glycogen in liver cells	Ressults of histochemical examination			
	Parenchymatous degeneration	Vacuolic degeneration	Focal cell necrosis	Congestion	Blood stasis	Swelling of stellate cells		Lipids in liver cells	Activity of acid phosphatase in the cells of hepar lobules		SDH activity in liver cells	
									in lobule centre	on lobule circumference	in lobule centre	on lobule circumference
I	+++ (8) + (2)	+++ (2) ++ (6) + (2)	+ (2) - (8)	++ (4) + (4)	++(2)	++ (4) + (6)	+ (7) +- (3)	+(10)	++++ (4) +++ (6)	++++ (8) +++ (2)	++(6) +(4)	+++(4) ++(6)
II	+++ (6) ++ (2) + (2)	+++ (2) + (2) -+ (4) - (2)	- (8) + (2)	++ (2) + (4) - (2)	++ (2)	+ (4) - (6)	++ (4) + (4) +- (2)	+(10)	+++ (4) ++ (4) + (2)	+++ (4) ++ (2) + (4)	++++(2) +++(6) + (2)	++++(4) +++(4) ++ (2)
III	+ (6) - (4)	+ (6) - (4)	- (10)	+ (4) - (2)	++(4)	+ (2) - (8)	++ (6) + (4)	+(10)	++ (6) + (4)	+ (10)	+++(8) ++++(2)	++++(6) +++(4)

Explanations to Table 2: Parenchymatous and vacuolar degeneration: (-) not noted; (+) noted in 30-40% of cells; (++) noted in 40-70% of cells; (+++) noted in 70-100% of cells. Congestion: (-) not noted; (+) comprises central veins and some interlobular veins. Stasis: (++) comprises intra and interlobular vessels to a large degree. Swelling of stellate cells: 8-) not noted; (+) noted in some cells. Presence of glycogen grains: (-) none; (+-) trace amounts; (+) single grains in a cell; (++) numerous grains. Lipids: (+) single cells with lipid vacuoles. activity of acid phosphatase: (+) physiologic; (++) considerable; (+++) high; (++++) very high. SDH activity: (+) trace; (++) decreased; (+++) physiologic, (++++) increased. Number of animals given in brackets.

Table 3 List of histopathological changes in fox kidneys and heart

Animal group	Histopathological changes in kidneys					Histopathoological changes in heart		
	Conges-tion	Parenchymatous degeneration	Hyalin degeneration	Necrosis of tubule epithelial cells	Thickening of basal membranes in glomerules	Parenchymatous degeneration	Congestion	Presence of glycogen grains
I	+(10)	++(10)	+ (10)	+ (8) - (2)	+ (10)	+ (10)	+ (8) - (2)	+ (4) +- (6)
II	+(6) -(4)	+(10)	+ (6) - (4)	+ (4) - (6)	+ (6) - (4)	+ (2) - (8)	+ (2) - (8)	+ (8) - (2)
III	+(4) -(6)	+(10)	+ (4) - (6)	- (10)	+ (8) - (2)	+ (2) - (8)	- (8) - (2)	+ (8) +- (2)

Parenchymatous degeneration: (-) not noted; (+) noted in 30-40% of cells; (++) noted in 40-70%. Congestion: (-) not noted; (+) noted. Presence of glycogen grains: (-) none; (+-) trace amounts; (+) single grains in a cell. Necrosis of tubule epithelial cells: (-) not noted; (+) noted: Thickening of basal membranes in glomerules: (-) not noted; (+) noted. Number of animals given in brackets.

Table 4 List of histopathological changes in spleen and pancreas

Animal group	Histopathological changes in spleen				Histoppathological changes in pancreas		
	Congestion	Enlargement of spleen nodules	Haemosiderosis	Presence of PAS - positive substances in follicles	Congestion	Increased acidophily of extrasecretive cells	Colliquative necrosis of extrasecretive cells
I	++ (2) + (2) - (6)	+ (8) - (2)	+ (2) - (8)	++ (6) + (4)	+ (8) - (2)	+ (2) - (8)	+ (2) - (8)
II	+ (4) - (6)	+ (4) - (6)	- (10)	++ (2) + (8)	+ (4) - (6)	+ (6) - (4)	- (10)
III	+ (10)	+ (10)	- (10)	+ (10)	+ (10)	+ (4) - (6)	- (10)

Congestion: (-) not noted; (+) noted. Presence of PAS positive substances: (+) single grains; (++) numerous. Haemosiderosis: (-) not noted; (+) noted. Number of animals given in brackets.

Table 5 List of histopathological changes in digestive tract

Animal group	Histopathologiical changes in stomach				Histopathological changes in duodenum				Histopathological changes in jejunum			
	Congestion of mucous membrane	Cell infiltration	Epithelium peeling	Multiplication of connective tissue	Congestion of mucous membrane	Cell infiltration in villi	Epithelium peeling	Villi deformation	Congestion of mucous membrane	Cell infiltrations in villi	Epithelium peeling	Villi deformation
I	+ (8) - (2)	+ (4) - (6)	++ (2) + (8)	++(2) + (2) - (6)	+ (8) - (2)	++ (2) + (4) - (4)	++(4) + (6)	++(6) + (4)	-(10)	+ (2) - (8)	++(10)	+ (6) - (4)
II	+ (6) - (4)	+ (4) - (6)	++ (2) + (8)	-(10)	++(4) +(6)	+ (4) - (6)	++(4) +(6)	++(2) +(6) -(2)	+(2) -(8)	- (10)	+ (10)	+ (4) - (6)
III	++(2) + (4) - (4)	+ (2) - (8)	++ (2) + (8)	-(10)	++(6) +(4)	+ (6) - (4)	++(6) +(4)	+(4) -(6)	+(4) -(6)	+ (4) - (6)	+ (10)	- (10)

Explanations: (-) no changes noted. (+) morphologic change (epithelium peeling in physiologic amounts, physiologic amount of mucus). (+++) intensified morphologic change. Number of animals given in brackets.

No significant differences were noted between the animal groups as to lipid content in the hepatic cells. Hepatic cells in foxes from group I contained single vacuoles with lipids near the veins of the central lobules. In groups II and III only single hepatic cells possessed vacuoles containing lipids.

Histopathological and histochemical studies of the internal organs of polar foxes proved that animal feeding with the standard diet was less satisfactory than feeding with the diet supplemented with "ERAFET" fat. Coagulative necrosis, vacuolic and parenchymatous degeneration were observed in the liver in the group of animals fed the standard diet, accompanied by congestions or blood stasis and changes in the activities of cell enzymes.

Activity of acid phosphatase increased in liver cells, while there was a decrease in the activity of succinic acid dehydrogenase, viz. of one of the most important enzymes for cell respiration. The kidneys, heart muscle, and digestive tract were less damaged. Although similar morphologic changes were observed in the group of animals receiving a 4% fat supplement, their intensity was much lower and comprised less liver cells than in the control group.

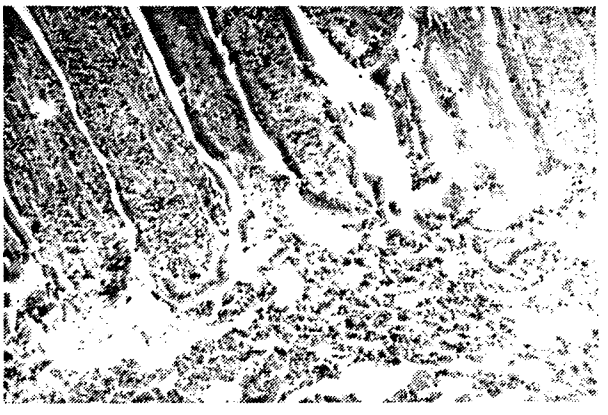

Fig. 4. Peeling of numerous epithelial cells, deformation of the vili and abundant mucus in the duodenum of fox from group II. HE staining, magn. 240 x.

Internal organs showed proper condition in group III which received an 8% "ERAFET" fat supplement. It should be noted that the diets affected less the digestive tract than the other organs. In the physiological conditions the mucous membrane of stomach and intestines is covered with a layer of mucus which plays an important protective role. Its amount may increase when the mucous membrane is irritated, inflamed etc. The epithelium of the stomach and intestine glands is renewed in the course of the peeling process, which is most intensive in the duodenum. Cell peeling or exfoliation reflects the processes of the adaptation to feeding conditions. Larger amounts of mucus, more intensive peeling of the gland epithelial cells, deformations of duodenum and jejunum villi and lymphoidal cell infiltrations were observed in the digestive tract of the animals from group I and in some foxes from group II; these changes suggest intensified adaptation processes and acute mucitis (Fig. 4).

Morphologic kidney damage is fairly frequent in foxes. It is induced by a number of toxic and pathogenic factors and is often manifested as damage to the kidney glomerules and tubule epithelium.

Examinations of the kidneys in the experimental animals revealed thickening of the basal membranes in kidney glomerules, symptomatic of an inflammatory process, as also parenchymatous degeneration of the kidney tubule epithelium and presence of hyalin protein deposits. This suggests increased excretion of proteins with the primary urine and its inhibited resorption in the kidney tubules. Changes in the kidneys were most intensive in the animals from group I.

In all experimental animals there were changes observed in the spleen, consisting of enlarged splenic follicles, multiplication of lymphoidal cells and congestions. Such changes are usually related to the stimulation of the immunological system. Use of improper fat in the diet for carnivourous fur-bearing animals has a negative effect on lipid accumulation. Studies by Rouvinen et al. (1991) and Rouvinen (1992) on different foxes given fish fat confirmed this effect. Improper fats in the diet may lead to morphologic damages of many internal organs of these animals. Studies by Beare-Rogers (1977) and Kinsella (1987) on rats showed that fish oil and rapeseed oil in erucic acid caused destruction processes and inflammation of many internal organs. Also Rouvinen and Niemelä (1992) observed degenerative changes in the liver and heart muscle, chronic colitis and kidney damages in blue foxes fed a diet containing fish oil.

Our own studies showed that the commercial fat "ERAFET" had a beneficial effect on the animals, reflected in the physiological activity of lysosomal and mitochondrial enzymes in the cells, and normal morphological picture of the internal organs.

Conclusion

Diets containing 4% and 8% powdered commercial fat "ERAFET" had a beneficial effect on the organism of polar foxes, stabilized the metabolism in liver cells and prevented morphological changes in the liver, kidneys and heart muscle.

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Original Report

Investigation of enzyme proteolytic activity in the fitch digestive tract

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Summary

The authors made an attempt to study a correlation between protein level in fitch diets and enzyme proteolytic activity in different sections of their digestive tract.

Fitches were fed diets at three protein levels: 14, 16.5, and 19 g of protein/1 MJ M.E., respectively. From animals killed at 25, 35, 90, 150, and 210 days (30 per term), samples were collected from the stomach mucous membrane, pancreas and small intestine. Total enzyme proteolytic activity was tested 'in vitro', by a modified Anson's method (*Bergmeyer, 1963*). The results obtained indicate that protein is digested mostly in the stomach and pancreas. There is a tendency to increased reaction activity with an increase in protein level in the feed. In the stomach, the highest activity was recorded in 90-day-old animals, 39.7, 40.0, and 41.1 μmol/min/g of protein, respectively. The pancreas enzymes displayed the highest activity in 150-day-old animals (19.1, 22.0, and 26.4 μmol/min/g of protein, resp.). Fitches and polar foxes show a similar type of digestion.

Introduction

A decrease in demand for mink pelts which has been observed in the world markets recently, caused some increase of interest in fitch pelts which are cheaper but also valuable from the fur market point of view, and have an original colour. This change in interest resulted in a growth of the fitch breeding business (*Bednarz and Szostak, 1983; Burberry, 1983; Korhonen, 1983*) and, consequently, a demand for information on breeding methods, feed, reproduction, and veterinary intervention has arisen.

Very little research work on feeding fitches has been completed so far (*Barabasz, 1986*), so it is important to learn and then introduce into practice basic principles of the use of particular nourishment components by the animals, as well as to investigate their demand for these components.

For a few years now, the problems of nitrogen balance and retention, as well as coefficient of protein digestion have been studied. Additionally, scientists have been studying problems of correla-

tion between the activity of proteolytic enzymes and the amount of protein in the diet of foxes and mink (*Asari et al., 1988; Bierestov and Olejnik, 1987; Elnif and Enggaard-Hansen, 1988*).

Since the results seem to be interesting, the authors have undertaken research work on these relations in fitches. The aim of this study was to investigate the dependence of protein levels in the diet upon proteolytic enzyme activities in various sections of the alimentary tract in young animals during their growth period.

Materials and methods

Female specimens with their youngsters were studied, all divided into three groups - each fed diets with different amounts of protein: group I - 14 g/l MJ M.E. of protein, group II - 16.5 g/l MJ M.E. and group III - 19 g/l MJ M.E. Components of the diet and their dietary values are presented in Table 1.

The activity of the proteolytic digestive enzymes was studied in 150 fitches, killed for research at the following ages: 25 days, fed dam's milk only, 35 days, fed the experimental diet, in their so-called transition period, and in somatic maturity period - at 90, 150, and 210 days. After slaughtering, samples of pancreatic tissue, mucous membrane from the stomach and small intestine were taken, frozen and stored at -25°C.

The enzyme activity in the samples was measured in vitro by the modified Anson's method (*Bergmeyer, 1963*). Samples were also homogenized in a physiological solution with addition of an activator (trypsine and triton X-100), then incubated at 37°C (stomach and intestine membrane for 20 min., pancreas for 15 min.) in a substrate solution (cattle albumen) in the presence of a lemon buffer which ensured a constant pH: 2.5 for stomach and 7.8 for pancreas and intestine samples. When incubation occurred, the process was stopped with 5% TCA.

Table 1 Dietary value and chemical composition of the diet (%)

Ingredients	I	II	III
1. Fish offal	8	8	8
2. Slaughter cow offal	25	25	25
3. Casein	3	3.5	4
4. Fish meal	3	4	5
5. Beef tallow	4	4.5	4
6. Barley, cooked	47	43	40
7. Yeast	6	6	6
8. Vegetables	4	6	8
- Digestible protein, %	8.1	9.4	11
- Digestible fat, %	8.3	7.5	7.4
- Carbohydrates, %	6.5	6.8	5.6
- Protein % of M.E.	26	30	35
- Fat % of M.E.	54	50	48
- Carbohydrates % of M.E.	20	20	17
Metabolizable Energy (MJ/kg)	580	580	580
Energy-protein ratio (g digest. protein/l MJ M.E.)	14.0	16.5	19.0

Both experimental and comparative samples (which contained non-incubated enzyme material) were centrifuged and the enzyme activity was evaluated based on changes in optical density of the solution, with the use of a spectrophotometer (wave-length 280 nm).

Results

A. Activity of proteolytic enzymes of stomach mucous membrane

The results obtained are given in Table 2. It was found that the enzyme activity was the highest in 90-day-old animals. This was assumed as 100% activity and compared with younger and older animals.

In 25-day-old specimens which were fed dam's milk only, the enzyme activity was low. It attained about 56-73% of the value observed in adult animals. However, we found a tendency to an increased activity in animals whose dams were fed a diet with a higher protein level. In the transition period, i.e. in 35-day old animals, when they had both dam's milk and some initial quantities of solid food, the enzyme activity increased substantially, up to 93-95% of 90-day-old animals' value. This can be considered as evidence of essential growth in the digestive function of the alimentary system.

The results obtained with 90-day-old animals, and the older ones, indicate that proteolytic activity of enzymes is nearly constant, since fluctuations in its value were small and not statistically significant.

Throughout the experiments a higher activity of enzymes was noted in animals which were given a diet with a higher amount of protein (groups II and III).

The enzyme activity observed in 150-day-old fitches, 38-65-42.17 μmol/min/g, was slightly different from the value obtained by Bierestow and Olejnik (1987), whose results were: 34.75 in mink, and 47.87 in foxes. This difference might be caused by various levels of protein in the diet, and by specific varieties (*Asari et al., 1988, Krogdahl and Holm, 1982*).

B. Activity of proteolytic enzymes of the pancreas

The proteolytic activity of pancreatic enzymes is presented in Table 2. As can be seen, this activity is highest in 5-month-old animals and amounts to 19.1-26.5 μmol/min/g. It indicates that digestive functions are fully developed in this period of life. In 25-day-old animals the enzyme activity was 65-80% and in 35-day-old ones 76-92% of the value observed in 5-month-old fitches.

Similar results were obtained in the studies on maturity of the digestive system in foxes and mink (*Bierestow and Olejnik, 1987; Olejnik, 1984; Ostaszkowa, 1982*), which indicates that adult mink (52.1) and polar foxes (44.3 μmol/min/g of protein) make better use of feed protein. These substantial differences, according to these authors, might be caused by species features or by various levels of protein in the feed used.

We also noted a high correlation between the amount of protein in the feed and enzyme activity in the pancreas. In animals of group II it was higher by about 25% and in group III - by 60%. Results obtained for group III were significantly different from those for group I. This correlation has been described in other publications (*Gawlikowska and Barabasz, 1989*), where the authors found that different amounts of protein in the feed resulted in varying pancreatic enzyme activity in the initial period. It was also suggested by these authors that in adult animals with fully developed pancreatic digestive functions, a decrease in the amount of protein in the feed does not cause a decrease in enzyme activity.

This conclusion seems to be proved by the present study. An increase in protein level in the feed caused an increased pancreatic enzyme activity in all animals including the adult ones (5-7 months of age). Statistical analyses did not show significant differences between age groups or any interaction between factors under consideration.

C. Activity of proteolytic enzyme in the small intestine

The authors carried out this research on mucous membrane taken from the jejunum and the results are presented in Table 2.

Table 2 Activity of proteolytic enzymes in the alimentary canal in fitch (μmol/min/g of protein)

Age (days)	x	s	Energy-protein ratio in diets					
			14.0		16.5		19.0	
			Enzyme activity	%	Enzyme activity	%	Enzyme activity	%
Stomach mucous membrane								
25	26.46	7.52	22.503	56.7	26.775	66.9	30.104	73.3
35	38.04	24.47	37.078	93.5	38.102	95.2	38.948	94.8
90	40.25	15.27	39.669	100.0	40.006	100.0	41.082	100.0
150	40.07	10.24	38.648	97.4	39.387	98.5	42.168	102.6
210	39.86	14.33	38.492	97.0	39.440	98.6	41.636	101.4
Pancreas								
25	14.74	7.36	12.460	65.2	15.270	69.4	21.211	80.2
35	19.65	8.85	14.605	76.4	19.963	90.8	24.387	92.2
90	20.60	12.56	15.953	83.5	20.841	94.8	25.530	96.5
150	22.52	9.54	19.107	100.0	21.994	100.0	26.448	100.0
210	22.66	8.38	20.720	108.4	21.098	95.9	26.173	98.9
Small intestine								
25	1.28	0.55	0.93	40.4	1.21	52.2	1.71	68.1
35	1.78	0.63	1.72	74.8	1.83	78.9	1.80	71.1
90	1.84	0.43	1.90	82.6	1.80	77.6	1.81	72.1
150	2.33	0.27	2.30	100.0	2.32	100.0	2.51	100.0
210	2.18	0.37	2.25	97.8	2.11	90.0	2.19	87.2

They found that the activity of these enzymes was relatively low, about 20 times lower than in the stomach, and almost 10 times lower than in pancreas. The highest activity of up to 2.3-2.5 μmol/min/g of protein was observed in 150-day-old animals, fully grown, and it was significantly different from 25-day-old specimens. The differences were important when age groups were analysed, but did not exist between groups fed diets with different amounts of protein. No interactions between studied factors were noted, either. According to Bierestow and Olejnik (1987), in adult mink the enzyme activity should be 3.17 and in foxes 2.53 μmol/min/g of protein. These results are similar to those obtained in fitches, presented above.

Also Rachimow et al. (1972), as well as Elnif and Enggaard-hansen (1988) have carried out research on enzyme activity in the small intestine in mink and foxes.

They agree that the proteolytic activity of enzymes in carnivorous animals is of medium intensity compared with that of omnivorous animals, in which it is much higher.

Conclusions

Our studies have proven the fact that in fitches the process of protein digestion is highly effective. A positive correlation between the proteolytic enzyme activity in the alimentary tract and the amount of dietary protein was also recorded. Since the authors used only diets containing 14, 16.5, and 19 g of protein/1 MJ M.E. the enzyme activity was increasing. However, the maximum value of activity was not attained. Possibly, the maximum point could be achieved by a comparatively small increase in dietary protein.

Our knowledge of digestive possibilities of enzymes might allow scientists to describe more precisely a more intensive use of protein in animals, and to find out the range of demand for it (*Bierestow and Olejnik, 1987; Gawlikowska and Barabasz, 1989*). Attempts are made to evaluate enzyme activity in the alimentary tract in vivo (*Olejnik, 1987*), making use of a correlation between this activity and the one of serum enzymes. The results obtained so far indicate that this correlation does exist.

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Biochemical and physiological investigations of the meal and syrup fractions from aqueous enzymatic rapeseed processing

Søren Krogh Jensen

The aim of the present study was to undertake biochemical and physiological investigations of the nutritional quality and value of the meal and syrup obtained by aqueous enzymatic rapeseed processing. The glucosinolates in rapeseed are considered to cause the most serious quality problems concerning optimal utilization of rapeseed. A large part of the work has thus been concentrated on these compounds. The high content of dietary fibre in rapeseed is evaluated in relation to the digestibility of protein and energy. Various other rapeseed constituents of importance for the quality of rapeseed, including aromatic choline esters and tannin are also considered and discussed briefly.

The physiological effects caused by seven pure glucosinolates ± myrosinases added to a casein based standard diet and fed to rats in N-balance trials have been investigated. The most sensitive effect observed was the decrease in biological value (BV) of the protein. A 23% decrease in BV was observed by feeding the rats with a diet containing 12.5 μmol sinapolygucoraphenin/g DM. Glucocheirolin, glucoraphanin, epiprogoitrin, glucotropaeolin, sinalbin and (2R)-glucobarbarin caused a decrease in BV at the level of 0.5 μmol/g diet DM, whereas other glucosinolates tested did not affect the animals at this dietary level. A level of 2.5 μmol glucosinolate/g DM affects most often BV. Other effects caused by too high concentrations of glucosinolates in the feed, are on the liver, thyroid and kidneys. Reduced weight gain and poor feed palatability were often observed. The presence of active myrosinases generally aggravates the negative effect 2-3 times. Transformation products of glucosinolates are therefore considered to be more harmful than the intact glucosinolates.

The development of a rapid and efficient isolation and purification procedure, also made it possible to isolate appreciable amounts of the chemically unstable 4-hydroxyglucobrassicin. This compound is the quantitatively dominating glucosinolate in many double low rapeseed varieties, and the importance of this glucosinolate in relation to the quality of rapeseed has been given appreciable attention during several years. The results now obtained with intact 4-hydroxyglucobrassicin showed only a slight negative effect on BV and other effects investigated. However, too high concentrations of transformation/degradation products of 4-hydroxyglucobrassicin are for various reasons considered unacceptable. Oxidation products of this glucosinolate also give rapeseed products an unwanted dark colour.

Results from studies of epimeric 2-hydroxy substituted glucosinolates have revealed appreciable differences in their physiological effects in rats. HPLC and NMR investigations of intact epimeric 2-hydroxy substituted glucosinolates, revealed the structural reasons for some of the characteristic differences between these epimers. HPLC has been developed as an efficient and easy method of analysis for these epimers. The results showed that (2R)-2-hydroxy substituted glucosinolates always elute earlier than the corresponding (2S)-form in HPLC of both intact and desulfo glucosinolates.

The aqueous enzymatic method of rapeseed processing is based on enzyme catalyzed cell wall degradation of dry milled rapeseed in an aqueous slurry. The process performed in a Pilot Plant scale comprise decanter and centrifuge separations following the enzyme catalyzed reaction and results in four fractions: oil, protein rich meal, syrup, and hulls. The process involves heat inactivation of myrosinases prior to addition of cell wall degrading enzymes, which are a multi-activity enzyme mixture from a selected strain of *Aspergillus niger*. This aqueous enzymatic process is performed under gentle conditions, without use of

organic solvents, and with a final spray drying of the meal fraction.

Effects on the rapeseed constituents caused by the processing have been investigated by use of chemical-biochemical analyses and animal trials. The low molecular weight (LMW) rapeseed constituents are extracted into the syrup fraction. This fraction contains the majority of the water soluble LMW rape constituents including glucosinolates, aromatic choline esters, phytate and phenolics. The meal fraction contains only low concentrations of the LMW compounds, a high content of protein, fat and insoluble dietary fibre. Compared to traditionally used methods of rapeseed processes this method results in a limited degradation of glucosinolates, even for the unstable 4-hydroxyglucobrassicin. The problems caused by harmful glucosinolate degradation products in rapeseed oil and meal are thus reduced correspondingly.

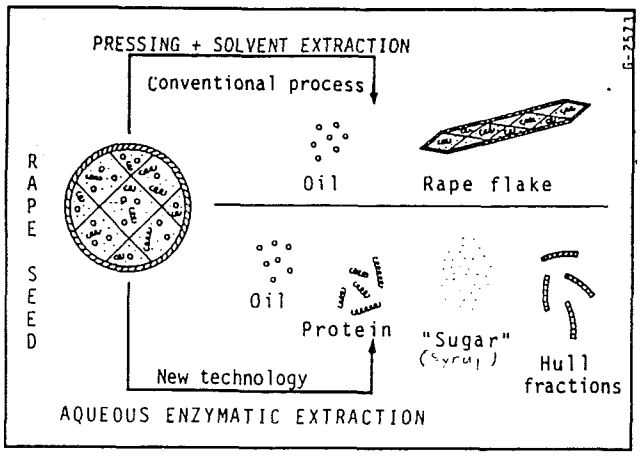

Fig. 5.1. Fractions obtained by the enzymatic procedure and the traditional procedure (Olsen, 1988).

Feeding experiments have been used for evaluation of the nutritive value and quality of the meal. These experiments comprised balance trials with young growing rats, piglets, chickens and long term studies with mink. No adverse effects from glucosinolates and products thereof have been revealed in the animal trials. This is in accordance with the chemical-biochemical results and the high biological values (BV) for the meal, BV = 98.3%, which have been obtained with appropriate feed mixtures. The low digestibility seen when feeding rapeseed has often been correlated to the high content of hulls. The meal obtained with the new type of rapeseed processing can, however, be further improved with respect to the digestibility. These remaining problems are most likely caused by protein association to the dietary fibres in the meal. Irrespective of the possibility of further quality improvements, the meal obtained in the aqueous enzymatic processing technique has a potential value as animal feed. In particular for young animals such as piglets, chickens, mink, and eventually small calves, where the requirements for protein quality are high.

Ph.D. Thesis, NOVO Nordisk A/S, Enzyme Process Development and the Royal Veterinary and Agricultural University, Chemistry Department, 1990. In ENGL, Su. DANH. 45 tables, 20 figs., 170 refs, 130 pp. Author's summary.

The importance of feed salt content for the incidence of nursing sickness

Tove N. Clausen, Søren Wamberg, Otto Hansen

Different investigations on salt in the feed for mink were discussed. Two groups of each 100 mink females were fed the same feed ration except for the content of NaCl. The energy distribution in the feed (protein:fat:carbohydrate) was 40:50:10). In one group the total salt content in the feed was 0.52 g NaCl/100 kcal. In the other group it was 0.22 g Nacl/100 kcal. The contents of Na and Cl in the urine and aldosterone in the blood were measured and the results discussed.

In the group with low salt content in the feed the average female weight loss at weaning were 100 grams more than in the group with high salt content and the incidence of nursing sickness in the respective two groups were 22% versus 7%. The conclusion was that the feed salt content in the nursing period should be about 0.40-0.45 NaCl/100 kcal.

In DANH. 6 tables, 2 figs., 11 refs. Technical Year Book 1993/94, pp 15-26, 1995.

The use of barley and peas for mink in the growing period

Tove N. Clausen, Niels Therkildsen

In the growing period 15 groups of each 80 males and 80 females Wild mink were fed barley, peas and combinations thereof at levels of 17, 21, and 25 per cent of the energy from carbohydrates. The amounts of energy from protein in the feed were 30%. A level of 25% of the energy from carbohydrates gave smaller pelts and a reduced pelt quality. when peas were used in the rations the skin were more silky.

The conclusion was that barley and combinations of barley and peas could be used at 17 and 21% of the energy from carbohydrates, but that a carbohydrate level of 25% of the energy and exclusively peas at all levels should be avoided.

In DANH. 7 tables, 3 figs. Technical Year Book 1993/94, pp 27-38, 1995.

Chicken offal for mink females from January to June

Niels Therkildsen, Tove N. Clausen

Four groups of mink of each 120 females were fed varying amounts of chicken offal from January to June. The energy distribution of the feed was about 53:37:10 and 11%, 20%, 29%, and 38% of the feed was chicken offal. The results showed a decrease in kit weight at weaning with increasing amounts of chicken offal in the feed.

In DANH. 4 tables. Technical Year Book 1993/94, pp 49-53, 1995.

The nursing period 1994: Experiments with increasing amounts of poultry offal in mink feed

Tove Clausen, Niels Therkildsen, Anette Svendsen

From the beginning of January 1994 until 42 days after birth, 6 groups of each 114 wild type mink females were given feed containing 8, 18, 23, 28, 34, and 38% boiled poultry offal, respectively. The energy content was approximately 125 kcal/100 g and the energy distribution 54:36:16 (protein:fat:carbohydrate).

The weight loss of the dams from parturition until weaning was least for the groups whose feed contained 18-28% poultry offal. With regard to the kit growth rate, this study seems to indicate that it cannot be recommended to surpass a level of 30% boiled poultry offal in the nursing period.

In DANH. 5 tables, 2 figs. Technical Year Book 1993/94, pp 54-61, 1995.

Attempt to reduce fish offal content in mink feed from January to June

Tove N. Clausen, Niels Therkildsen

Four groups of mink of each 118 females were fed varying amounts of fish offal and industrial fish from January to June. The fish offal content varied from 0 to 44 per cent and the industrial fish content varied from 54 to 0 percent.

The energy distribution (protein:fat:carbohydrate) was 52:37:11 (60:29:11 for the control group). The group fed no fish offal had the lowest kit weights at 42 days.

In DANH. 3 tables. Technical Year Book 1993/94, pp 62-66, 1995.

Recommendations for the supply of protein and amino acids in the growing-furring period of the mink. I: Experiments performed during 1992

Christian Friis Børsting, Tove N. Clausen

The mink has a very high protein requirement due to its evolutionary history, because mink are carnivorous animals. It has therefore - untill recently - been unknown whether the amino acid (AA) composition is of importance for the health and performance of the mink at the required level of dietary protein.

However, in 1990 we demonstrated the importance for both fur quality, skin size and health of AA

composition. Low content of essential AA resulted in short skins of low quality and the health of the animals was subnormal. During the present experiment it was demonstrated that maximal fur quality and skin length as well as normal health can be achieved with 30% of metabolizable energy (ME) from protein with a diet containing a relatively high proportion of essential amino acids, this proportion being similar to what is commonly used in Danish mink feed at the present time.

In 1992, an experiment was run during the growing-furring period, where up to half of each of the essential amino acids were supplied in crystalline form, allowing us to remove the amino acids individually, while the requirements of the other amino acids were met at the same time. It proved possible to use this concept when a basic mixture with only 15% of ME from protein was used.

Most emphasis was put on examining the requirements of the sulphur containing amino acids, methionine and cysteine, and of lysine, tryptophan, and threonine. For the remaining amino acids only an upper limit for the requirements could be given because only two levels were tested.

As previously shown, deficiency in sulphur containing amino acids resulted in the most pronounced effects on fur quality. However, it was also possible to show negative effects on fur quality, skin length or health due to deficiencies of the other essential amino acids.

The following recommendations for amino acids (g apparent digestible amino acids per MJ) during the growing-furring period were given: methionine: 0.382; cysteine: 0.167; lysine: 0.717 (0.645); tryptophan: 0.143 (0.120); threonine: 0.454 (0.406); histidine: 0.359 (0.382); phenylalanine: 0.693; tyrosine: 0.430; leucine: 1.195; isoleucine: 0.621; valine: 0.837 (values in ( ) are requirements for AA where the recommended supply differs from the requirements measured).

In DANH. 8 tables, 2 figs., 7 refs. Technical Year Book 1993/94, pp 79-99, 1995.

Recommendations for the supply of protein and amino acids in the growing-furring period of the mink. II: Experiments performed during 1993

Christian Friis Børsting, Tove N. Clausen, Niels Therkildsen

The experiments of this year were undertaken to further demonstrate the effects of different levels of protein from the presently common feedstuff composition and to more accurately asses recommendations for the amino acids (AA) methionine, cysteine and tryptophan.

In accordance with the findings of 1992 as described in the previous paper maximal skin size and quality was found when around 30% of metabolizable energy was from protein.

Methionine was - as in 1992 - shown to be by far the most important AA regarding fur quality, and therefore the recommendation was not altered.

The effect of cysteine was less clear despite there was a slight decline in fur quality when the level was reduced. There was not found a methionine sparing effect when cysteine suppply was increased. The recommended level was the same as given in the previous experiment.

Fur quality was reduced when tryphophan supply was decreased and therefore the recommended level of supply was unchanged compared to the findings of the previous year. The recommended levels comply with the present Danish Feed Table for feedstuffs for mink. However, in the experiments of 1992 and 1993 the measured levels of apparent digestible methionine, cysteine and tryptophan was lower than the levels computed from this table. Hence, the true requirements may be somewhat lower than the recommendations given here, which are (g apparent digestible amino acids per MJ): methionine: 0.382; cysteine: 0.167 and tryptophan: 0.143.

In DANH. 9 tables, 1 fig., 4 refs. Technical Year Book 1993/94, pp 100-120, 1995.

Effect of protein content in feed on blood variables and health condition in mink

Birthe Damgaard, Tove N. Clausen, Christian Friis Børsting

The effect of different protein content in the feed on blood parameters, fatty infiltration in the liver, frequency of wet belly disease, and the number of dead mink was measured in groups of male scanblack mink in two consecutive years from weaning until pelting.

The investigation included 5 groups fed 15%, 20%, 25%, 30%, and 35% of metabolizable energy (ME) from protein and one group fed 15% of ME from protein and supplied with essential amino acids to the levels in the diet with 30% of ME from protein.

Blood samples were collected in September and at pelting in December. Hematocrit was determined in whole blood and plasma was analysed for activity of the enzymes ALAT and CK, and for concentration of total protein, urea, total lipids, bile acids, and glucose. At pelting, the liver was weighed and the content of fat was estimated.

The activity of ALAT was increasing at decreasing content of ME from protein in the feed. The degree of hepatic fatty infiltration was significantly higher at a low protein level than at a high protein level in the feed.

No differences were found in degree of hepatic fatty infiltration between groups fed 15% ME from protein and 15% ME from protein supplemented with essential amino acids.

It is concluded that low protein (<25% of ME)/-high fat (>58% of ME) content in feed for mink affects physiological variables and health condition negatively.

In DANH. 8 tables, 2 refs. Technical Year Book 1993/94, pp 121-130, 1995.

Variations in fat and carbohydrate content of mink feed at a low amount of energy from protein and its importance to animal health

Tove N. Clausen, Birthe M. Damgaard

Four groups of mink in the growing period were fed the following energy distributions (protein:fat:-carbohydrate): 29:54:17, 15:68:17, 15:64:21, and 15:60:25. Blood samples were taken in September and at pelting, and the livers were examined at pelting.

Generally, the low protein content had a negative effect on all the parameters examined at all carbohydrate levels. Especially the degree of fatty infiltration in the liver increased with decreasing feed protein content. For some parameters, the negative effect of low protein could, to a certain degree, be compensated by an increase in carbohydrates and a decrease in fat.

In DANH. 5 tables. Technical Year Book 1993/94, pp 131-136, 1995.

Examination of feed consumption and female weight at the end of the nursing period

Tove N. Clausen, Søren Wamberg, Otto Hansen

Two groups of each 15 female mink were weaned at 6 or 7 weeks, respectively. The female weights during the last part of the nursing period at the end of lactation and after weaning were noted. Urine samples were taken. The feed was analyzed for sodium among other things. The amount of feed consumed was evaluated by judging the sodium excretion in urine.

It seems that females wean themselves by reducing their feed consumption. 45 days after birth they are physiologically weaned and increase their feed consumption again. The physical removal of the females from their kits often makes the females stop eating for a couple of days. If the female at

the same time is very thin because of many kits, she will very likely contract nursing sickness.

In DANH. 3 tables, 3 figs. Technical Year Book 1993/94, pp 137-145, 1995.

Transitional feed rations for mink: Investigation of the optimum start and finish dates, 1994

Anette Svendsen, Tove N. Clausen, Niels Therkildsen

The aim of the investigation was to determine the optimum start and finish dates for transitional feed rations for mink. Transitional feed is defined as the gradual change from nursing period feed to growing feed.

Combinations of three start dates (6/6, 13/6, 20/6) and three finish dates (4/7, 11/7, 18/7) were studied. In each of the 9 groups there were 76 standard black females with kits. All groups started and ended with the same start and finish rations. The number of intervening feed rations depended on the length of the transition periods.

Evaluation of the optimum start and finish dates was based on the average male and female kit weight gains on a litter basis in the period 2/6-19/7 and the dam weight loss in the same period. The latter was significantly less if the transitional feed ration started 6/6 or 13/6 compared to 20/6. However, the finish date had no significant effect on the dam weight loss. With respect to the male kit growth rate, starting the transitional feed on 20/6 was too late. The female kits in the group that started with transitional feed on 6/6 and ended 18/7 fared poorly with regard to their growth rates.

All in all, the two groups whose transition periods were from 6/6-4/7 and 13/6-18/7, respectively, had the best results with regard to kit growth rate, dam weight loss as well as faeces consistency and condition. The study will be repeated in 1995.

In DANH. 9 tables, 11 figs. Technical Year Book 1993/94, pp 146-163, 1995.

Palatability: ground alfalfa in the feed for grown male mink

Anette Svendsen

A study was undertaken to determine if mink have an aversion to the taste of ground alfalfa in the feed at a level of 5% on a wet basis. there were two groups in the study, each groups consisting of nine standard black males. In the first and fourth week of the experiment both groups could choose between feed containing alfalfa and feed without.

In the second and third weeks of the experiment, one of the groups was offered only feed containing alfalfa while the other groups were offered only feed without alfalfa. The study demonstrated a very significant preference for feed without alfalfa.

In DANH. 5 tables, 6 figs. Technical Year Book 1993/94, pp 164-170, 1995.

Determination of fat in feedstuffs and feed mixtures for mink

Christian F. Børsting, Birgit Hansen, Bent Munkøe

Acurate determination of the fat concentration in samples of mink feed can be very difficult due to high content of fat and due to very heterogeneous samples. In the present experiment fish silage, industrial fish, poultry offal, and 7 feed mixtures were analysed at both the Centrallaboratorium at the Danish Institute of Animal Science (DIAS) and at the Feed Laboratorium at Fur Center Vest (FCV). At FCV 2 different set of conditions for the fat analysis were applied and at DIAS one of these analysis were tested together with three other sets of conditions for the analysis which in all cases were performed with a SOXTEC HT 1043 EXTRACTION UNIT utilizing petroleum ether as the extraction solvent.

The method tested in both laboratories included HCl-hydrolysis directly in the wet samples without any pre-drying or pre-extraction. In the other method tested at FCV 5 g of sample was dried and grinded before 1 g was taken directly to the extraction unit without HCl-hydrolysis.

The remaining of the three methods used at DIAS all included freeze drying and HCl-hydrolysis, and one of these further included a pre-extraction step before HCl-hydrolysis and the last method included pre-extraction of 50 g of sample followed by a grinding step before HCl-hydrolysis.

Except for the method performed in wet samples including HCl-hydrolysis at FCV all methods gave very similar results with correlations from 0.96 to 1.00. For these 5 methods ratios between mean values of the 10 samples varied only between 1.000 and 1.014 compared to 0.979 for the method which differed slightly more. For all methods very good repeatabilities were found with mean CV-values as low as 0.4-1.6%.

It was concluded that both laboratories can perform fat analysis in mink feed with high repeatability when care is taken regarding sampling, grinding, and all of the steps in the analysis. Further, it was concluded that the results of the standard procedures of the two laboratories corresponds very well (r=0.97).

In DANH. 2 tables, 1 ref. Technical Year Book 1993/94, pp 171-178, 1995.

Microbiological activity in the intestine of mink

Bent Borg Jensen, Tove N. Clausen

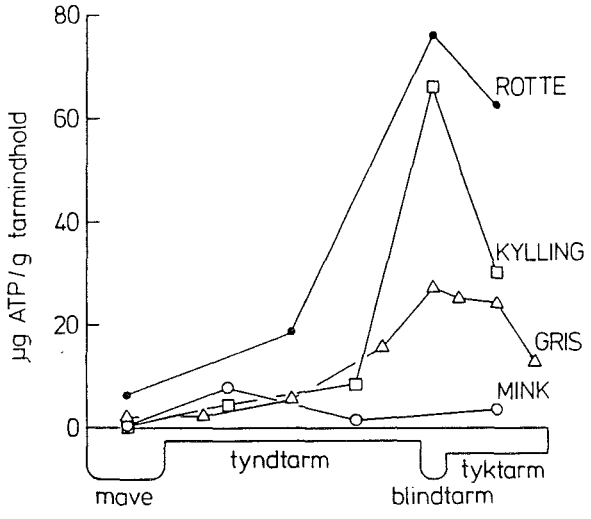

Fig. 2.

The amount of bacteria and the microbiological activity in the intestine of grown male mink were examined. Compared to pigs, the bacteria population in mink was 1.000 time slower in all parts of the digestive system. The highest bacteria concentration was found in the colon, and the highest microbial activity in the first part of the intestine.

In DANH. 6 tables, 2 figs. Technical Year Book 1993/94, pp 186-198, 1995.

Energy intake of captive adult-sized arctic foxes, *Alopex lagopus*, in Svalbard, in relation to body weight, climate, and activity

K. Frafjord

Food intake, change in body weight, and rate of inactivity were studied in two groups of arctic foxes *Alopex lagopus*, caged at Ny-Ålesund on the western coast of Svalbard (79°N). One group consisted of five "tame" foxes held in captivity for 9 to 28 months, and the other group consisted of 20 "wild" foxes held in captivity for 4 to 23 days. Daily energy intake varied between individuals, but no significant seasonal differences were found. Throughout the year mean energy consumption in tame foxes was 623.8 kcal·day^{-1} and in wild foxes 530.2 kcal·day^{-1}. Maintenance requirement was about 120 kcal·kg weight^{-1}·day^{-1}, or 360 kcal·day^{-1} for a 3 kg fox. Yearly mean weight for tame foxes was 3.37 kg, and no seasonal differences in weight were detected. Change in body weight (g/day) was correlated with energy intake (kcal·kg weight^{-1}·day^{-1}) in wild foxes, and for a single tame fox that was caught as an adult. Foxes were generally inactive 60-90% of the day, with no seasonal differences. Relationships between energy intake, inactivity, and weather (temperature and wind velocity) were weak or absent.

Z. Säugetierkunde 58, pp.2 66-274, 1993. 3 tables, 3 figs., 18 refs. Authors' abstract.

Original Report

The welchiosis (anaerobic enterotoxaemia) pathogenesis in mink

II. The virulence of C.welchii strains isolated from mink

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Summary

The authors studied the virulence of 182 C.welchii strains from healthy mink and mink with welchiosis and enteritis.

Determining the virulence of C.welchii type A and non-toxigenic, on an intramuscular route, in guinea pig, rat and mink, a positivity of 75%, 52.08% and 64%, respectively resulted, while on an intraperitoneal route, in mink, the result was 68%.

The ligated intestinal loops in mink and rabbit were positive by 37.86% and 30.76%, respectively.

The strains isolated from the mink with welchiosis and infectious enteritis gave maximum values in virulence tests, compared to the type A strains and non-toxigenic, with other origins.

Introduction

The pathogenicity of most germs from Clostridium genus which includes C.welchii, is manifested by toxicity and virulence. The studies dedicated to the C.welchii strains from different species of animals focused mainly on the toxicity, in order to typify them, particularly the toxigenic types like B, C and D. On the other hand, the type A strains, the most frequent C.welchii type, isolated from animals, human beings and in nature, were focused on in the virulence study (refs. 2, 3).

In a previous study, we have described the results of the toxicity of C.welchii strains isolated from mink (ref. 8).

Materials and methods

1. Strains. 182 C.welchii strains were studied. They were isolated from fresh corpses (intestine, liver, kidney, spleen and bone) and from the faeces, from sick and healthy animals.

2. Animals used for the experiments. a) guinea pigs, 350-500 grams weight; b) rabbits, 2.400-3.500 grams; c) adult mink, 850-1.000 grams; d) rats, 100-200 grams.

3. Methods. a) Virulence in animals. The strains were cultivated in liver broth, inoculated at 37°C,

for 17-24 hours and inoculated i.m. in the thigh muscle - 1 ml, 0.5 ml and 0.5 ml, respectively, in guinea pig, rat and mink, and 2 ml i.p. in mink. b) The ligated intestinal loops in mink and rabbits. The enteropathogenicity was determined on ligated intestinal loops of mink and rabbits, according to the method of E.coli (ref. 4). 2 ml of C.welchii toxin were inoculated/intestinal loop. A positive result (an enteropathogenic strain) was identified when a dilation index of the intestinal loop bigger than one was obtained, and a negative result (a non-enteropathogenic strain) when the dilation index was smaller than one.

Results and discussion

Table 1a shows the results of the determination of the virulence in guinea pigs and rats. 75% of 92 strains of C.welchii were virulent. In this case, a similar high level of activity of the C.welchii type A, isolated from the mink with welchiosis and with infectious enteritis could be seen. The strain from the healthy mink and those with non-infectious enteritis showed a moderate virulence; 57.14% and 44.44%, respectively. All strains produced myolysis in guinea pig. The local lesion was a gaseous oedema, which sometimes included both the opposite thigh and the subcutaneous conjunctive tissue in the abdominal wall and thoracic areas producing the characteristic lesion of gaseous gangrene. In most cases the development was 1-3 days. With some strains with a negative virulence, developed after inoculation to guinea pigs, only a local oedema occurred and the animals recovered.

The maximum values were determined also on the strains isolated from the rats with welchiosis and infectious enteritis, while minimum values were obtained on the non-toxigenic strains. Compared to the guinea pig, the rat is more resistant after being inoculated i.m. The majority of the inoculated animals showed congestions and bleedings, with a gaseous infiltration, with a weak intensity of the myolysis, without the typical image of the gaseous gangrene lesion determined in the guinea pig. The average development of the infection was 2-4 days, with the alteration of the general condition. 7 (30.4%) inoculated rats, with a negative reaction after inoculation, showed only a slight local reaction at the very point where inoculation took place. Compared to the tests done on the strains of different origins, the virulence of the strains was higher, especially in the type A strains. Out of a total of 555 type A strains, isolated from domestic animals (cattle, sheep, pigs, poultry) and from nature, 75.1% were virulent, 60.72% out of 220 non-toxigenic strains and 100% out of 103 strains of types B, C and D (ref. 4).

Table 1a The virulence in guinea pig and rat

Strain origin	Guinea pig						Rat					
	Examined strains		Positive		Negative		Examined strains		Positive		Negative	
	No	Type	No	%	No	%	No	Type	No	%	No	%
1. Mink with welchiosis	12	A	12	100	-	-	7	A	5	71.42	2	28.57
2. Mink with infectious enteritis	46	A	45	97.82	1	2.22	19	A	11	57.89	8	42.11
	8	NT	1	12.5	7	87.5	6	NT	1	16.66	5	83.33
a) pseudomonas	15	A	14	93.33	1	6.66	12	A	7	58.33	5	41.66
b) other infectious enteritis	31	A	31	100	-	-	7	A	4	57.14	3	42.85
	8	NT	1	12.5	7	87.5	6	NT	1	16.66	5	83.33
3. Mink with non-infectious enteritis	7	A	4	57.14	3	42.85	5	A	3	60.0	2	40.0
	2	D	2	100	-	-	2	D	2	100	-	-
	8	NT	1	12.5	7	87.5	4	NT	1	25.0	3	75.0
4. Healthy mink	9	A	4	44.44	5	55.55	5	A	2	40.0	3	60.0
Total	92		69	75.0	23	25.0	48		25.0	52.08	23	47.91

NT = non-toxigenic strains

Table 1b The virulence in mink

No	Examined strains		I.M. inoculation				I.P. inoculation			
			Positive		Negative		Positive		Negative	
	No	Type	No	%	No	%	No	%	No	%
1.	4	A	4	100	-	-	4	100	-	-
2.	8	A	5	62.5	3	37.5	7	87.5	1	12.5
A)	3	NT	1	33.33	2	66.66	2	66.66	1	33.33
B)	5	A	4	80.0	1	20.0	4	80.0	1	20.0
	3	A	1	33.33	2	66.66	3	100	-	-
	3	NT	1	33.33	2	66.66	2	66.66	1	33.33
3.	2	A	2	100	-	-	2	100	-	-
	4	NT	2	50.0	2	50.0	2	50.0	2	50.0
4.	4	A	2	50.0	2	50.0	-	-	4	100
25			16	64.0	9	36.0	17	68.0	8	32.0

In the case of strains isolated from mammals with welchiosis, enteritis and healthy, the positivity for rat was 69.82% (ref. 7). Out of 25 strains inoculated i.m. and i.p. in mink, 16 (64%) and respectively 17 (68%) were virulent, with a 3-4 days development of the infection and muscular flushed-bleeding lesions, with a weak gaseous infiltration and without myolysis (table 1b). With the i.p. inoculation, the development of the infection was 1-2 days, with haemorrhagic and degenerative lesions in the heart, liver and kidneys. The virulence of the strains isolated from mink with non-infectious enteritis was determined in most cases as 2-3 MLD/ml (ref. 5). At the time, the inoculation of these strains, in sub-lethal doses, also caused a synergistic lethal effect (ref. 6).

The test of ligated intestinal loops of mink and rabbit pointed out the presence of the enteropathogenity of 39 (37.86%) respectively 56 (30.76%) strains, with maximum values in the strains isolated from the animals with welchiosis (table 1c). None of the 24 non-toxigenic strains were enteropathogenics. The most virulent reactions, with the maximum quantity of liquid, were obtained from the middle third of the jejunum. The loops with a positive answer were dilated, with an enlarged volume with inflated veins and with the walls under pressure. Inside there were gases and liquid coloured yellow-grey or from bleedings, with mucous. The maximum liquid quantity obtained in mink and rabbit was 19 ml/loop and 27 ml/loop, respectively .

Table 1c Ligated intestinal loop assay in rabbit and mink

No	Rabbit						Mink					
	Examined strains		Positive		Negative		Examined strains		Positive		Negative	
	No	Type	No	%	No	%	No	Type	No	%	No	%
1.	14	A	7	50.0	7	50.0	14	A	10	71.42	4	28.57
2.	115	A	45	39.13	70	60.85	63	A	25	39.68	38	60.31
A)	18	NT	-	-	18	100	5	NT	-	-	5	100
B)	34	A	12	35.29	22	64.7	26	A	9	34.61	17	65.38
	81	A	33	40.74	48	59.25	37	A	16	43.34	21	56.75
	18	NT	-	-	18	100	5	NT	-	-	5	100
3.	15	A	-	-	15	100	8	A	1	12.5	7	87.5
	3	D	3	100	-	-	3	D	3	100	-	-
	6	NT	-	-	6	100	4	NT	-	-	4	100
4.	11	A	1	9.09	10	90.0	6	A	-	-	6	100
182			56	30.76	126	69.23	103		39	37.86	64	62.13

From our previous investigations, we found that the maximum enteropathogenicity was obtained from the type B and C strains (78.2%), known as having an intense bleeding-necrosis action on the intestinal mucous membrane, both in the natural disease and in the experimental one (refs. 1, 3). On the other hand, the type A strains, isolated from animals with welchiosis, showed a virulence of only 38%, the type D strains 27.3% and the non-toxigenic strains 2.3%.

In other experiments, type A strains isolated from pigs and calves, clinically healthy and with diarrhoea, 2% and respectively 12.7% were enteropathogenics (ref. 1).

Acknowledgements

The invaluable technical assistance of Mrs. Mariana Mailat, Mr. Barsan N., Mrs. Maria Borcea and Mr. Martin Iakob is gratefully acknowledged.

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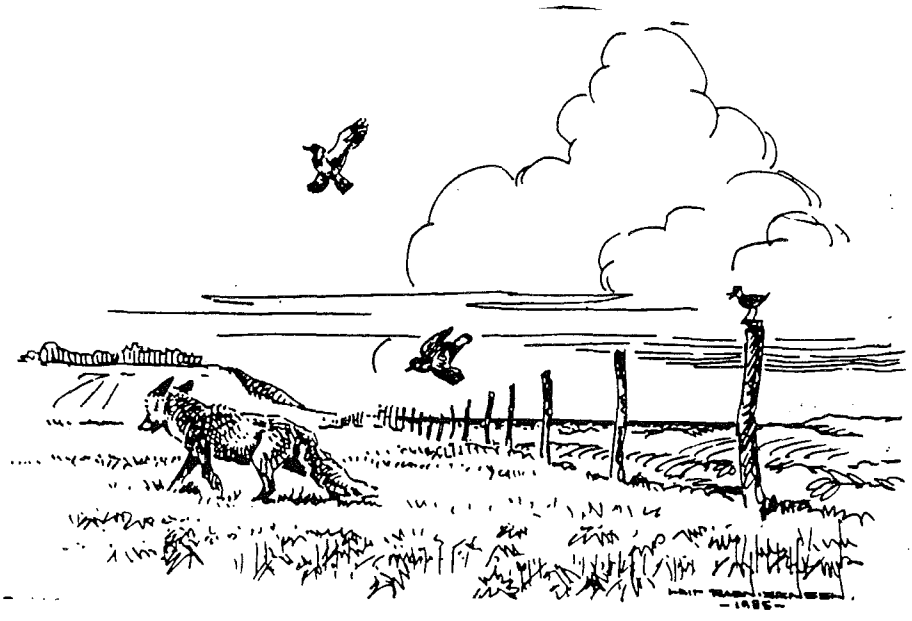

Original Report

Occurrence of the coccidia *Isospora laidlawi* and *Eimeria vison* in Danish farm mink, 1987-1993; Age related resistance to the infection

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Abstract

Infection with the 2 species *Isospora laidlawi* and *Eimeria vison*, was detected in Danish farmed mink from 1987 to 1993. In mink kits the *I. laidlawi* infection was present at a comparable level in all years with a mean prevalence of 24.1%, whereas the *E. vison* infection fluctuated at a lower level, with a mean prevalence of 6.4%. Infection of *I. laidlawi* was significantly lower in adult mink than in mink kits and no infection at all of *E. vison* was detected in adult mink. An age dependent resistance in the mink against both coccidia species was therefore evident and seemed to occur during the autumn.

Introduction

In continuation of investigations on coccidial infections in foxes on a Danish fur farm (*Hindsbo, Andreassen and Nielsen, 1991*), an investigation on the coccidial parasites of mink was carried out. The investigation was repeated yearly until 1993, when the farm was closed. Results from eight consecutive years are presented.

Materials and methods

From 1986 to 1993, a total of 159 cages, each containing a male and a female mink kit, and a total of 139 adult mink were examined on Gre-Ca Farm, situated in the middle of the island of Zealand.

In the first year, 1986, samples were taken in the autumn (Oct. 28.) from 8 kit cages and from 14 adult mink. From 1987 faecal samples were taken every year one day during late summer, usually in the last week of August. Collection and examination of the faeces were carried out as previously described (*Hindsbo, Andreassen and Nielsen, 1991*).

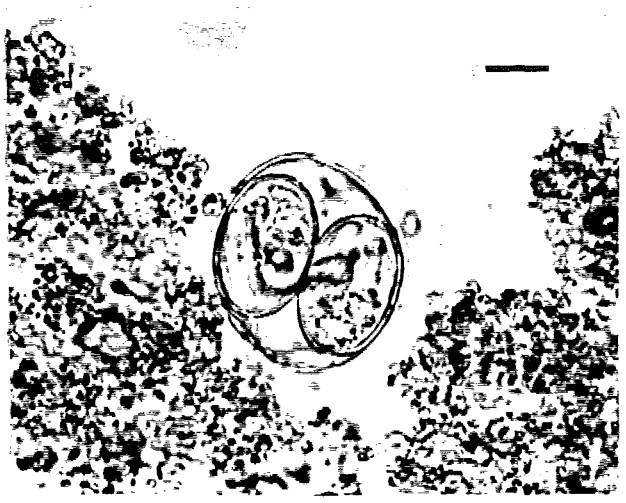

Fig. 1. Oocyst of *Isospora laidlawi*, fully sporulated. Note the two-layered outer wall and the two sporocysts containing banana-shaped sporozoites and granular material (the residuum). Bar represents 10 microns.

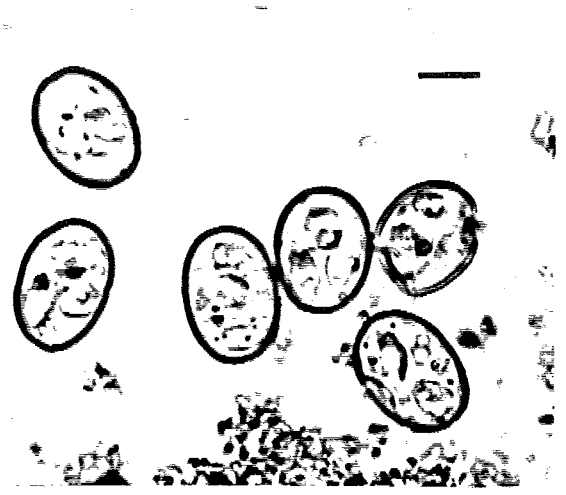

Fig. 2. Oocysts of *Eimeria vison*, fully sporulated. Three of the four sporocysts are seen in most of the oocysts. Bar represents 10 microns.

In all years, the mink kits were born within a narrow time range of about 2 weeks. In 1991 and 1992, the mean date of birth was the 2nd of May. Therefore, the samples were taken from 6-month-old kits in 1986 and from 4-month-old kits in the other years.

Oocyst-positive faeces from the 1993 samples were suspended in a 2% potassium dichromate solution and left to sporulate for size measurements as described by Long et al. (1976). The long axis of oocysts selected for measurement should lie in the phocal plane of the microscope. The correct position for measurement was obtained when the sporcysts were lying parallel to the long axis of the oocyst (*Long et al., 1976*). This was easily obtained for the *Isospora* oocysts, but for the *Eimeria* oocysts, both whole sample (random), as well as correctly positioned (selected) oocyst measurements, were carried out. Photos of a microscopic preparation of the sporulated oocyst were obtained, including a scale. Length and width of 100 different oocysts of each species were measured from the photos.

Results

In 1986, the coccidia were not identified, whereas, in 1987, oocysts from two species of coccidia were found. The morphology (Figure 1 and 2) of these oocysts was in agreement with the description of *Isospora laidlawi* and *Eimeria vison* by Levine & Ivens (1981). The range and maximum of the length frequency of the present random sample measurements of *E. vison* oocysts were also in agreement with the description by McTaggart (1960), as shown in Fig. 3, whereas the selected oocyst length measurements were in agreement with the description by Levine (1948). The length measurements of *Isospora* (Fig. 3) were in acceptable agreement with the measurements given by McTaggart (1960). The distribution of the calculated length/width ratios of the oocysts confirmed the identifications (Fig. 4a and 4b).

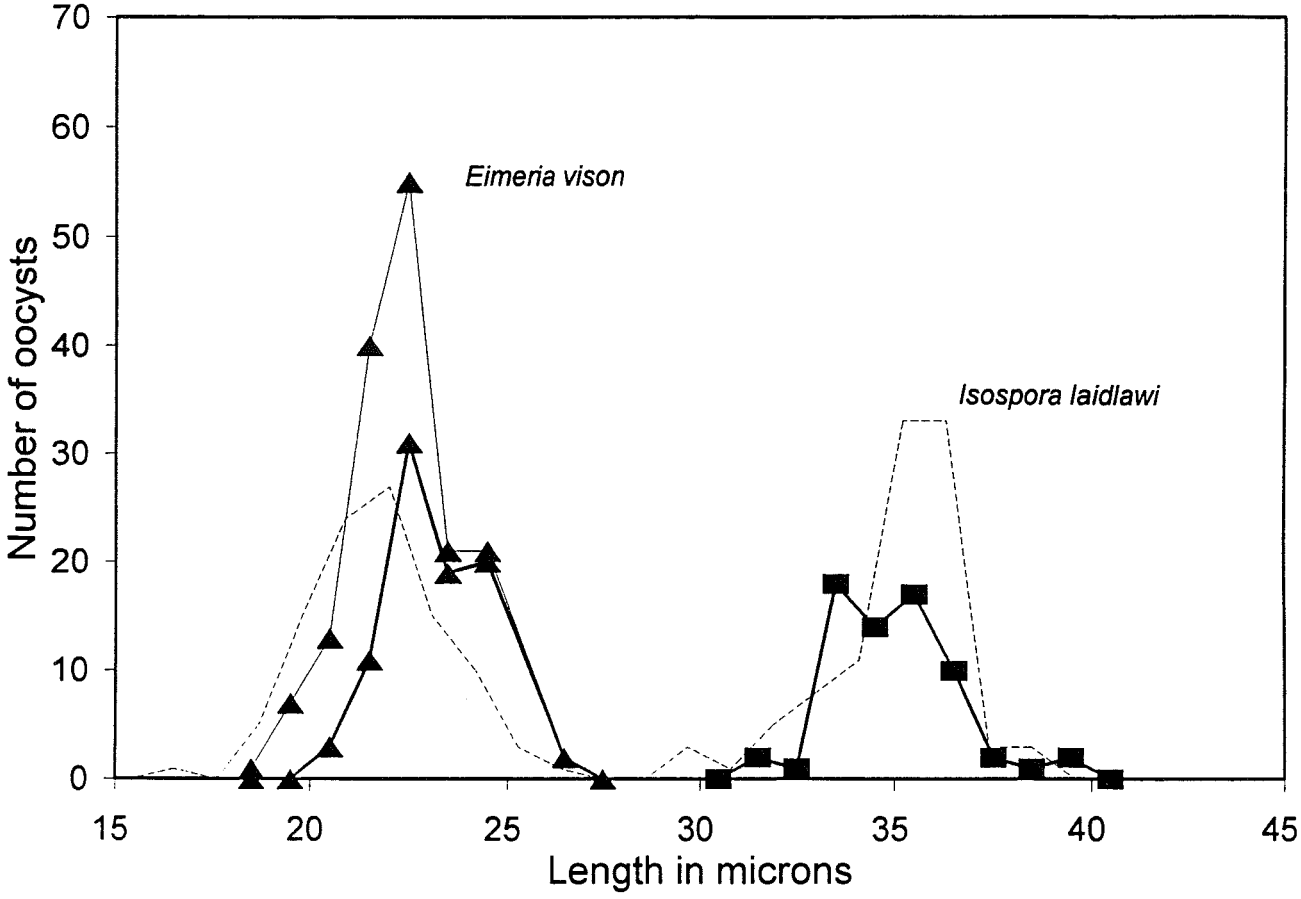

Fig. 3. Frequency distribution of length of oocysts from faeces collected at Gre-Ca farm: (▲——▲) *E. vison* selected oocysts, (▲——▲) *E. vison* random oocysts, (■——■) *I. laidlawi* selected oocysts, (----) results from McTaggart (1960).

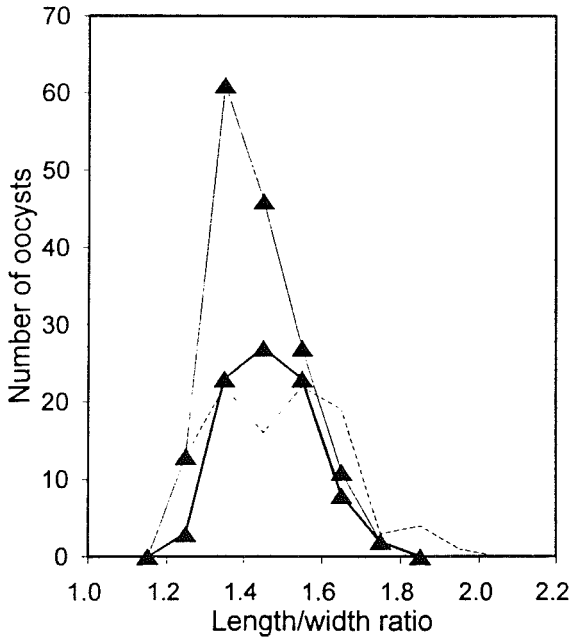

Fig. 4a. Frequency of length/width ratio of *Emeria vison* oocysts from mink faeces collected at Gre-Ca farm: (▲——▲) selected oocysts, (▲——▲) random oocysts, (----) from McTaggard (1960).

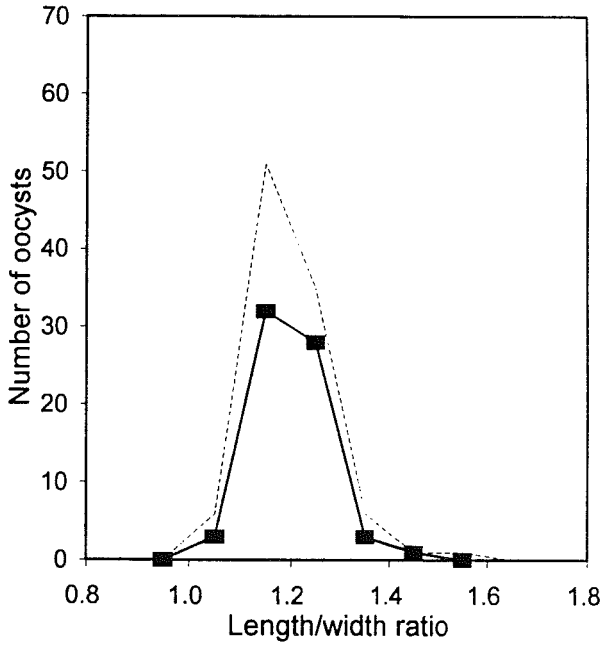

Fig. 4b. Frequency of length/width ratio of *Isospora laidlawi* oocysts in mink faeces: (■——■) from Gre-Ca farm, (----) from McTaggart (1960).

Table 1 Prevalence of coccidia in mink kits on Gre-Ca Farm

Year	Sample date	Number of pairs examined	Percent positive *Isospora*	Percent positive *Eimeria*
1987	Aug. 26.	29	24 %	10 %
1988	Aug. 24.	20	35 %	0 %
1989	Aug. 23.	21	29 %	5 %
1990	Sep. 5.	11	18 %	0 %
1991	Aug. 28.	19	16 %	0 %
1992	Aug. 26.	24	17 %	8 %
1993	Aug. 25.	27	30 %	22 %

Table 2 Prevalence of coccidia in adult mink on Gre-Ca Farm

Year	Sample date	Number of pairs examined	Percent positive *Isospora*	Percent positive *Eimeria*
1987	Aug. 26.	17	0 %	0 %
1988	Aug. 24.	21	10 %	0 %
1989	Aug. 23.	22	5 %	0 %
1990	Sep. 5.	10	0 %	0 %
1991	Aug. 28.	18	0 %	0 %
1992	Aug. 26.	17	0 %	0 %
1993	Aug. 25.	20	5 %	0 %

In the mink kits, *I. laidlawi* was present in all years (Table 1), the mean prevalence in 1987-1993 being 24.1%. Infections of *I. laidlawi* were also detected in adult mink (Table 2), the mean prevalence in 1987-1993 being 2.9%. The prevalence of *I. laidlawi* in the adult mink was significantly lower than in the kits ($p < 0.001$, chi-square test, Siegel, 1956) indicating an age-dependent resistance of the mink. In the kits, the incidence of *E. vison* was lower than *I. laidlawi*, the mean prevalence in 1987-1993 being 6.4%. In some years, the infection of *E. vison* was not detected at all but an exceptionally high incidence was seen in the kits in 1993. No *E. vison* was found in the adult mink.

In October 1986, the coccidia prevalence was 13% in the kits (Figure 5) and 7% in the adult mink.

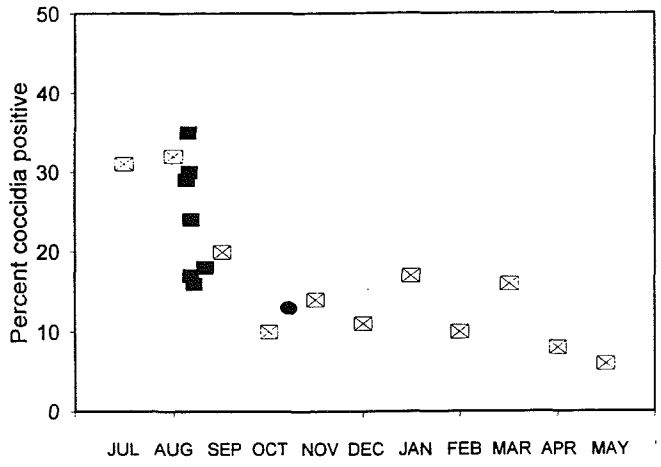

Fig. 5. Seasonal prevalence of coccidia oocysts in mink faeces from Denmark: (●) coccidia sp. in kits from Gre-Ca farm in 1986, (■) *Isospora laidlawi* in kits from Gre-Ca farm in 1987-1993, (☒) results from Henriksen and Andersen.

Prevalences of coccidia from farmed mink in Denmark have previously been reported by Henriksen and Andersen (1986). These data are rearranged and depicted in Figure 5, together with prevalences of *I.laidlawi* in kits from 1987-1993 in the present study. Henriksen and Andersen (1986) did not state the age of the mink, but the data fit well and show a marked decline during August and September. The few *I. laidlawi* positive adult mink were represented by both males and females; 2 females and 1 male were one year old and 1 female was 2 years old.

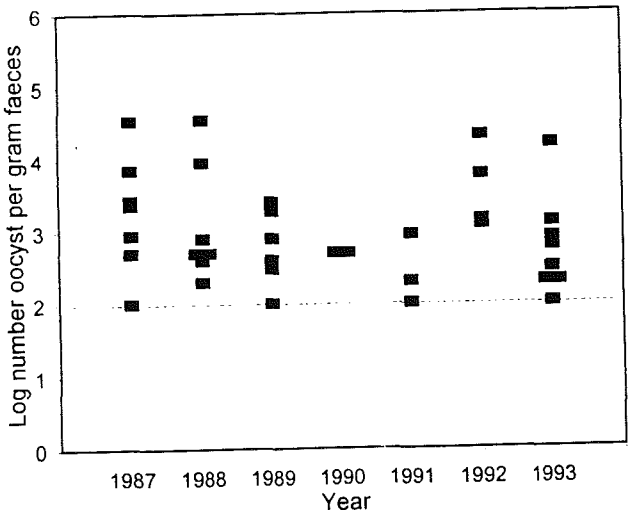

Fig. 6. Intensity of Isospora *laidlawi* oocyst output (■) from individual mink kits on Gre-Ca farm 1987-1993, (----) indicates lower limit of possible detection by the method used.

Intensity of the infections of *I. laidlawi* each years is given in Figure 6.

The frequency distribution of the intensity of *I. laidlawi* oocysts (Figure 7) demonstrate a remarkable similarity to the observation by Henriksen and Andersen (1986). Only 2.6% of the present mink kits examined excreted the highest concentrations of *Isospora* oocysts per gram faeces (15,500-34,800 o.p.g.), but on the other hand they comprised about 11% of the *Isospora*-infected mink kits. No mink kit produced more than 5,700 *Eimeria* o.p.g. except in 1993 where 1 kit produced 446,400 o.p.g.

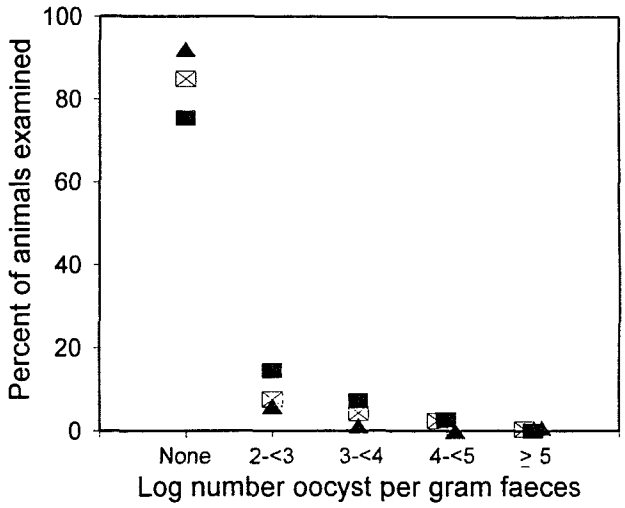

Fig. 7. Frequency distribution of oocyst intensity in mink kit faeces: (▲) *Eimeria vison* and (■) *Isospora laidlawi* from Gre-Ca farm 1987-1993, (☒) results from Henriksen and Andersen (1986). Due to the lower method limit of detection, oocyst concentrations below 100 oocysts per gram faeces are not detected.

Discussion

In the present investigation coccidia infections were found in mink kits during 8 consecutive years.

Tinar (1985) reported figures on prevalence to be 20% and 19% for *I. laidlawi* and *E. vison*, respectively, but stated neither the season of sampling nor the age of the mink. Nevertheless the prevalence figure (20%) reported on *I.laidlawi* is within the present range (16-35%) of observations from the mink kits and may fit even better if adult mink are included. In contrast most of the present observed prevalences of *E. vison* are lower than report-

ed by Tinar (1985) and much lower than recently reported (83.5%) by Jatusevich and Gerasimchik (1995). Except for 1988 there is a trend of positive correlation between the prevalence of *I. laidlawi* and *E. vison* in the kits, but the material is not conclusive on this point.

Likewise conclusive correlations to meteorological data are not possible although the external development to the fully sporulated infective oocyst is temperature dependent (*Soulsby, 1968*). But it can be mentioned that in 1993 when a high prevalence of *E. vison* was observed, the mean temperatures in May and August were the highest and lowest respectively of all the years. Likewise the highest August mean temperature occurred in the years 1990, 1991, and 1992 which were also the years when the lowest prevalences of *I. laidlawi* were observed.

Although an age dependent resistance was observed for both *Isospora laidlawi* and *Eimeria vison*, 1- and 2-year-old adult mink could still be observed infected with *I. laidlawi*. This agrees with the results of McTaggart (1960) who only found kits infected with *E. vison*, whereas *I. laidlawi* was found in adults "several years" of age.

As all mink kits are born almost simultaneously, a seasonal variation in prevalence should be expected, due to the occurrence of age dependent resistance. The highest values could be expected after weaning and, thereafter, a decline should be seen during the first year of life. This was confirmed (Figure 5) by comparing the present results with data from Henriksen and Andersen (1986). The data show a marked decline during August and September. This indicates that, although the present prevalences seem comparable between the years (Table 1), they are likely to be in an unstable decreasing phase. Recently, Jatusevich and Gerasimchik (1995) have observed 57% coccidia infection in 3-month-old kits and less than 12% infection in adult mink. Although the four species of coccidia involved were not differentiated according to the age of the mink this is most likely reflecting the present observed age dependent resistance of the mink. Specific acquired resistance, i.e. immunity, is a common event in coccidial infections. This has been studied mostly in *Eimeria* infections, but is suggested to be similar in infections with *Isospora* (*Rose, 1987*). It is therefore likely that the age dependant resistance of mink is due to immune responses. As almost all adult mink show the resistance this would further imply that probably all the mink acquire the infection as kits. The present prevalences makes this likely for the *Isospora* infection but for the *Eimeria* infections a much higher prevalence should then probably be expected in kits younger than the present 4-months-old-kits investigated.

Despite a difference in the selection of the mink material, both the prevalence of the coccidia infection of the mink (Figure 6) and the overall frequency of the intensity of oocyst production in infected mink (Figure 7) reported by Henriksen and Andersen (1986) are in remarkable agreement with the present data on *I. laidlawi* infection of the kits. Henriksen and Andersen (1986) state that coccidial infection intensities of more than 10,000 oocysts per gram faeces usually are regarded as an inducer of diarrhoeal disease in mammals. This was not clearly supported by their own results, but it could on the other hand be due to the fact that all their mink were sent for investigation because they had died for some reason that also could be reflected in the overall high prevalence of about 50% and 60% diarrhoea in the coccidia uninfected and infected mink respectively.

Only 2.6% of all the mink kits had an infection intensity of more than 10,000 oocysts per gram faeces. On the other hand these animals comprised a substantial part of the infected animals (11%) in the present investigation and 20% as calculated from the data presented by Henriksen and Andersen (1986). Considering that all the mink kits are expected to acquire the infection and that higher prevalences could be expected in the younger kits during the summer, then diarrhoeal disease due to coccidial infections could be significant in the mink kits.

Acknowledgement

We wish to thank cand. scient. Mogens Brandt for introducing us to Gre-Ca mink farm, Sylvia Holm for technical assistance and Grethe Drewsen for linguistic correction of the manuscript.

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Evaluation of protein A and protein G as an indicator system in an ELISA for detecting antibodies in mink to *Pseudomonas aeruginosa*

E. Rivera. M. Jackert-Jernberger, T. Meyerland, K.A. Karlsson

A modified, indirect enzyme-linked immunosorbent assay (ELISA) was developed and applied in the detection of mink antibodies to *Pseudomonas aeruginosa*. In this assay, peroxidase conjugated protein A and protein G were evaluated as indicator systems for detecting antigen-antibody complexes. It was found that protein A has a strong affinity for mink immunoglobulins. In contrast, protein G showed no such affinity. The affinity of protein A for mink immunoglobulins was further demonstrated by immunoprecipitation assays.

Veterinary Microbiology 42, pp. 265-271, 1994. 3 tables, 11 refs. Authors' abstract.

Copper toxicosis in sibling ferrets

James G. Fox, David H. Zeman, James D. Mortimer

Diagnosis of copper toxicosis in adult ferrets is based on high copper concentrations and excessive copper deposits in hepatic tissues, as well as characteristic hepatopathy.

This disease in ferrets may have an inheritable component.

JAVMA, vol. 205, No. 8, pp. 1154-1156, 1994. 2 figs., 14 refs. Authors' heading.

Emerging chloramphenicol resistance in *Staphylococcus lentus* from mink following chloramphenicol treatment: characterisation of the resistance genes

Stefan Schwarz

A total of 26 staphylococcal strains isolated from mink with urinary tract infections as well as from the environment of the mink were examined for antibiotic resistance and prevalence of plasmids mediating resistance to the antibiotics applied for prophylactic or therapeutic purposes. Chloramphenicol resistance (Cm^r) which occurred in fourteen of the eighteen *Staphylococcus lentus* strains, but in none of the *Staphylococcus intermedius* and *Staphylococcus xylosus* strains, was shown to be mediated by small plasmids of 3.6 to 4.6 kb. On the basis of restriction endonuclease mapping and hybridization experiments, four different types of Cm^r by encoding the Cm-inactivating enzyme chloramphenicol acetyltransferase (CAT). In all four types of Cm^r plasmids from *S. lentus*, the expression of the *cat* gene was inducible with Cm, as demonstrated by enzymatic assay and polyacrylamide gel electrophoresis.

Veterinary Microbiology 41, pp. 51-61, 1994. 2 tables, 3 figs., 29 refs. Author's summary.

A technique for vasectomizing male ferrets

L.M. Ryland, E. Lipinski

A surgical technique is described for vasectomizing male ferrets. An uncomplicated, rapid procedure that does not require invasion of the abdominal cavity or specialized instrumentation is outlined. No apparent adverse effects of surgery on the vasectomized male ferrets of the study were noted.

Canine Practice, Vol. 19, No. 1, pp. 25-27, 1994. 8 figs., 9 refs. Authors' summary.

Requirements for hemagglutination inhibition test for diagnosis of parvovirus infections of carnivores

Jerzy Górski, Andrzej Daniel, Beata Mizak, Jan Zwierzchowski

19 sera samples collected from dogs were tested by hemagglutination inhibition (HI) and serum neutralization (SN) tests with attenuated dog parvovirus. The results obtained were consistent only for sera inactivated at 56°C for 30 min and absorbed with 25% kaolin suspension. Non-specific hemagglutination inhibitors were removed neither by heat inactivation, absorption on porcine

erythrocytes nor by addition to the serum sample 20 μg of kaolin *in substantia*. A total of 255 samples of blue fox (*Alopex lagopus*) sera were tested by HI with canine and feline parvoviruses (CPV and FPV). Sera were either absorbed with 25% kaolin suspension or heat inactivated. The percentage of positive titre in sera testing with CPV was similar when tested with FPV, but it differed depending on the method of serum preparation for the test. 95% of the positive results were noted when unabsorbed sera were used while 455 positive results were observed when sera were absorbed. This difference was statistically significant.

Bull. vet. Inst. Pulawy 38, pp. 59-66, 1993. 3 tables, 16 refs. Authors' summary.

Prevalence of parvoviral antibodies in fox breeding farms

Jerzy Górski, Jan Zwierzchowksi, Beata Mizak, Andrzej Daniel

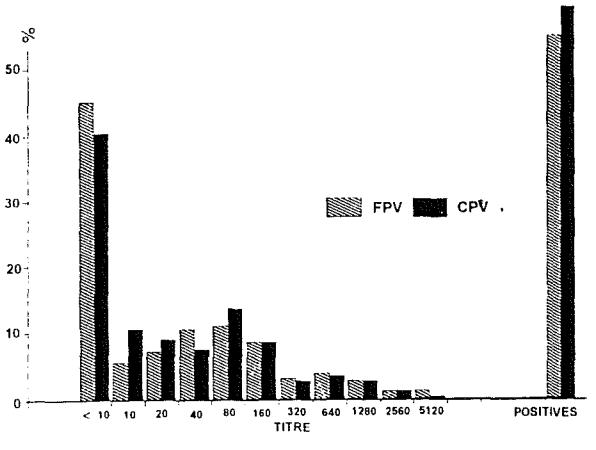

Fig. 1. Antibody level by HI in 255 fox sera tested with FPV and CPV.

A total of 392 sera of blue foxes and 12 sera of silver foxes was collected from 17 farms where foetus mummifications, abortions, neonatal deaths and, in consequence, reduced litter size were seen. In haemagglutination inhibition test (HI), dog and cat parvoviruses (CPV and FPV) were used as antigens. In 14 out of 17 fox farms prevalence of HI antibodies in titres ranging between 10 to 5120 were noted in sera tested with both viruses. In 3 farms, antibodies were not detected.

Acta Microbiologica Polonica 42, 2, pp. 157-162, 1993. 2 tables, 1 fig., 14 refs. Authors' abstract.

Diagnosis and treatment of campylobacteriosis on a fox farm

Jerzy Górski, Piotr Bugajak

The authors reported clinical and anatomopathological syndromes of an atypical diarrhoea in blue and silver foxes on the farm. More than 50% of the young animals were sick and 20% of them died within 2 weeks. Campylobacter jejuni was isolated on a differential medium incubated at 43°C.

The animals were treated with gentamycin (8 mg per day) and mineral premix with bentonite. During the recovery period an increase of the meat content up to 80% in the food was recommended.

Medycyna Weterynaryjna, 48, 11, pp. 504-505, 1992. In POLH, Su. ENGL. 1 table, 12 refs. Authors' summary.

Intracellular *campylobacter*-like organism from ferrets and hamsters with proliferative bowel disease is a *desulfovibrio* sp.

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Proliferative bowel disease is an intestinal disorder of a variety of domestic animals associated with the presence of an intracellular *Campylobacter*-like organism (ICLO). We have identified the ICLO obtained from a ferret with proliferative colitis by 16S rRNA sequence analysis. In this ferret, proliferative bowel tissue containing the ICLO had translocated to the mesenteric lymph nodes, omentum, and liver. The 16S rRNA genes of the ICLO were amplified from an infected fragment of extraintestinal tissue by using universal prokaryotic primers. Approximately 1,480 bases of the amplified 16S rRNA gene were sequenced by

cycle sequencing. Comparison of the sequence of the ICLO with those of over 400 bacteria in our data base indicated that the sequence of the ICLO was most closely related to that of *Desulfovibrio desulfuricans* (87.5% similarity). Phylogenetic analysis with 12 *Desulfovibrio* species and 20 species from related genera placed the ICLO in a subcluster within the genus *Desulfovibrio* with *D. desulfuricans* and 5 other *Desulfovibrio* species. We will refer to this organism as the intracellular *Desulfovibrio* organism (IDO). Specific primers were produced for PCR amplification of a 550-base fragment of the 16S rRNA gene of the IDO in proliferative intestinal tissue samples. This unique 550-base segment was amplified from samples of frozen intestinal tissue from nine ferrets and three hamsters with ICLO-associated disease but not in four intestinal tissue samples from animals without the ICLO-associated disease. The 550-base amplified products from the bowel tissues of one hamster and one ferret were fully sequenced. The ferret IDO partial sequence was identical to the previously determined 16S rRNA sequence over its length, and the hamster IDO sequence differed by a single base. The same intracellular organism has been identified in proliferative intestinal tissues of swine and the organism has been successfully maintained in tissue culture. The availability of specific primers for PCR-based detection of this intracellular *Desulfovibrio* organism will aid in the determination of its role in the pathogenesis of proliferative bowel disease in a variety of infected hosts.

Journal of Clinical Microbiology, Vol. 32, No. 5, pp. 1229-1237, 1994. 3 tables, 3 figs., 56 refs. Authors' summary.

Control of scabies in breeding foxes

Stanislaw Paciejewski

The samples of skin scrapes taken from foxes of three farms revealed the presence of Sarcoptes scabie var. canis and Otodectus cynotis. Out of 1648 silver and polar foxes their lesions on the head, legs and back of 169 animals were observed.

For treatment a dose of 300-400 meg Ivomec per 1 kg of body weight and a solution of Biocyd (0.1 per cent) were used. Ear scabies was treated with a 0.4% solution of Biocyd in paraffinum liquidum. It was found that Ivomec at a rate of 300-400 meg per 1 kg of body weight was safe and highly effective in the control of scabies in breeding foxes. However, to get a therapeutic effect it was necessary to carry out disinfection of the premises for the animals and to spray the drug over the surface of animals with no signs of lesions. The process of hair renewal was remarkably shorter and the quality of skins was higher if the feed was enriched with minerals and vitamins. A solution of Biocyd (0.4 per cent) proved to be very effective both for prevention and therapy of scabies in breeding foxes.

Medycyna Wet. 48 (11), pp. 506-508, 1992. In POLH, Su. ENGL. 1 table, 2 figs., 10 refs. Author's summary.

Encephalomyocarditis virus infection in raccoons (*Procyon lotor*)

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To determine the susceptibility of raccoons (*Procyon lotor*) to infection with encephalomyocarditis virus (EMCV), 1-yr-old raccoons were exposed intramuscularly (n=1) or orally (n=6). Serum samples were collected at 3-7-day intervals beginning 14 days prior to exposure and continuing to postexposure day (PED) 107. EMCV-specific antibody titers were measured by a serum virus neutralization (SN) test. In the intramuscularly (i.m.) exposed animal, elevated and stable SN antibody titers (1:64 to 1:128) were present from PED 11 through 107. Among orally exposed raccoons, antibody titers were detected in one of six animals. In contrast to the i.m.-exposed animal, antibodies in this individual were low ($\leq$1:16) and transient, falling to undetectable levels by PED 64. Fecal samples for virus isolation were collected four times before exposure, for 10 days after EMCV exposure, and at biweekly or weekly intervals for an additional 82 days. Whole blood for virus isolation was collected in sodium citrate on PED 3, 6, 8, and 11. No virus was detected in

fecal samples or whole blood by mouse inoculation assay from any of the raccoons. No overt signs of disease were observed in raccoons over the course of the experiment following exposure by either route. No gross lesions or histopathologic changes attributed to EMCV infection were detected. Serum samples (n=380) from a free-ranging population of raccoons trapped in Guthrie County, Iowa over a 5-yr period (1984-1988) were tested for neutralizing antibodies against EMCV. The population cross section included adults and juveniles of both sexes. Antibody titers were $\leq$1:4 in all samples by the SN test. The absence of elevated neutralizing antibody titers suggested that EMCV was not circulating in the wild population. Although raccoons were shown to be susceptible to infection with EMCV, the cumulative results of the experimental and field studies suggest that raccoons are a dead-end host for EMCV and do not participate in the epidemiology of the disease.

Journal of Zoo and Wildlife Medicine, 25 82), pp. 233-239, 1994. 1 table, 1 fig., 30 refs. Authors' summary.

Characterization of chimeric full-length molecular clones of aleutian mink disease parvovirus (ADV): identification of a determinant govering replication of ADV in cell culture

Marshall E. Bloom, Bradley D. Berry, Wu Wei, Sylvia Perryman, James B. Wolfinbarger

The ADV-G strain of Aleutian mink disease parvovirus (ADV) is nonpathogenic for mink but replicates permissively in cell culture, whereas the ADV-Utah 1 strain is highly pathogenic for mink but replicates poorly in cell culture. In order to relate these phenotypic differences to primary genomic features, we constructed a series of chimeric plasmids between a full-length replication-competent molecular clone of ADV-G and subgenomic clones of ADV-Utah 1 representing map units (MU) 15 to 88. After transfection of the plasmids into cell culture and serial passage of cell lysates, we determined that substitution of several segments of the ADV Utah 1 genome (MU 15 to 54 and 65 to 73) within an infectious ADV-G plasmid did not impair the ability of these constructs to yield infectious virus in vitro. Like ADV-G, the viruses derived from these replication-competent clones caused neither detectable viraemia 10 days after inoculation nor any evidence of Aleutian disease in adult mink. On the other hand, other chimeric plasmids were incapable of yielding infectious virus and were therefore replication defective in vitro. The MU 54 to 65 *Eco*RI-*Eco*RV fragment of ADV-Utah 1 was the minimal segment capable of rendering ADV-G replication defective. Substitution of the ADVG *Eco*RI-*Eco*RV fragment into a replication-defective clone restored replication competence, indicating that this 0.53-kb portion of the genome, wholly located within shared coding sequences for the capsid proteins VP1 and VP2, contained a determinant that governs replication in cell culture. When cultures of cells were studied 5 days after transfection with replication-defective clones, rescue of dimeric replicative form DNA and single-stranded progeny DNA could not be demonstrated. This defect could not be complemented by cotransfection with a replication-competent construction.

Journal of Virology, Vol. 67, No. 10, pp. 5976-5988, 1993. 1 table, 7 figs., 82 refs. Authors' summary.

Transmissible mink encephalopathy species barrier effect between ferret and mink: PrP gene and protein analysis

Jason C. Bartz, Debbie I. McKenzie, Richard A. Bessen, Richard F. March, Judd M. Aiken

Experimental infection of transmissible mink encephalopathy (TME) in two closely related mustelids, black ferret (*Mustela putorius furo*) and mink (*Mustela vison*), revealed differences in their susceptibility to the TME agent. When challenged with the Stetsonville TME agent, a longer incubation period was observed in ferrets (28 to 38 months) than mink (4 months). Wester blot analysis of ferret and mink prion proteins (PrP) demonstrated no detectable differences between the proteins. Northern blot analysis of ferret brain RNA indicated that PrP mRNA abundance is similar in infected and uninfected individuals. We amplified the PrP coding region from ferret DNA using the polymerase chain reaction and compared the

deduced amino acid sequence of the ferret PrP gene with the mink PrP gene. This comparison revealed six silent base changes and two amino acid changes between mink and ferret: Phe → Lys at codon 179 and Arg → Gln at codon 224, respectively. these changes may indicate the region of PrP that is responsible for the species barrier effect between mink and ferret.

Journal of General Virology, 75, pp. 2947-2953, 1994. 2 tables, 4 figs., 4 refs. Authors' summary.

Expression of Aleutian mink disease parvovirus capsid proteins in a baculovirus expression system for potential diagnostic use

Wai-Hong Wu, Marshall E. Bloom, Bradley D. Berry, Michael J. McGinley, Kenneth B. Platt

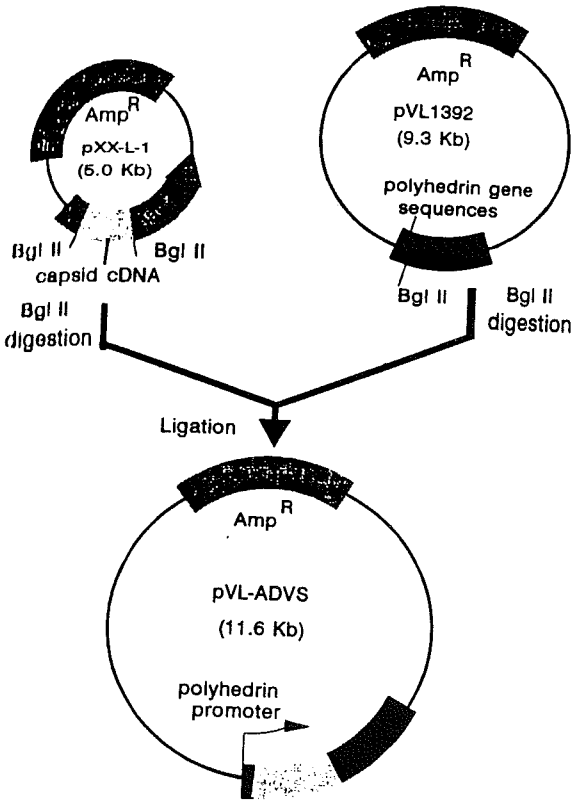

Fig. 1. Construction of the recombinant baculovirus transfer plasmid pVLADVS.

A 2.3-kb cDNA clone encoding Aleutian mink disease parvovirus (ADV) structural proteins VP1 and VP2 was inserted into the polyhedron gene of *Autographa californica* nuclear polyhedrosis virus (AcNPV) and expressed by the recombinant virus. AcADV-1, in *Spodoptera frugiperda*-9 cells. Sodium dodecyl sulfate-polyacrylamide gel electrophoresis and western immunoblot analysis (W1A) indicated that synthesis of both VP1 and VP2 was being directed by AcADV-1. Fluorescence microscopic examination of AcADV-1-infected *S-frugiperda*-9 cells indicated that the recombinant protein was present within the nucleus of the cells, and electron microscopic examination of these cells revealed the presence of small particles 23-25 nm in diameter. Structures resembling empty ADV capsids could be purified on CsC1 density gradients, thus indicating that the ADV proteins were self-assembling. The antigenicity of recombinant VP1 and VP2 was evaluated by W1A. Sera collected from 16 mink prior to infection with ADV did not react with VP1 and VP2. Ten sera collected from mink with counter current immunoelectrophoresis (CIE) titers greater than 4 (log_2) reacted with VP1 and VP2 in W1A. Two of 6 sera with CIE titers of 4 and 1 of 14 sera with CIE titers <4 reacted with the recombinant proteins. These results suggest that baculovirus recombinant ADV capsid proteins may be useful as diagnostic antigens.

J Vet Diagn Invest 6, pp. 23-29, 1994. 1 table, 6 figs., 22 refs. Authors' abstract.

Sequence comparison of the non-structural genes of four different types of Aleutian mink disease parvovirus indicates an unusual degree of variability

E. Gottschalck, S. Alexandersen, T. Storgaard, M.E. Bloom, B. Aasted

The present work shows that at least four different sequence types of Aleutian mink disease parvovirus (ADV) are present in ADV isolates from mink. We here report the nucleotide sequences of these four types of ADV from nucleotide 123 to 2208 (map unit 3 to 46). This part of the genome encodes three non-structural (NS) proteins of ADV. Comparison of the deduced amino acid sequences of these NS proteins showed that the ADV proteins are much less conserved than the NS proteins from other members of the autonomous group of parvoviruses. In general, we found that the middle region of the ADV NS-1 protein

was relatively well conserved among the types, while both the amino- and carboxy-terminal ends of the protein had higher amino acid variability. Interestingly, the putative NS-3 protein from type 3 ADV is truncated in the carboxy-terminal end. The molecular evolutionary relationship among the four types of ADV was examined. This analysis, taken together with the unusually high degree of variability of the ADV types, indicates that the ADV infection in mink is likely to be an old infection compared to the other parvovirus infections or, alternatively, that ADV accumulates sequence changes much faster than other parvoviruses.

Arch Virol, 138; pp. 213-231, 1994. 1 table, 4 figs., 70 refs. Authors' summary.

Practical venipuncture techniques for the ferret

Glen Otto, William D. Rosenblad, James G. Fox

As the number of ferrets (*Mustela putorius furo*) used in research and kept as pets continues to rise, so does the need for simple, humane research and diagnostic techniques. We have developed venipuncture methods for the ferret utilizing the jugular and cephalic veins. Using these methods it is possible to repeatedly sample moderate volumes of blood and to perform intravenous injections in both conscious and sedated ferrets.

Laboratory Animals, 27, pp. 26-29, 1993. 4 figs., 13 refs. Authors' summary.

A technique for catheterization of the urinary bladder in the ferret

R.P. Marini, M.I. Esteves, J.G. Fox

The technique of catheterization of the urinary bladder, an important clinical skill for the diagnosis of urinary tract disorders, has not been described for the ferret. The bladder was catheterized in 23 ferrets (10 intact females, 11 spayed females, and 2 intact males) using a 3½ French, red rubber urethral catheter fitted with a steel wire stylet. Ferrets were anaesthetized with isoflurane or ketamine (30 mg/kg IM) and zylazine (3 mg/kg IM). Females were positioned in ventral recumbency with the rear quarters elevated by a rolled surgical towel. The urethra was catheterized by direct visualization of the external urethral orifice using a vaginal speculum. The orifice was approximately 1 cm cranial to the clitoral fossa on the ventral floor of the vestibule. Blind passage was used in several spayed females. In males, the distal end of the penis was exteriorized from the prepuce and the external urethral orifice cannulated without stylet. No difficulty was encountered in advancing the catheter past the os penis. This catheterization technique allows urinary tract access for urine collection, pneumocystography, contrast cystography, double contrast cystography, and urine output determination in pharmocologic studies or in critical care of debilitated animals.

Laboratory Animals, 28, pp. 155-157, 1994. 2 figs., 3 refs. Authors' summary.

Airborne particulate matter, fungi, bacteria and endotoxins in fur farming

Rainer W. Schimberg, Jukka Uitti, Marjut Kotimaa, Riitta Sarantila

Epithelial and excremental matter from animals, feed and bedding material are sources of exposure to airborne organic matter, especially fungi, bacteria and endotoxins in animal farming. Exposure can cause organic dust toxic syndrome, and repeated exposure can cause allergic alveolitis. On one farm with 80,000 mink and 56,000 foxes the concentrations of airborne total particulate matter, respirable particulates, fungi, bacteria and endotoxins were determined for different work phases. High concentrations of fungi and bacteria were found during skinning (135,000 cfu/m^3) and the replacing of bedding material in mink nests (64,000 cfu/m^3). The highest endotoxin concentration (1.95 $\mu g/m^3$) was measured during the latter of the two operations. During the killing of mink, the endotoxin concentration was 0.23 $\mu g/m^3$. The risk for fever reactions and irritative symptoms among the workers during these tasks was

increased. Because these tasks are seasonal, it is suggested that the workers use personal protective equipment to diminish the level of exposure.

Staub-Reinhaltung der Luft, 52, pp. 457-460, 1992. 4 tables, 22 refs. Authors' summary.

Megaesophagus in nine ferrets

Michael C. Blanco, James G. Fox, Karen Rosenthal, Elizabeth V. Hillyer, Katherine E. Quesenberry, James C. Murphy

Megaesophagus develops in adult ferrets. The etiopathogenesis is unknown.

Diagnosis of megaesophagus can usually be made on the basis of radiography. Esophagoscopy or positive-contrast esophagography may be used to detect trauma, stricture, or obstruction.

Megaesophagus in ferrets is often complicated by dehydration, malnutrition, hepatic lipidosis, and aspiration pneumonia. Treatment includes parenteral administration of fluids and antibiotics, and furnishing nutrients by feeding a semiliquid diet while maintaining the ferret in an upright posture.

JAVMA, Vol. 205, No. 3, pp. 444-447, 1994. 3 figs., 10 refs. Authors' summary.

Animal spongiform encephalopathies - an update. Part 1. Scrapie and lesser known animal spongiform encephalopathies

B.E.C. Schreuder

The present article (part 1) reviews recent developments in animal spongiform encephalopathis (SEs), with the exception of bovine spongiform encephalopathy (BSE), which is dealt with in part II.

The article focuses on scrapie and describes epidemiological aspects and the prospects for a preclinical diagnosis. Up to now, confirmatory diagnosis of scrapie depended on histological examination of the brain, collected during post-mortem examination from sheep with clinical signs of the disease. An altered protein, PrP^{Sc}, can be detected in the brain of diseased animals. The demonstration of the same protein in the spleen and in peripheral lymph nodes of infected animals seems to offer interesting possibilities of arriving at a method for a preclinical diagnosis, and thus a diagnosis in the live animal.

Progress has also been made in our understanding of the relationship between the genetic constitution and susceptibility of the host. Susceptibility is expressed as the survival time of sheep inoculated with scrapie. This was thought to be determined by a single genetic locus designated the *Sip* gene (scrapie incubation period gene). Putative markers for the two alleles of the *Sip* gene, sA and pA, have been discovered, consisting of restriction fragment length polymorphisms (RFLPs). In field tests, however, the link between these markers and the length of incubation time was far from consistent. These RFLPs were found to be situated outside the prion-protein-coding region of the ovine gene. In later studies, RFLPs were detected inside this region. These markers appear to be more informative, i.e. they correspond with a difference in the length of the scrapie incubation period.

Finally, the article briefly describes recent developments in other, lesser known, animal spongiform encephalopathies: chronic wasting disease and other spongiform encephalopathies in exotic ungulates, transmissible mink encephalopathy, and feline spongiform encephalopathy, focusing on their possible links with scrapie or bovine spongiform encephalopathy.

The Veterinary quarterly, Vol. 16, No. 3, pp. 174-181, 1994. 2 tables, 3 figs., 71 refs. Authors' summary.

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