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Production of Mink

The influence of various management, environment and nutritional elements on behaviour, physiology and production in mink

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The influence of various management, environment and nutritional elements on behaviour, physiology and production in mink

Indflydelse af forskellig driftsledelse, miljø og ernæring på adfærd, fysiologi og produktion hos mink

Med dansk sammendrag

Manuskriptet afleveret december 1990

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PREFACE

This report represents summaries and conclusions of all experimental sections of the largest single project this department has carried out. The project was entitled: Production factors and production control. The results of the investigations on the farms regarding management and environment will be published in a special report.

The project was financed by the Ministry of Agriculture through The Joint Committee for Agricultural Research and Experiments as part of the research programme: Production Systems in Agriculture (1984-88), The Danish Fur Breeders Association, and the appropriations allocated to the department by the government.

The official title of the very broad project was: The influence of management and environmental factors on behaviour, health and productivity of fur animals, illustration of the applicability of various blood parameters for health control as well as an investigation of the influence of minerals on fur characteristics and blood parameters in mink. Investigations regarding foxes were, however, from the beginning separated into an independent internordic project.

The work within the different areas of the project has been diversified. Within environment, behaviour, and production the experimental work has primarily been concentrating on behaviour and reactions of the mink to different production environments. A considerable part of this work has included development and testing of measuring methods. A method for determination of behaviour by a stick test, sampling and analysis of blood as well as methods for determination of light reduction were developed. Choice of drinking water temperature was tested under experimental conditions. Greatest importance was, however, placed on experiments with mink in different environments.

The comparative work with mineral content in feed and hairs included a great deal of method development as regards analysis of minerals in mink hairs. Morphological methods were applied for description of hair types and used for basic studies on normal variation between farms.

The work with haematology and clinical-chemical parameters has mainly consisted of development

and implementation of analyses on mink and determination of normal values.

A common factor for the parts of the project included in the present report has therefore been the high degree of development work. This is an illustration of the status of fur animal research when we are talking about areas outside the traditional subjects. In areas like nutrition, disease control, breeding and reproduction, methods and tools are available, as well as knowledge about the normal reactions and characteristics of the animals.
In new fields such as environment, behaviour and production, hair and skins as well as health control, tools and methods were sparsely developed. Knowledge of the normal condition of the animals and the variations herein was therefore sparsely or not at all documented.

Besides leading to a number of other projects, this project gave the following main results and effects:

1. Behavioural research within the fur animal field started early enough for the debate regarding the welfare of fur animals to be based on facts instead of assumptions. The results of these investigations have been used extensively as documentation in discussions and legislation in this field. The team responsible for this part of the research is today internationally well recognized when fur animal welfare is discussed.

2. The management and environment section has clearly revealed important areas to be covered and cleared up a number of "myths" as regards mink production, (screening of females, watering conditions, kit behaviour before weaning, light conditions etc). The results of these investigations were also included in the above mentioned work of elucidation and legislation.

3. The genetic examinations related to pelt properties and blood parameters have contributed considerably to the understanding of the importance of objective criteria in the breeding work. The results have also confirmed that genetics play an important role in the variations between mink farms (mink populations).

4. The haematological and clinical-chemical section of the project has helped to

develop and strengthen this very important tool as an aid in research as well as in health control.

It has not been possible to establish a complete check-up system for health control - if such a system can at all be found.

An important result of the efforts of the department in this area has been that in cooperation with The Soviet Union and Finland, and edited by Asbjørn Brandt, we have been able to publish a book of 150 pages entitled: "Haematology and Clinical Chemistry of Fur Animals - A Current Treatise (SCIENTIFUR, 1989).

5. The subproject concerning mineral substances revealed considerable variation in the mineral content of feed and illustrated in which cases the mineral content in hair depended on the feed. Furthermore, correlations between mineral content in the hair and pelt characteristics were illustrated.

6. The establishment of hair morphological research in connection with this project has made it possible to start up basic research in this area. In fur animals as well as in rabbits it has proved to be an important tool in the research as well as in the breeding work. The immediate result of this project has been the description of genetic differences in hair types from scanblack mink.

In the areas mentioned, the project has given considerable contributions as regards practical methods and actual experimental results. As the weighing of method development and applications has varied between the three different parts of the project, it is difficult to make a combined discussion about all subprojects. The broad aim of the project has also made it difficult to get an overall view or to make a common conclusion.

Discussion as well as summaries and conclusions have therefore been given within each of the 4 main sections of the report. Besides, we refer to the more than 70 publications incl. papers at congresses and meetings, on which this report is based.

The project has been carried out in cooperation with Bioteknisk Institut, The Government Veterinary Serum Laboratory, The University of Copenhagen, dept.

of Population Biology, and The Danish Fur Breeders Association. Besides, we have been in contact with a number of institutions and persons in Denmark and abroad.

All technical employees of the department on the farm as well as in our laboratories have taken part in this project.

The manuscript has been typed by Hanne Artved and Dorthe V. Nielsen. The major part of the editorial and lay out work has been done by Steen H. Møller.

The department wants to thank all collaborators and employees for a fine cooperation as regards the implementation of the project and the preparation of the report.

Foulum, December 1990

Gunnar Jørgensen

CONTENTS

1. MANAGEMENT AND ENVIRONMENT IN MINK PRODUCTION

by Steen Møller

Summary

In this part of the project "Management and Environment" experiments were conducted concerning light, temperature of drinking water, water supply, visual isolation, scent communication, reaction to repeated weighings, and the correlation between time of birth, litter size, weight development, body length, skin size, and pelt quality.

A method for evaluation of light conditions in mink sheds was developed and tested. This method measures the quantity of light in the shed in relation to the quantity of light outside, and the difference expresses the light reducing effect of the shed. The light reduction is connected with the number of light panels in the roof and the cleanliness of these panels. The method gives the same results under different weather conditions. The method is therefore found to be applicable in practice. The significance of the light reduction is discussed, and it is found that mink can be kept under very dark conditions without any negative consequences as regards production. Negative consequences are more likely to occur when moving animals between varying light conditions or when using artificial light.

The intake of 40°C warm water by mink in comparison to water from the tap has been examined with adult males, with kits, and with pregnant and nursing females. In all cases the warm water was drunk in the same quantities as the cold water. If the adult males were offered one temperature at a time, they drank more warm water. If both temperatures were offered at the same time, they drank the same quantities, or in one case more cold water. The mink drank more frequently but less per session of cold water if both temperatures were available. If there was only one temperature at a time, no difference in the intake pattern was seen. In all cases a higher quantity of cold water was spilled, both totally and per drinking session. Mink kits drank equal amounts of warm and cold water, but wasted

more cold water. Before giving birth the females preferred warm water, but after birth they drank mostly cold water. The physiological significance of the temperature of drinking water is discussed. It is concluded that mink like to drink water up to 40°C. The waste of water is lowest if the water is warm.

In the latter part of the lactation period a supplement to the ordinary supply of drinking water is often used. A drip watering system was tested throughout two lactation periods, and the weight development, drinking behaviour and activity of the animals were recorded. In the cold and wet lactation period of the first year the system had no effect. In the warm and dry lactation period of the second year, the weight loss of the females was reduced, and the weight gain of the kits was faster in the group with drip watering system. Kits with drip water supply take in water earlier than the control group, by licking water from the tongue of the valve. However, they do not learn to release the valve earlier. Saliva licking occurred most frequently in the group without drip water supply. A difference of approx 2 weeks was observed from the kits start eating and until they start drinking. It is concluded that the opportunity of the kits to take in water early improves with drip watering. In warm and dry lactation periods this will increase gain and reduce the stress on the females.

Previous experiments with visual isolation of mink females in the gestation and lactation periods have indicated a positive effect on the whelping result. The experimental groups have, however, been too small to demonstrate any reliable differences in reproduction results. Therefore, four experiments with visual screening of mink females were carried out. The females were isolated before mating or before whelping. The separation consisted of an empty cage or a cage filled with straw. Whelping results, weight development, and activity were recorded. No systematic differences in whelping results were found that could be related to the screening, and kit gain was not affected. The females in the control group were more active than the separated groups. The whelping results are discussed. They conflict with previous results, but are confirmed by later investigations. It is concluded that visual isolation of mink females does not influence whelping result or kit gain but reduces the activity of the females in the lactation period.

The significance of scent communication between males and females was investigated by spraying male urine on the cages of the females before mating. The period of latency from the time animals are put together until mating starts, was 4 minutes shorter in the group stimulated by scent, but no difference was found in mating success or whelping result. It is concluded that scent stimulation is of no practical importance as regards the way in which males and females are placed before mating.

The reaction of mink kits to repeated weighings was examined by means of a stick test on 17 farms distributed all over Denmark. In general, the weighed animals reacted more timidly than the not-weighed animals. Females reacted in general more timidly than males. On a few farms the weighed animals were less timid, which shows that handling can be regarded as a positive experience.

The correlation between weight development, body length, skin size, and pelt quality was examined on scanblack males. Already at the age of 9 days weight is correlated to body length, skin length and weight at pelting. After weaning, the correlations are increasing until pelting. The correlations between size measurements and quality of the skins show that weight is responsible for the decrease in quality, whereas body length plays a secondary role. It is concluded that a long skin from a long mink is of better quality than a long skin from a fat mink. The body length can therefore with advantage be included in the breeding work, instead of body weight.

Body length and weight development were examined in relation to time of birth and litter size. The importance of time of birth decreased quickly and had almost been balanced out in August-September. Litter size was significant for both body length and weight all the time until pelting, and the biggest kits came from litters of 3-7 kits. Kits from large litters do catch up somewhat in size after weaning, whereas kits from small litters have no compensatory gain.

Sammendrag

Under delprojektet "Management og Miljø" er der gennemført undersøgelser vedrørende lysforhold, temperatur af drikkevand, vandforsyning, visuel adskillelse, duftkommunikation, betydning af gentagne vejninger og sammenhænge mellem fødselstidspunkt, kuldstørrelse, vægtudvikling, kropslængde, skindlængde og kvalitet.

En metode til vurdering af lysforholdene i minkhaller blev udviklet og afprøvet. Ved metoden måles lysstyrken i hallen i forhold til udenfor, og forskellen tages som udtryk for hallens dæmpende effekt. Lysdæmpningen står i relation til antallet af lysplader i taget og pladernes renhed. Metoden giver samme resultater under forskellige vejrforhold, og den skønnes derfor anvendelig i praksis. Lysdæmpningens betydning diskuteres, og det skønnes, at mink kan holdes under meget mørke forhold uden negative produktionsmæssige konsekvenser. Disse kan snarere forventes ved flytning af dyr mellem forskellige lysforhold, eller ved brug af kunstigt lys.

Minkens optagelse af 40°C varmt drikkevand i forhold til vand fra hanen er undersøgt med udvoksede hanner, med hvalpe samt med drægtige og diegivende tæver. I alle tilfælde blev det varme vand drukket i samme mængder som det kolde vand. Hvis de voksne hanner blev tilbudt en temperatur ad gangen, drak de mest varmt vand. Hvis begge temperaturer blev givet samtidig, drak de lige meget eller, i et tilfælde, mest koldt vand. Minkene drak hyppigere men mindre per gang af koldt vand, hvis begge temperaturer var tilgængelige. Hvis der kun var en temperatur ad gangen, var der ingen forskel i optagelsesmønsteret. Der blev i alle tilfælde spildt mest koldt vand, både i alt og per drikkesekvens. Minkhvalpe drak lige meget varmt og koldt vand, men spildte mest koldt vand. Indtil fødsel foretrak tæverne varmt vand, men efter fødslen drak de mest koldt vand. Den fysiologiske betydning af drikkevandets temperatur diskuteres. Det konkluderes, at mink gerne drikker vand op til 40°C, og at vandspildet er mindst, hvis vandet er varmt.

I sidste del af diegivningsperioden anvendes ofte et supplement til den almindelige drikkevandsforsyning. Et drypvandingssystem blev afprøvet gennem to diegivningsperioder, og dyrenes

vægtudvikling, drikkeadfærd og aktivitet blev registreret. I det første års kolde og fugtige dieperiode havde systemet ingen effekt. I andet års varme og tørre dieperiode var tævernes vægttab reduceret og hvalpenes vægtudvikling hurtigere i holdet med drypvand. Hvalpe med drypvand optager vand tidligere ved at slikke vand fra ventilens læbe, men de lærer ikke at udløse ventilen tidligere end kontrolholdet. Spytslikning forekom hyppigst i holdet uden drypvand. Der blev observeret en forskel på ca. to uger, fra hvalpene begynder at æde, til de begynder at drikke. Det konkluderes, at hvalpenes mulighed for tidligt at optage vand forbedres ved drypvand. I varme og tørre dieperioder vil dette forøge tilvæksten og reducere belastningen af tæven.

Visuel adskillelse af minktæver i drægtigheds- og diegivningsperioden har ved tidligere forsøg med meget få tæver tydet på en positiv effekt på hvalperesultatet. Der blev derfor gennemført fire forsøg med visuel adskillelse af minktæver. Tæverne blev isoleret før parring eller før hvalpning, og adskillelsen bestod af et tomt eller et halmfyldt bur. Hvalperesultater, vægtudvikling og aktivitet blev registreret. Der var ingen systematiske forskelle i hvalperesultaterne, der kunne henføres til adskillelsen, og hvalpenes tilvækst var ikke påvirket. Tæverne i kontrolholdet var mere aktive end i de adskilte grupper. Baggrunden for hvalperesultaterne diskuteres. De er i strid med tidligere resultater, men bekræftes af senere undersøgelser. Det konkluderes, at visuel isolation af minktæver ikke har betydning for hvalperesultat eller hvalpenes tilvækst, men reducerer tævernes aktivitet.

Betydningen af duftkommunikation mellem hanner og tæver blev undersøgt ved at sprøjte urin fra hanner på tævernes bur forud for parring. Latenstiden fra dyrene sættes sammen, til parringen begynder, var 4 minutter kortere i den duftstimulerede gruppe, men der var ingen forskel i parringssucces eller hvalperesultat. Det konkluderes, at duftstimulering ikke har nogen praktisk betydning med hensyn til, hvordan hanner og tæver skal gå i forhold til hinanden op til parring.

Minkhvalpenes reaktion på gentagne vejninger blev undersøgt ved en pindetest på 17 farme fordelt over hele Danmark. De vejede dyr reagerede generelt mere frygtsomt end ikke vejede, og tæver reagerede generelt mere

frygtsomt end hanner. På enkelte farme var de vejede dyr mindst frygtsomme, hvilket viser, at håndteringen kan opfattes som noget positivt.

Sammenhængen mellem vægtudvikling, kropslængde, skindstørrelse og pelskvalitet blev undersøgt på scanblack hanner. Allerede ved 9 dage er vægten korreleret til kropslængden, skindlængden og vægten ved pelsning. Efter fravænning er korrelationerne stigende frem til pelsning. Sammenhængene mellem størrelsesmålene og kvaliteten af skindene viser, at vægten er ansvarlig for den faldende kvalitet, mens kropslængden spiller en underordnet rolle. Det konkluderes derfor, at et langt skind fra en lang mink er af bedre kvalitet end et langt skind fra en fed mink. Kropslængden vil derfor med fordel kunne inddrages i avlsarbejdet i stedet for vægten.

Kropslængde og vægtudvikling blev undersøgt i relation til fødselstidspunkt og kuldstørrelse. Fødselstidspunktets betydning aftog hurtigt og var stort set udlignet i august-september. Kuldstørrelsen havde betydning for både kropslængde og vægt helt frem til pelsning, og de største hvalpe kom fra kuld på 3 til 7 hvalpe. Hvalpe fra store kuld indhenter noget i størrelse efter fravænning, mens hvalpe fra små kuld ikke har nogen kompensatorisk vækst.

1.1 Background

The purpose of the management and environment section of the project was to describe the variation in physical farm conditions and management on the participating mink farms. Based on an evaluation of the importance of these differences in relation to production, individual items were chosen for experimental investigation, either on a research farm or in practice.

Furthermore an examination of routines for management and control of the production was included as an essential element in the project.

As the project has been split up into many subjects and sections, results and discussion will be divided into paragraphs on each subject.

1.2 General farm conditions and management.

The physical frames of fur animal production are standardized to a very high degree in Denmark. Within the field of sheds and cages a few types are totally dominant on the market. This is primarily due to a very rational standard of the buildings, where farmers - contrary to other countries - buy readymade sheds instead of developing and building the sheds themselves. The design is based on a well operating type developed on basis of practical experiences. On the other hand the many different housing conditions used in mink farming around the world reflect that mink are very robust with regard to housing in general.

The type of shed most often used is the open double-row shed with an "Eternit" asbestos roof. Another much used type is the closed multi-row shed. This type exists in many different designs, but most often it consists of 6 rows and is mainly used for breeding animals during the winter. Afterwards the kits are placed in double-row sheds at weaning. The construction of the sheds is divided into 2.00 m wide sections, on which the cage sections are suspended.

The cage sections consist of either 6 or 8 cages. Typically 6-room cages measure 36 x 12 x 18 inches (approx 90 x 30 x 45 cm) (l x w x h) and are used for breeding, birth, lactation and kits after weaning. The 8-room cages measure 36 x 8 x 18 inches (approx 90 x 20 x 45 cm) and are used for breeding animals kept individually. Each cage has a nest box, placed outside the

cage of the same width as the cage, 8-10 inches (20-25 cm) long and 6-8 inches (15-20 cm) deep. Furthermore, there are cages with the nesters placed in the upper part of the cage. These cages are often shorter but taller than the standard 6- and 8-room cages. Design of cages and nest boxes has been described in "Guidelines of fur animal production" prepared and recommended by the Fur Breeders' Associations of the four Nordic countries. These common Nordic regulations which also deal with feed and water, hygiene, health, transportation, and killing, express the wish for a common, justifiable standard of fur animal production in the countries selling under the Saga label.

During the whelping period most breeders place a drop-in bottom in the nest box. The purpose is to help the female build the nest and to prevent the kits from getting lost in the corners. The drop-in bottom varies from a wooden plate and a brick to a moulded plastic or a hollowed wooden bottom with windscreen.

Fresh feed is delivered every day from feed kitchens. The feed is placed on top of the cage and the mink eat the wet feed through the wire netting.

The watering system consists of a hose running along the backside of the cages. The mink get water by releasing a drinking valve and sucking water. The valves are of different makes, but only details differ. An older system consisted of a cup with free water, but this type is not in use very much any more. The water hoses may be either insulated, screened or uninsulated, and the water may be circulating and/or heated in order to secure the water supply in frosty weather.

Management can be divided into the various production periods throughout the year.

The main impression from the project is that there is not only one correct way of doing things. What is important is that all jobs are carried out at the right time which means that many different systems may very well work perfectly. The most important thing is that the farmer has a system which suits him and which he knows will work.

The farms can be divided into different types or groups according to their way of handling each production period. This division is to a high degree geographical and is presumably the result of the local advisory ser-

vice and of communication with colleagues where farmers learn how things are done on other farms. The same goes for special cage and watering systems like for instance the use of nester cages and circulating watering systems.

The physical farm conditions and management on the project farms are described in a later Report from the Natl. Inst. of Anim. Sci.

Physical farm conditions as well as management have been the objects of experimental investigations. These have been published currently and will be presented in short here, to obtain a total presentation of the results of the project.

The following investigations are presented in this chapter:

Investigations of physical farm conditions:

1.3 Light conditions in mink sheds.
1.4 Drinking water temperature.
1.5 Supplementary watering systems.

Investigations of management conditions:

1.6 Visual isolation of females.
1.7 Scent communication in the mating season.
1.8 Weighing and behavioural response.
1.9 Weight development, body length, size and quality of fur.
1.10 Development of body length and weight in relation to time of birth and litter size.

1.3 Light conditions in mink sheds

1.3.1 Introduction

The time of fur development and the reproduction cycle of the mink are primarily controlled by light conditions.

Mink breeders therefore have a natural interest in being familiar with these and the importance of the changes caused by different housing arrangements in relation to normal daylight.

The objective of almost all light experiments has been to change the production towards earlier fur development (e.g. Adair & Stout, 1972) or to induce more heat cycles per year (e.g. Aulerich et al., 1963; Williams & Turbak, 1970; Reiten, 1977).

Only a few investigations deal with the conditions of daylight and/or artificial light required to maintain a normal production (Travis et al., 1971).

Poor light conditions have often been suspected to be the reason for problems, but there has been no measuring method or references which could help evaluate when problems might arise. On this background a measuring method has been developed and tested on the project farms.

1.3.2 Material and methods

This method measures the strength of the light in light value (lw) by means of a Gossen lunalite photometer. The light is measured at three places in multi-row sheds. The quantity of light is measured outside the shed before and after the measurements inside. The difference between the averages of measurements outside and inside is a direct expression of the light reducing effect of the shed.

1.3.3 Results and discussion

The method was found to be rather robust with regard to conditions at the time of measurement, i.e. weather, time of year and time of day. However, there should be no snow etc. on the roof.

The light reduction corresponds rather well to the first impression. The differences in the reduction are connected with the amount of light panels in the roof, windows in the walls, the cleanliness of the light panels and windows, and the strength of the light outside the shed. In two-row sheds light conditions are much better in the cages than in the middle, as light falls in through the side of the sheds.

The quantity of light in normal daylight varies from approx 8-14 lw. The reduction in the sheds measured varied from 2.8-5.6 lw with no detectable problems with the production caused by the light conditions. This is not surprising, as the mink respond to very low intensities of light (Travis et al., 1971). Moving mink between sheds with different light conditions might cause trouble with pelt priming and reproduction, as a shift in light conditions can alter the actual day length and thereby the production of light regulated hormones in mink.

1.3.4 Conclusion

It is concluded that the measuring method is applicable for

determination of light conditions in mink sheds. Within the limits of measured reduction there is no detectable effect on the production, and a reduction of up to 6 lw is therefore found acceptable. Light panels covering 10-15% of the roof give a satisfactory amount of light, if they are kept clean. Moving mink from good to poor light conditions or vice versa, may effect the production by altering the day length experienced by the mink. Light problems in double-row houses are regarded as very rare, as the light falls in through the side of the shed.

The results have been presented and published at the annual meeting of the Natl. Inst. of Anim. Sci. (Møller, 1989).

1.4 Drinking water temperature

1.4.1 Introduction

In Denmark, watering systems for mink consist almost exclusively of automatic equipment where a drinking valve at the rear end of the cage is screwed into a water hose running along the cage rows. Traditionally the water hose is a black 3/4" plastic hose. In sunlit hoses the water will quickly be heated up to 45°C or more, whereas in frosty weather it will quickly freeze. Various methods have been developed to remedy these problems.

The water in the hoses may be circulating and the hoses may be screened or insulated. Often heating possibilities are installed in the hoses and/or the circulating water.

While the water intake of the mink stops suddenly when the water freezes, no knowledge existed regarding the water intake of the mink when the water temperature was rising. A number of experiments for clarification of this problem have been carried out.

1.4.2 Material and methods

In all experiments the temperature of the warm water was 40°C, whereas the cold water from the tap varied from 6-17°C.

Experiments have been carried out according to two different methods. In one method the animals were offered both temperatures for 8 days. The animals could thus choose between two temperatures at the same time. The drinking nipples with the two temperatures were switched around at certain intervals in order to avoid habituation to

placement. In the other method the animals were offered the two temperatures alternately every second day, offering each temperature for 4 days.

The experiment ran over 5 periods of approx 8 days. In periods 1, 3 and 5 warm and cold water was offered according to the two methods. In periods 2 and 4 only warm and cold water, respectively, was offered.

Experiments have been carried out with adult males in February-March, with pairs of male and female kits from weaning, and with lactating females.

1.4.3 Results

1.4.3.1 Males

The adult males were given cold water of 6°C and warm water of 40°C according to both methods. In both methods the waste of cold water was significantly higher than the waste of warm water (tables 1.1 a and b). In the method with changing temperatures a higher amount of warm water was drunk in all three periods ($P<0.01$). At the method with both temperatures equal amounts of warm and cold water were drunk in periods 1 and 3. In period 5 a significant preference for cold drinking water was registered. The results of the habituation periods are summarized in table 1.1c. With great certainty it can be said that in both methods warm water was preferred ($P<0.001$).

The total water intake amounted to approx 90 ml/day except for the periods with only cold water, where smaller amounts were drunk. The individual variation between mink is considerable with regard to water intake, waste of water and temperature preference.

Table 1.1 Grams of water at 40°C or 6°C ingested and wasted by a group of five male mink.

a. One temperature at a time, changing from day to day. Each temperature offered for 4 days.

Period	g of water	40°C	6°C	P<
1	Wasted	50	178	0.01
	Ingested	103	81	0.001
3	Wasted	55	122	0.001
	Ingested	121	87	0.01
5	Wasted	56	250	0.001
	Ingested	121	69	0.001

b. Both temperatures offered simultaneously for 8 days.

Period	g of water	40°C	6°C	P<
1	Wasted	26	172	0.001
	Ingested	53	54	ns
3	Wasted	18	220	0.001
	Ingested	49	49	ns
5	Wasted	15	189	0.001
	Ingested	38	55	0.001

c. Each temperature offered for 7 days between experimental periods 1, 3 and 5.

Method	g of water	Period 2 40°C	Period 4 6°C	P<
Changing	Wasted	30	261	0.001
	Ingested	93	76	0.01
Both	Wasted	51	256	0.001
	Ingested	89	71	0.001

A video recording of the experiment with males was carried out in order to examine activity and drinking behaviour of the two methods and temperatures. The results appear from table 1.2.

Table 1.2 **Activity and behaviour of male mink in relation to intake and waste of water at 40°C or 6°C. Registered 24 hours with each temperature.**

	Temperatures offered simultaneously			Changing	
Behaviour	6°C	40°C	Total	6°C	40°C
Drinking sessions	18	7	25	28	30
g ingested	41	37	78	79	81
g wasted	138	11	149	230	35
Ingested g/ session	2.4	5.1	-	2.9	2.8
Wasted g/ session	7.9	1.4	-	8.4	1.2
Hours of sleep			18	17	17

The table shows that there was no difference in water intake on the days with video recording. Nevertheless, there was a difference in drinking behaviour at the two temperatures. Each time the mink drink, they take in twice as much warm as cold water if both temperatures are offered. There is no difference when only one temperature is offered. Both methods showed that the waste of cold water was 5 times as high per drinking session as the waste of warm water.

In both experimental methods the mink drank 25-30 times and took in approx 80 ml of water altogether. The mink ate 6-12 times per 24 hours followed by 1-2 drinking sessions and 1-2 hours of sleep. One of the mink played with the cold water valve but the difference in waste of water applied to all 4 mink.

If the results before and after a period of habituation with only one water temperature, or the start and ending of such a period, are analysed, the results will give an idea of the habituation to water temperature. The results before and after an exchange of temperatures will give an indication of habituation to placement.

The waste and intake of cold water (6°C) are not affected by habituation in the males. Apparently habituation has a small influence on waste and intake of warm water (40°C).

After 8 days with warm water, the intake of warm water is higher than before the period ($P<0.05$) when both temperatures are offered. When the temperatures are offered alternately, the same tendency towards increased intake is found ($P=0.12$), and the waste of warm water has also increased ($P=0.06$).

Eight days with cold water did not influence waste or intake of water regardless of temperature at the method with both temperatures offered simultaneously. The method with changing water temperatures showed a tendency towards a reduction of the intake of cold water after the 8 days ($P=0.13$).

From the start to the end of the 8 day period with warm water there was a tendency towards a reduction in both intake and waste of water, when both temperatures were offered ($P=0.17$) and similar for the intake when temperatures were changing ($P<0.05$). In none of the methods were there any differences in water intake or waste at the beginning or the end of the period with cold water.

The waste of water was independent of the placement of the drinking valves, whereas the water intake showed a moderate tendency towards changing after the exchange of the valves and therefore a possible habituation to placement ($P=0.14$).

1.4.3.2 Kits

Mink kits were offered water of 40°C and 17°C simultaneously.

Water intake per pair of kits increased by approx 5 ml daily during the experimental period (figure 1.1). The statistical processing showed no difference in water intake at the two temperatures. There was a significant difference in water intake between the five pairs of kits ($P<0.01$).

The water waste also increased a little with age, and the increase was largest for cold water. In the latter part of the experiment significantly more cold than warm water was wasted ($P<0.001$). There was also a significant difference in water waste between the 5 pairs of kits.

There seemed to be an effect of habituation of periods 2 and 4 with regard to waste of water.

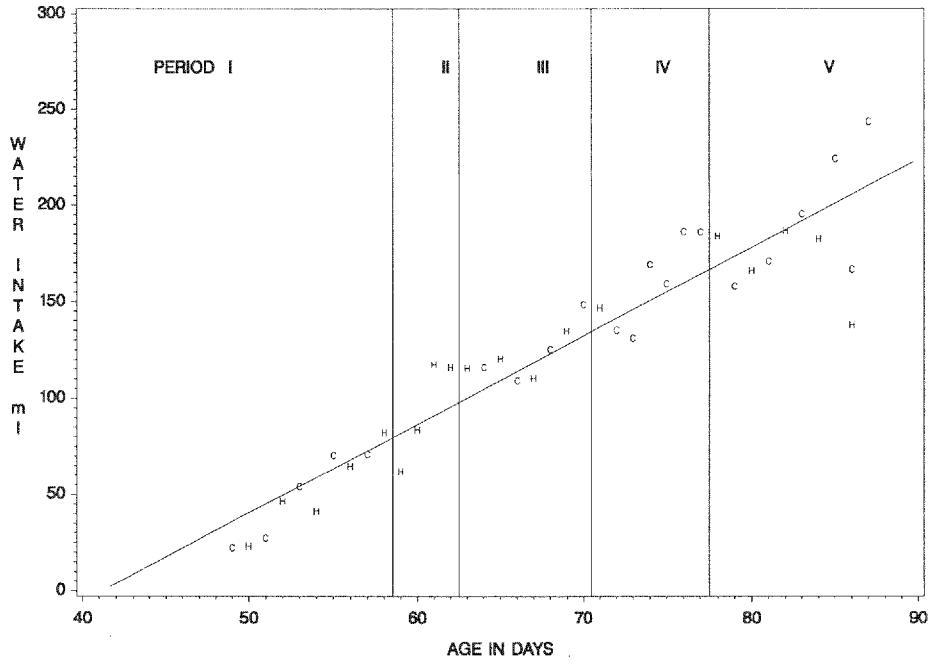

Figure 1.1 **Daily intake of 40°C (H) and 17°C (C) drinking water by 5 pairs of mink kits after weaning. Periods I, III and V include both temperatures simultaneously, period II with warm and IV with cold water.**

1.4.3.3 Females

In the experiment with females in the latter part of the gestation and lactation periods water of 40°C and 8-15°C was offered simultaneously during the entire experiment. This is due to the fact that differences in time of birth and number of kits makes an experimental plan with the previously used 5 periods impossible.

The whelping result was poor, and many kits were lost during the experiment. This cannot be caused by the water temperature, as the females could still drink cold water as they were used to. The experiment with the females started on April 27th, the first female gave birth on April 30th and the last on May 7th. On average the females had 6 live kits and 0.5 stillborn. Within the first week the females lost 4 of the 6 kits which must be a result of the transfer to the unknown environment indoors. Therefore the experiment will have to be repeated in the normal farm environment before any final conclusions can be drawn.

Due to the low number of kits and the many deaths it is impossible to calculate water consumption per kit or per g of kit gain.

Two females had 3 and 4 kits, respectively, during the entire experimental period. The results of these two females are illustrated in figure 1.2. It appears that the total water intake decreases until birth in order to increase again until weaning. Until the time of birth the females drank mostly warm water, but after the birth mostly cold water was drunk. The waste of water followed the curve of water intake, and after birth almost exclusively cold water was wasted (figure 1.3).

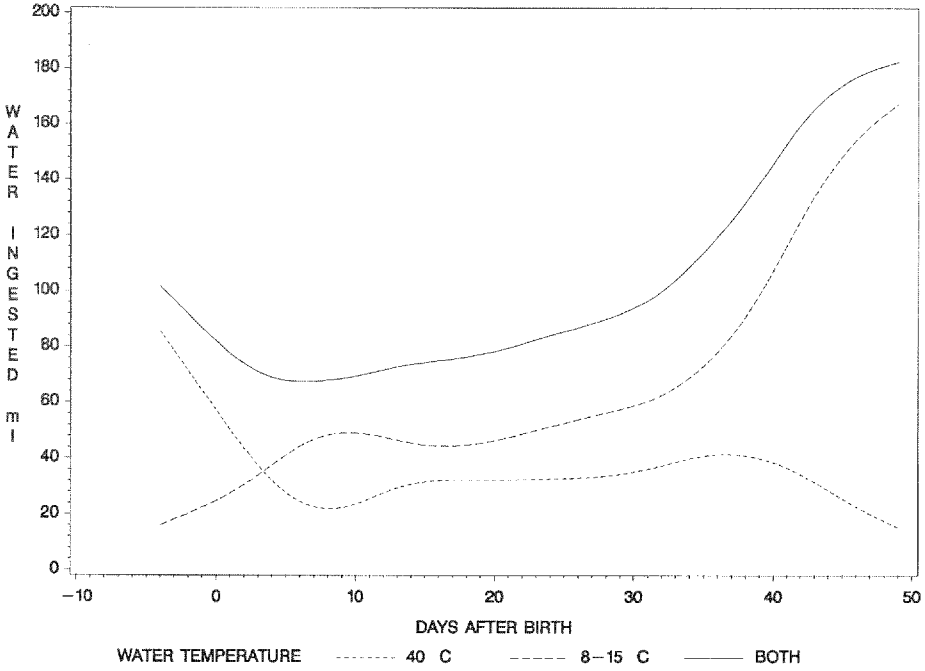

Figure 1.2 **Daily intake of 40°C and 8-15°C drinking water from 4 days before till 7 weeks after birth. Two females with 3 and 4 kits.**

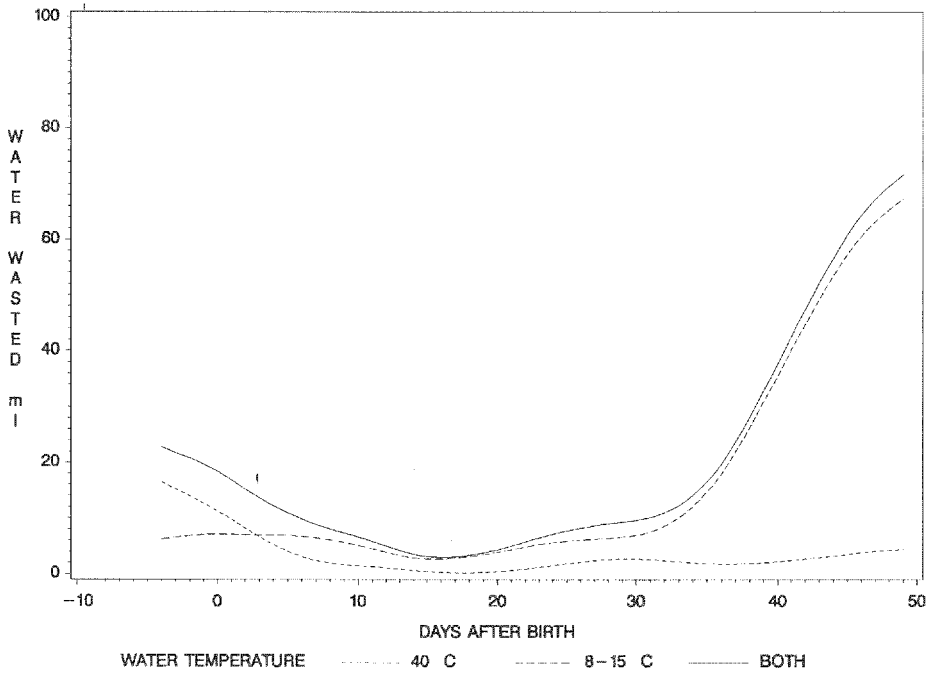

Figure 1.3 **Daily waste of 40°C and 8-15°C drinking water from 4 days before till 7 weeks after birth. Two females with 3 and 4 kits.**

1.4.4 Discussion

Both test methods work well and the choice of method must be based on the actual investigation. The method with both temperatures is a logical way to determine preferences, while the method with changing temperatures is realistic in relation to practice, where only one temperature is available at the time.

Physiologically it is understandable that mink like to drink 40°C warm water. Cold water is only let out of the stomach slowly, and the stomach will be

filled quickly. The feeling of thirst is thereby put out without removing the physiological reason for thirst (Deaux, 1973). Drinking water at body temperature passes through the stomach, allowing a greater volume to be ingested before stomach distention signals a satiation of thirst. Blood tests in rats have shown that drinking water at body temperature lead to a faster decrease in serum osmolarity than cold water (Deaux, 1973). Warm water is therefore a better thirst quencher, as it is absorbed more quickly in the body, and a higher amount is absorbed before the feeling of thirst is put out. This may be the explanation why mink prefer warm water, if the cold water is only 6°C, whereas there is no difference if it is over approx 10°C.

Waste of water is in all cases significantly higher for cold water. Of course there is a correlation between the intake and the waste, but also in relation to intake there is always more waste of cold water. There seems to be an insignificant habituation to water of unknown temperature, i.e. 40°C for adult mink, but also to cold water as far as the kits are concerned.

1.4.5 Conclusion

On basis of the experiments performed, it can be concluded that mink do not refrain from drinking 40°C hot water. In some cases the warm water is preferred, but in most cases no preference is noted. In a single experimental period cold water was preferred. It is still too early to say precisely, whether the difference in results is due to a difference in sex and age of the animals, in time of year and/or in differences in ambient temperature or temperature of the "cold" water. Waste of water is in all cases significantly higher for cold water than for warm water.

The results of the water temperature experiments were presented at the Annual Meeting of the Natl. Inst. of Anim. Sci., and at the 4th Int. Sci. Congr. in Fur Animal Prod., Toronto, as well as in the Danish Fur Breeders Journal (Møller, 1986, 1987, 1988b, 1988c; Møller & Lohi, 1989b).

1.5 Supplementary watering system

1.5.1 Introduction

Mink kits start eating and drinking in the latter part of the lactation period. In the same period loss of weight and dehydration of some females are seen, especially in hot summers. Both kits and females can therefore be relieved if the kits start to drink as soon as possible. Mink have no natural qualifications for drinking from a valve. Various devices have been developed to help the kits find the drinking valve and learn to drink. One type is a clamp intended to help the kits release the valve more easily. Another principle is valves supplied with a tongue which can either be tilted so that water will stay on the tongue or it can be equipped with drip watering to secure that there is water on the tongue all the time. A clamp with a bowl combines these two ideas.

1.5.2 Material and methods

During two lactation periods experiments have been carried out with a drip watering system. The drip watering equipment was turned on on May 3rd and set at 25-40 drips per minute to secure maximum effect of the system. Among 60 scanblack females with drip watering and 60 females in the control group, 15 females with 4-8 kits were chosen for weighing.

The animals were weighed at the age of 10, 20, 30, 40 and 50 days. The first year the kits were distributed at weaning with 60 pairs (male + female) in the drip watering system and 60 pairs in the control group. They were weighed every fortnight until August 26th when the experiment ended.

Activity and drinking behaviour of the kits were registered 6 times with short observations in two daily periods from June 9th until weaning. Until June 9th, the animals were observed once a day, and registrations started when the first kits became active near the drinking valve.

In the first year it was registered whether the kits were out in the cage, active, in which part of the cage compared to the drinking valve, and whether they were drinking or trying to drink. In the second year the observations were concentrated on the actual drinking behaviour and specifying whether the kits got water and whether they released the valve themselves. Further-

more, saliva licking from the corner of the female's mouth was observed.

1.5.3 Results

The results of the first year experiments showed no effect on weight development of females or of kits. Neither was the development of kits after weaning affected by the watering system. Behavioural observations showed that the kits with drip watering system had significantly less unsuccessful attempts to drink ($P<0.05$) and more attempts tended to be successful. There were no other differences in behaviour between systems.

The results from the second year showed that the females in the drip watering group lost less weight than the control group ($P<0.05$). The weights are shown in table 1.3, from which it appears that the difference was most significant around June 10th.

Table 1.3 **Weight of female mink with normal and drip watering system. Fifteen females weighed five times between birth and weaning.**

		date				
		11/5	20/5	30/5	10/6	21/6
Group	n	weight g ± sd				
Control	15	1026± 79	1006± 89	986± 72	889± 67	848± 63
Dripwater	15	1156±130	1146±114	1113±121	1054±109	980±127

Table 1.4 **Weight of male and female kits with normal and drip watering system. Kits weighed five times between birth and weaning.**

		date				
		11/5	20/5	30/5	10/6	21/6
Group	n	weight g ± sd				
Males						
Control	42	48±12	99±18	176±25	295±53	505±75
Dripwater	47	45± 9	98±15	185±23	294±42	541±78
Females						
Control	37	42±9	91±16	157±27	263±57	430±93
Dripwater	40	43±9	95±17	170±19	263±33	451±58

As far as the gain of the kits (table 1.4) is concerned there was a clear effect of drip watering ($P<0.001$). The effect started showing already between the age of 20 and 30 days and increased until weaning. As the kits in the experimental group are born one day later than the kits in the control group, the difference is actually larger than shown in tables 1.3 and 1.4.

Behavioural observations showed that kits with drip watering start sucking water from the age of 40 days, whereas kits without drip watering rarely have this opportunity. On the contrary more kits in the control group find the valve without getting any water from it.

All in all the kits with drip watering start taking in water earlier no matter whether they release the nipple or not which is shown in figure 1.4. This is due to the fact that they suck the dripping water. They do not learn to release the valve earlier than the control group. Saliva licking is significantly more common in the control group than in the drip watering group already from the age of 35 days as shown in figure 1.5. The water temperature in the hoses was 19.6 ± 4.0°C in the drip watering system and 20.4 ± 5.6°C in the control row. The difference was significant ($P<0.05$).

Observations of eating and drinking behaviour showed that

the kits start eating at the age of 4 weeks but only start drinking approx 14 days later. These terms have been confirmed by observing the time of eating and drinking during the whelping period 1989 on the research farm. Of 272 litters observed, 3.3% were seen eating before 4 weeks of age, whereas 73.9% started eating in their 5th week, between 28 and 35 days of age. The first time drinking was only registered in 42 litters. Of these 81% started drinking in the 7th week from 42 to 48 days, whereas 7.1% (3 litters) started the week before.

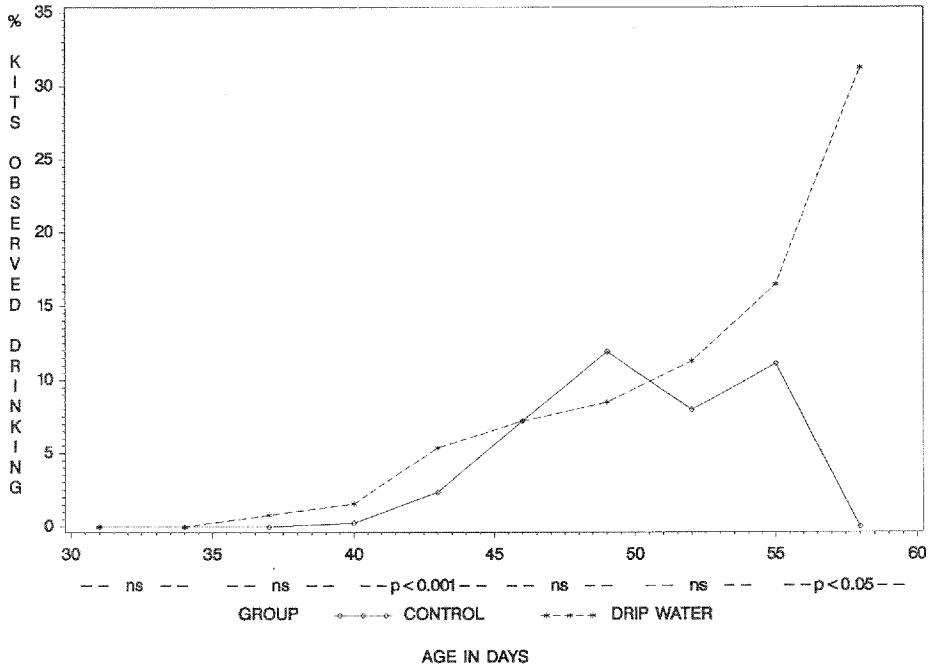

Figure 1.4 Development of drinking behavior of mink kits with and without drip watering system. Percentage of kits observed drinking, releasing the nipple or not.

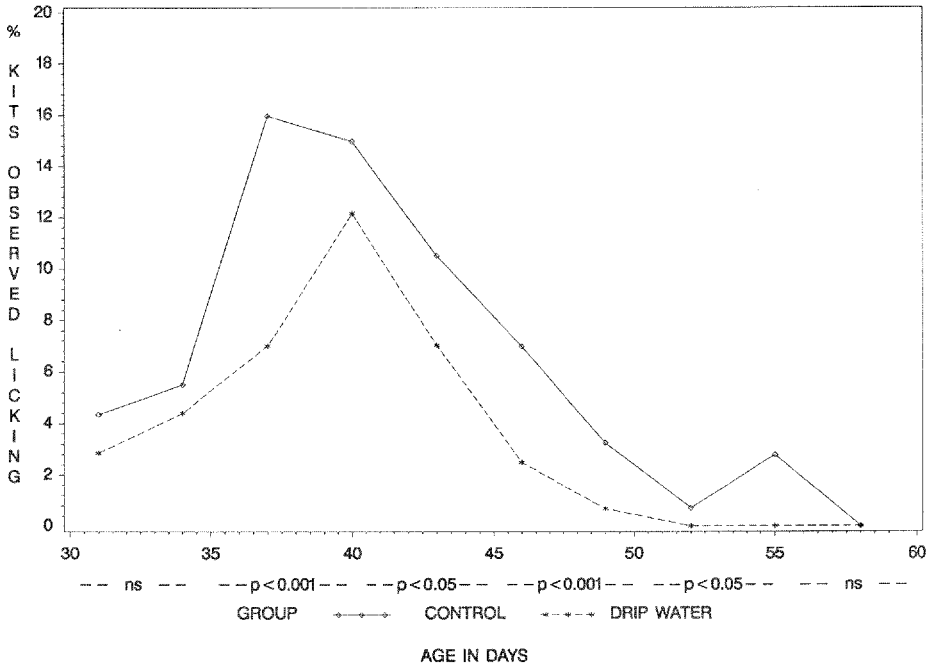

Figure 1.5 Development of saliva licking in mink kits with and without drip watering system. Percentage of kits observed licking saliva.

1.5.4 Discussion

The difference between the results of the two years is presumably due to the large difference in weather conditions during the lactation periods. In 1987 it was very cold with medium temperatures of 9.1°C against normally 11.2°C. The maximum temperature during daytime was around 15°C. In 1988 it was hot and dry with a medium temperature of 11.9°C and temperatures above 20°C during daytime in the month of May.

Even though an analysis of covariance shows a difference in the weight development of females of the two groups, the weight curves run almost paral-

lelly, except at the weighing on June 10th. Females in moderate condition around birth have earlier proved to keep their weight best during the lactation period (Tauson & Aldén, 1985). It was therefore to be expected that the rather heavy females in the drip watering group would suffer a heavier weight loss towards the end of the lactation period. This is not the case, which is probably due to the drip watering system and confirms the effect showed by the analysis of co-variance.

The weight of the kits is already differing at the age of 30 days. The gain has thus been quicker in the drip watering group, before the kits have started eating solid feed at the age of approx 4 weeks. This seems to indicate that the female somehow benefits from the drip watering system and passes this on to the kits. As the drinking behaviour of the females has not been investigated, the reason cannot be explained.

The improved weight development of the majority of the females as well as of the kits can be explained by their drinking behaviour. As the kits take in water earlier and more frequently in the drip watering group, they take in more feed and become less dependent on the female. This is confirmed by the reduction in saliva licking. The difference in water temperature can be explained by an increased flow in the drip watering system. The difference of approx 1°C is, however, quite insignificant for the water intake as documented in paragraph 1.4 on water temperature experiments.

The difference of 2 weeks in the time when the kits start eating and drinking illustrates why this period is so critical for the female as well as for the kits. During this period the kits must cover their need for liquid through water from the feed, milk and the saliva licking from the female. During this period the milk production of the female starts to decrease, and nursing disease in the female and cannibalism among the kits are seen.

1.5.5 Conclusion

All in all, it can be concluded that the water intake of mink kits is improved with access to free water. In hot and dry weather during the lactation periods this will improve their weight gain and reduce the weight loss of the females. The female herself may also profit from auxiliary installations at the water valves. The drip watering

system which was tested helped the kits take in water at an earlier stage, but they did not learn to release the valve themselves before the kits in the control group. Kits only start taking in water when they are approx 35 days old, and the first of them can release the valve at the age of 40 days. When they start eating at the age of approx 4 weeks, there is a period of 1-2 weeks after they have started eating when they have not yet learned to drink. It is therefore important to do anything possible to help them during this period.

The results of the drip watering experiment have been published as a Short Comm. from the Natl. Inst. of Anim. Sci. and in the annual report of the Danish Fur Breeders Association (Møller & Lohi, 1988, 1989a). The importance of good management during the lactation period has been discussed in the Danish Fur Breeders Journal (Møller & Lohi, 1989b).

1.6 Visual isolation of mink females during the reproduction period.

1.6.1 Introduction

Wild mink are known to be solitary animals, and presumably in most cases the females are rearing the kits alone. It could therefore be expected that the close presence of other mink would influence mink in captivity, and in the reproduction period be the cause of inferior parental care. Previous investigations have found that a screening from neighbouring females may influence breeding result and kit gain favourably (Gilbert & Bailey, 1967, 1970; Hernesniemi, 1976; Vestergård, 19??a). These investigations are, however, 10-20 years old and were performed with only very few animals (10-20 females per group). An investigation with large groups of mink without plasmacytosis was therefore needed in order to clarify, if a favourable effect can be obtained.

1.6.2 Material and methods

The experiments were performed over two years with scanblack and scanbrown females on two private farms. In 1985 3 experiments were carried out. In experiment I 79 scanbrown females were isolated immediately before mating by means of an empty cage, whereas the control group of 70 females occupied all the cages. In experiment II 198 females were separated by means of an empty cage during the latter part of the gestation peri-

od, whereas the control group of 72 females occupied all the cages. In experiment III 58 scanblack females were separated by means of a cage filled with straw, whereas the control group of 60 females occupied all the cages. Experiment IV was a repetition in 1986 of experiment I from 1985. In this experiment 142 isolated and 144 non-isolated females were included, all of scanbrown type.

In 1985 information about date of birth, whelping result, loss of kits and barren females was procured from the EDP-breeding system of the Danish Fur Breeders Association. In Experiment I the number of unsuccessful matings was recorded. Kits from experiments I and II were weighed before weaning. The activity was registered as number of animals moving in the cage. In experiment I the animals were observed for 24 hours just before weaning.

In 1986 (experiment IV) information about date of birth, number of kits and barren females was recorded at farm visits. This was due to uncertainty in the EDP-breeding information in case of kits which had been moved and females which had more kits at 2 weeks than at birth. A careful registration on the farm was therefore necessary in all cases.

The activity of the females was registered on four mornings during the week after birth. The degree of nest building was evaluated according to a grading from 1-4. The degree of territory marking was evaluated according to a grading from 1-4 based on the size and shape of the mounds of manure.

1.6.3 Results

Breeding results from all four experiments are shown in table 1.5. In experiment III there were significantly more living kits per fertile female in the isolated group than in the control group. This difference was balanced out at 2 weeks, when no difference in number of kits in experiment and control groups in any of the experiments was found. Percentage barren females and number of kits are shown in table 1.6. In experiment III there were significantly more barren females in the control group than in the isolated group. The same tendency existed in experiment II, where a division in young and old females showed a significant difference in first year females with 13.8% barren in the experimental group against 0% in the control group ($P<0.05$).

Table 1.5 Litter size in the control and visually isolated female mink. Only females giving birth.

Groups

I = scanbrown isolated by an empty cage prior to mating (1985).
II = scanbrown isolated by an empty cage prior to birth (1985).
III = scanblack isolated by a cage filled with straw before birth (1985).
IV = scanbrown isolated by an empty cage prior to mating (1986).

	----- Isolated -----		---- Control ----				
a. Live at birth	No. of fem.	Mean ± SD	No. of fem.	Mean ± SD	t-value	DF	P<
I	73	5.44 ± 2.32	65	5.88 ± 1.84	-1.219	136	ns
II	183	6.17 ± 1.89	77	6.35 ± 2.11	-0.686	258	ns
III	43	5.67 ± 2.02	55	6.35 ± 1.88	-1.698	96	0,05
IV	130	6.19 ± 2.15	131	6.18 ± 1.86	0.037	259	ns
b. Live at two weeks							
I	73	5.27 ± 2.33	65	5.57 ± 1.78	-0.829	136	ns
II	183	5.85 ± 1.89	77	5.88 ± 2.38	-0.107	258	ns
III	43	5.19 ± 2.08	55	5.18 ± 1.99	0.010	96	ns
IV	130	5.72 ± 2.37	131	5.78 ± 2.34	-0.217	259	ns

Table 1.6 **Percentage of barren females and lost kits in the control and visually isolated female mink**

Group	Isolated	Control	χ^2 value	P<
a. Barren females %				
I	5.1	5.7	0.017	ns
II	7.6	2.5	1.695	ns
III	24.1	6.6	5.677	0.01
IV	10.8	8.4	0.416	ns
b. Lost kits %				
I	3.02	5.24	1.723	ns
II	5.14	7.36	2.358	ns
III	8.61	18.34	7.764	0.01
IV	7.58	6.30	0.719	ns

In experiment III there were significantly more lost kits in the control group than in the group of screened females calculated by means of χ^2 tests on living and lost kits in the experiment and control groups. The same tendency was seen in experiment II, but the difference was not significant at the 5% level. A division in young and old females did not show any significant difference, either.

There was no difference in the number of unsuccessful matings in any of the experiments.

The activity of the animals before weaning in experiment I is illustrated in figure 1.6. The activity of the females after birth in experiment IV is illustrated in figure 1.7. Mink in the control groups were at all times of day and in all observed periods more active than the visually isolated mink.

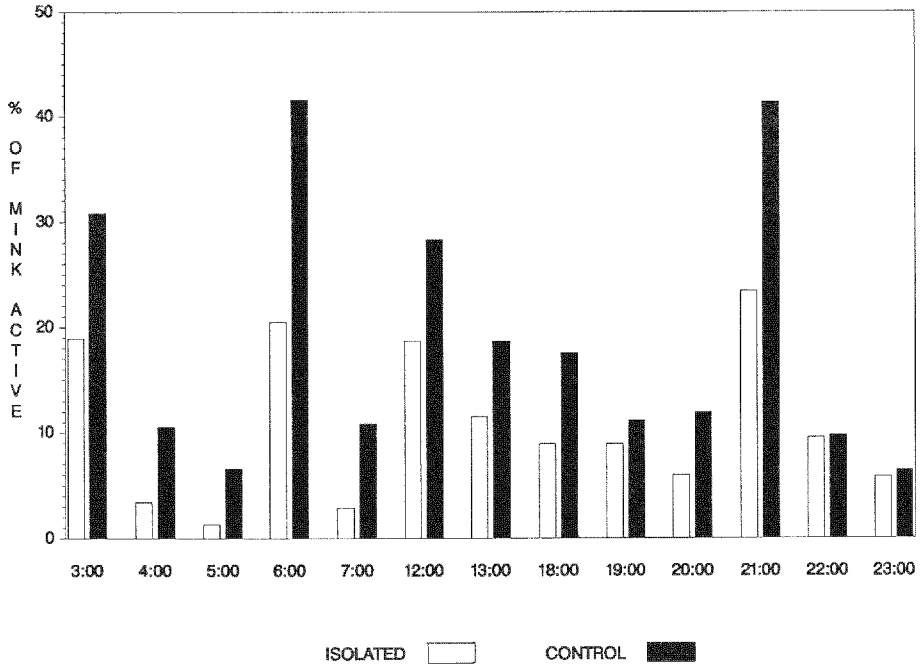

Figure 1.6 **Percentage of mink females and kits active in the cage at different times of the day in isolated and not isolated cages. Observations from experiment I before weaning.**

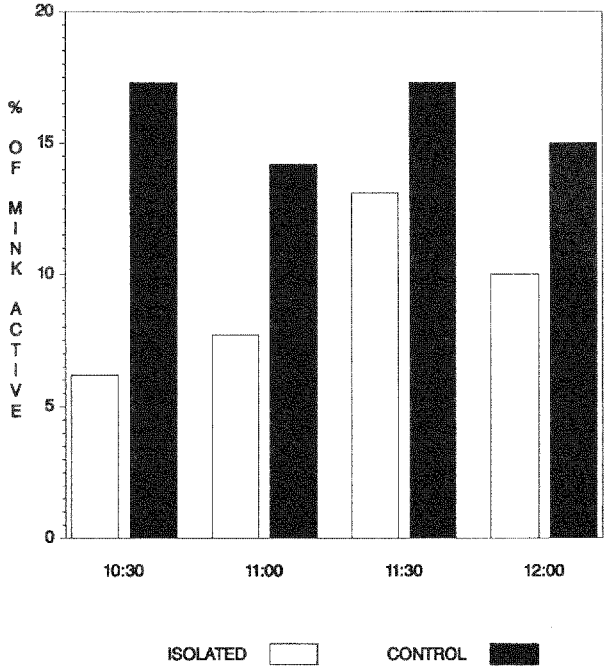

Figure 1.7 Percentage of mink females active in the cage during the morning in isolated and not isolated cages. Observations from experiment IV the week after birth.

There was no difference in the degree of nest building or territory marking with mounds of manure.

1.6.4 Discussion

In earlier investigations lower percentages of barren females and better whelping results have been found in females visually isolated from neighbouring females shortly before mating (Gilbert & Bailey, 1967, 1979; Hernesniemi, 1976; Vestergård, 19??a). In experiments I and IV, screening before mating showed none of these differences. There is no immediate explanation of the differences in number of living kits, barren females and loss

of kits in experiment III. As there is no difference in the number of kits at 2 weeks, this is no doubt due to the generally large uncertainty regarding the counting of kits at birth. As the screened females are less active in the cage, their kits have presumably been counted later. Stillborn and weak kits will therefore often be gone, which will influence both barren percentage, kits at birth, and loss of kits.

There may be many reasons why this investigation did not show the same differences in whelping results as previously. The nest box system has been changed and mink are more quiet today, because restless animals have been removed by selection. Animals without plasmacytosis are less sensitive to the mild stress factor constituted by neighbours. The large number of animals in the present investigations meant that poorly working males and the generally large variation in breeding results of mink females were evenly distributed on experimental and control groups. Later experiments have confirmed that no difference in whelping results of isolated and control females can be expected (Neil, 1989).

Vestergård (19??a) found that isolated females had heavier kits at weaning than control females. As an age difference of 9 days between the kits weighed was not evenly distributed on the experimental and control groups, the result is hardly contrary to the result that there is no effect of screening on the weight development of the kits. The author's conclusion also varies in the article (Vestergård, 19??b) and the abstract (Vestergård, 19??a). Neil (1989) did not find any difference in kit weight, either.

The increased activity in the control groups confirms earlier findings by Vestergård (19??a).

1.6.5 Conclusion

It can be concluded that a separation of mink by means of an empty cage results in lower activity of the animals. Neither breeding result nor kit gain are influenced by the isolation or the difference in activity.

The results of the experiments with visual isolation have been published in the annual report of the Danish Fur Breeders Association (Hoffmeyer & Møller, 1987) and discussed at the annual meeting of the Natl. Inst. of Anim. Sci. (Møller, 1987).

1.7 Scent communication

1.7.1 Introduction

During the mating season two different ways of placing the males in relation to the females are commonly used. In a section of six cages, one male is placed in a cage along with the five females he is meant to serve, or a group of males is placed beside the group of females they shall mate. Any communication between sexes that might influence heat development in females, is more effective when males are placed among the females than if placed in groups.

1.7.2 Material and methods

In order to examine whether the communication of scent between sexes influences the heat development in mink, cages were sprayed with urine from males prior to mating.

The cages of 40 pastel females were sprayed twice a day for four days before the mating started, while 40 females were kept as control. The time from introduction of the female to the male until the beginning of mating was measured, as well as the number of females that were not mated at the first attempt. Mating recordings were obtained from 37 females in the experimental group and from 34 in the control group.

1.7.2 Results and discussion

No significant changes in the number of unsuccesful mating attempts, nor in the whelping result were found on account of scent stimulation. The period from introduction till mating was significantly reduced from 15.9 till 11.8 minutes in the scent stimulated group ($P<0.01$). The placing of males in relation to females during the mating season does therefore not seem to be of importance in normal mink farming.

1.8 Weighing and behavioural response

1.8.1 Introduction

A weighing program was conducted each year to register the weight gain of kits on the project farms. From birth till weaning 25 litters were weighed every 10 days, and from weaning till pelting 25 pairs of kits were weighed every 2 weeks or every month. In connection with the many weighings, some farmers found that the weighed animals became more difficult to handle whereas other farmers did not observe any change.

1.8.2 Material and methods

The stick test has been developed as a fast and simple way of measuring the reaction of the animals to human beings. It was therefore used to describe the effect of the weighings on the 17 farms where the kits were weighed after weaning in 1987.

In the month of September 10 male and female kits from weighed groups and 10 control pairs were stick tested. The kits were weighed every 11 days until weaning and from then every fortnight. Not all of them were weighed each time, but on average the kits had been weighed 7 times before the stick test. Totally 310 pairs of kits of scanblack type were tested. At the stick test the kits were shut off from the nest box, a tongue spatula was thrust into the cage and their immediate reaction such as escape or exploration was noted.

1.8.3 Results and discussion

The distribution of behavioural response of weighing and control groups appears from table 1.7.

Table 1.7 Frequency of behavioural response to the stick test in the weighed and control groups.

	Escape	Exploration	Total
Control	47	263	310
Weighed	94	216	310
Total	141	479	620

Twice as many of the weighed animals as of the not weighed animals react with escape, which is strongly significant ($P<0.001$).

The distribution of behavioural response with regard to sex appears from tables 1.8 and 1.9.

Tabel 1.8 Frequency of behavioural response to the stick test in males and females.

	Escape	Exploration	Total
Males	53	257	310
Females	88	222	310
Total	141	479	620

Table 1.9 Frequency by sex of behavioural response to the stick test in the weighed and control groups.

Group	Sex	Escape	Exploration	Total
Control	Males	15	140	155
	Females	32	123	155
Weighed	Males	38	117	155
	Females	56	99	155
Total		141	479	620

It appears from the tables that considerably more females than males react by escaping ($P<0.001$). The difference is significant for the entire material and when split up in control and weighed animals ($P<0.05$). A certain reservation must, however, be made for the significances, as a test of independence was significant for the ani-

mals of the weighed group ($P<0.05$). For both sexes the weighed animals are significantly more timid than the control animals ($P<0.01$).

The reaction to the stick test is shown in figure 1.8 for both weighed and control groups.

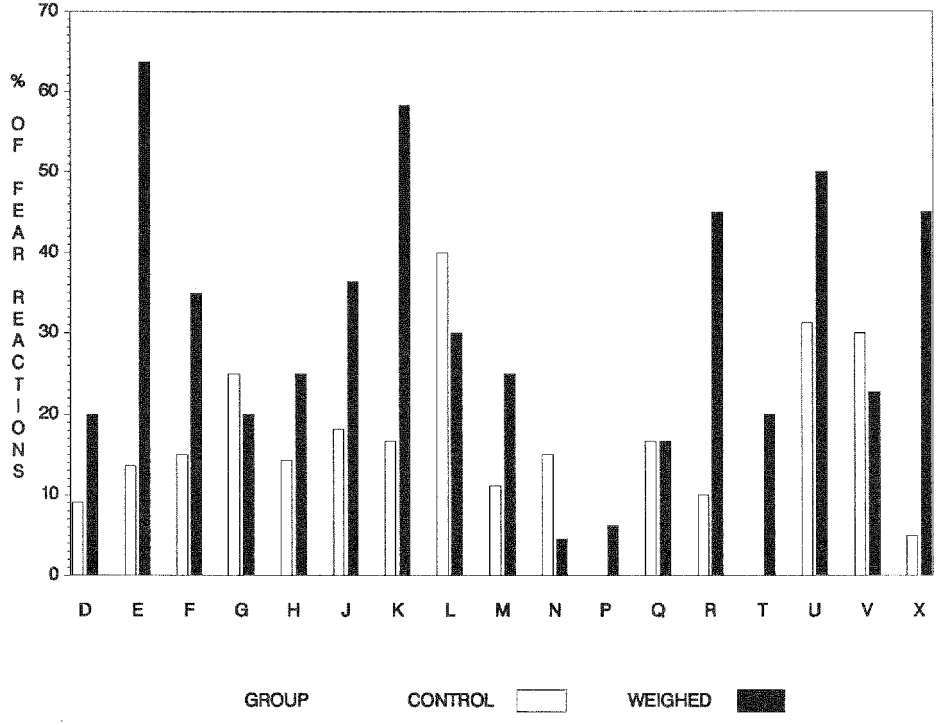

Figure 1.8 Percentage of fear reactions in weighed and control groups of mink kits. Found by stick test in September on 17 farms.

The level of escape reactions as well as the relation between weighed and control groups differ very much from farm to farm. A few farms have more escape reactions in the control group, but on all farms with significant difference between groups, the share of escaping animals is higher in the weighed group.

1.8.4 Conclusion

Weighed animals generally react more timidly than animals that have not been weighed. In general, mink must therefore regard weighing as a strain, but the large difference between farms shows that the strain depends on how the weighing is carried out and by whom. On some farms weighed animals were even less timid than not weighed animals. The results show that the reaction of the animals is not only heriditary but is to a high extent an expression of the acquired experience with handling.

In general, females react more timidly than males. The dependence between sexes makes it "harder" to prove this fact because both animals from a cage tend to react the same way. The difference between sexes is significant in spite of this and the dependence does not impair the conclusion. The dependence must be due to the fact that the animals are influenced by the reaction of each other to strain, as this is only found in the group of weighed animals. The stick test therefore illustrates the amount of handling the animals have been subjected to, whereas the "basic level" in not handled animals expresses the genetic difference between strains and the general relation of the animals to human beings.

The results have been published in a Short Comm. from the Natl. Inst. of Anim. Sci. (Møller & Hansen, 1988).

1.9 Weight development, body length, skin length and pelt quality.

1.9.1 Introduction

It has been known for a long time that there is a negative correlation between skin size and pelt quality (Nielsen, 1972). However, breeding efforts have been concentrated on both traits without having an exact description of the correlation and the possibilities of improving both at the same time.

The body development of the mink ends at the beginning of September, and gain will from then on almost exclusively be deposits of fat (Hansen et al., 1981).

The importance of body length and weight to the length of the skin can be separated and illustrate the possibilities of breeding for different ways of obtaining skin length.

1.9.2 Material and methods

Thirty litters of 5-8 scanblack kits were weighed every 10 days in the lactation period. After weaning the weighed group was extended to 120 male-female pairs that were weighed every fortnight until August 26th and thereafter on September 24th and October 27th when the animals were killed and body length measured from tip of nose to tail root. Pelt quality was evaluated on a 1-15 scale, density from 1-5. Leather thickness was measured on the back line with a micrometer screw, and "weak hips" were evaluated according to a scale from 0-3.

1.9.3 Results and discussion

Weight development of the mink was normal in the lactation period as well as after weaning. Skin size and quality, body length and weight are shown in table 1.10. It appears that there is a rather large variation in skin length, weight and quality, whereas body length varies less.

Table 1.10 Skin size, pelt quality, body weight and length of 91 scanblack male mink.

	Unit	Min.	Max.	Mean	SD
Skin size	cm	67	83	75.43	3.40
Pelt quality	1-15	1	15	9.24	3.65
Body weight	g	1620	2810	2505.5	253.7
Body length	cm	43	50	46.65	1.58

Correlations between weight development of the kits until weaning and body and skin length at pelting, and weight at pelting are shown in table 1.11.

Table 1.11 Correlation between weight before weaning and length of body and skin and weight at pelting of 48 males.

Age in days	9	19	30	40	52
Body length	0.48***	0.43**	0.41**	0.37**	0.44***
Skin length	0.32*	0.21ns	0.24ns	0.18ns	0.32*
Weight at pelting	0.32*	0.23ns	0.26ns	0.19ns	0.40**

*P<0.05; **=P<0.01; ***=P<0.001; ns=P>0.05

It appears that already from the age of 9 days there is a high correlation between weight and body length at pelting, whereas the correlation with skin length is lower but still significantly different from zero at the age of 9 and 52 days. The weight at 9 and 52 days is also correlated to the weight at pelting.

Correlations between kit weight after weaning and body length, skin length, weight at pelting and pelt quality are shown in table 1.12.

Table 1.12 Correlation between weight after weaning and length of body, skin length, weight at pelting and pelt quality of 91 males.

Date Age in weeks	15/7 10	29/7 12	12/8 14	26/8 16	24/9 20	27/10 25	At pelting
Body length	0.58	0.64	0.64	0.66	0.68	0.73	0.72
Skin length	0.54	0.64	0.69	0.73	0.79	0.84	0.87
Weight at pelting	0.57	0.67	0.73	0.78	0.85	0.91	1.00
Pelt quality	-0.15	-0.21	-0.33	-0.36	-0.42	-0.44	-0.38

All correlations are significant (P<0.001).

Correlations between weights after weaning and body length, skin length and weight at pelting are increasing until pelting. From the middle of August the correlation between weight and skin length is higher than between weight and body length illustrating that body length is fully developed and that from September gain mainly consists of fat. The increasing negative correlation between weight after weaning and pelt quality illustrates that fat deposits are responsible for the reduction in pelt quality.

The three measurements of mink size at pelting have been correlated to four measurements of skin quality and are shown in table 1.13.

Table 1.13 Correlation between different quality measures and sizes of 91 male mink.

	Quality	Density	Thickness of leather	"Weak hips"
Skin length	-0.42***	-0.29**	0.43***	0.38***
Body length	-0.20ns	-0.14ns	0.26*	0.15ns
Body weight	-0.38***	-0.23*	0.41***	0.33**

*=P<0.05; **=P<0.01; ***=P<0.001; ns=P>0.05.

In several cases a negative correlation between skin length and fur quality has been reported. This investigation revealed that the negative correlation is due to weight rather than body length. The same applies for fur density. Long skins from the heaviest mink have the thickest leather, whereas body length is less important. The same applies for the skin defect "weak hips".

A multiple regression shows that both body length and weight at pelting contribute significantly to explain the variation in skin size.

The regression equation looks as follows:

Skin length cm = 26.4 + 0.63 · body length cm + 8.8 · weight kg.

All three terms are different from 0 ($P<0.001$) and $R^2 = 0.79$.

A similar regression with skin quality as dependent variable shows that only the weight contributes significantly to the variation in quality. The regression equation looks as follows:

Skin quality = 7.2 + 0.35 · body length cm - 6.41 · weight kg.

Only the weight term is significantly different from 0 ($P<0.001$) and $R^2 = 0.16$.

The calculation can be defended, as the subjectively evaluated skin quality is normal distributed.

The degree of fattening varying between 37.7 and 59.1 g/cm is a very good expression of the relationship between weight and length. However, regression with degree of fattening gives a poorer description of the variation in skin size and quality than the multiple regressions with weight and length. The results show that the negative correlation between skin size and quality is primarily due to weight, whereas body length plays a secondary role.

The correlation between average skin size and quality for more than 4000 mink farms in the database from the Danish Fur Auctions was calculated for 1985, 86 and 87. The correlations were between 0.10 and 0.06, but still significant. Thus it seems possible in practice to improve both traits simultaneously.

1.9.5 Conclusion

All in all it can be concluded that a long skin from a long and slim mink is of better pelt quality and density, has thinner leather and is less liable to develope "weak hips" than a long skin from a fat mink. Skin length is correlated more to weight at pelting ($r = 0.87$) than to body length ($r = 0.76$). With present feeding routines mink can fully utilize their capacity for growth of length. A longer mink can therefore only be obtained by breeding for body length.

The problem in connection with application in practice is that variation in weight is larger and above all easier to measure than variation in length. It is therefore easier to breed for weight which also gives the best response in skin length. Unfortunately the effect on quality is usually negative. In practice, however, it is possible to improve skin size and quality at the same time.

1.10 Development of body length and weight in relation to time of birth and litter size

1.10.1 Introduction

Variation in length and weight of the mink is the basis for selection according to these factors. It is therefore important to know the reasons for the variation in order to find the right way and the right time for selection.

1.10.2 Material and methods

The importance of time of birth and size of litter for length and weight development was examined on 345 scanblack male kits. The kits were weighed approx every 3 weeks from July 6th until pelting and length was measured at the age of 7, 13 and 16 weeks and at pelting (28 weeks). Time of birth was divided into 3 groups: 1) born in April, 2) born May 1st-5th and 3) born after May 5th.

There were 88, 211 and 52 kits in groups 1, 2 and 3, respectively.

Litter size was corrected for an uneven distribution of sexes to 50% male and 50% female kits according to the formula:

corrected litter size = (male kits · 105 + female kits ·95)/100

and afterwards split up in intervals of 2 kits from corr. litter size of below 2.5 till over 8.5.

1.10.3 Results and discussion

The kits developed normally and reached a maximum mean weight of 2085 g in October and 2018 g at pelting. Length increased between all measurements until 47.8 cm at pelting.

Development of length in relation to time of birth is illustrated in figure 1.9.

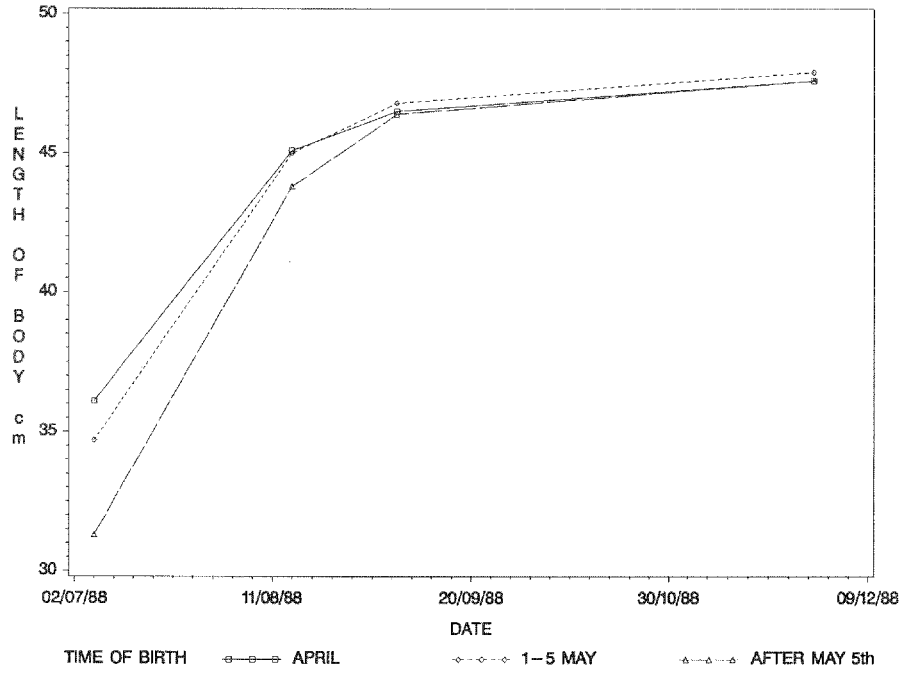

Figure 1.9 Growth in body length in relation to time of birth of male mink kits from weaning till pelting.

It will be seen from the figure that the length at the first two measurements is dependent on time of birth but that the kits born late have caught up with the early born kits at the measurement on September 9th, and from then on there is no difference.

Weight development in relation to time of birth is illustrated in figure 1.10.

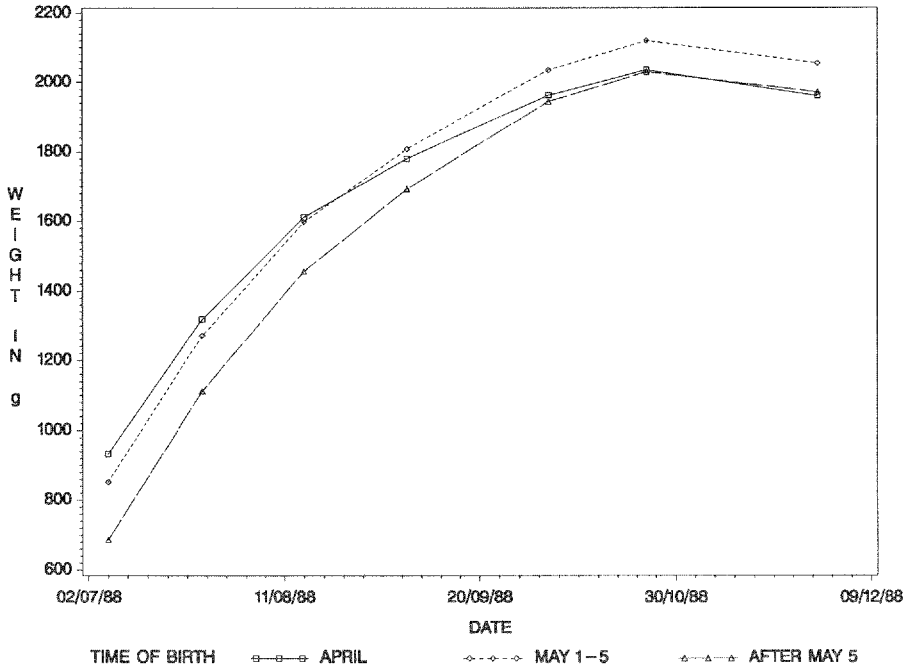

Figure 1.10 Weight gain in relation to time of birth of male mink kits from weaning till pelting.

It appears from the figure that weight is influenced by time of birth all the time until pelting. The difference is significant until August 15th, and from then on the difference between the earliest and latest born kits is balanced out.

Kits born May 1st-5th stay approx 100 g ahead of the others until pelting, but the difference is not significant.

Development of weight at different litter sizes is illustrated in figure 1.11.

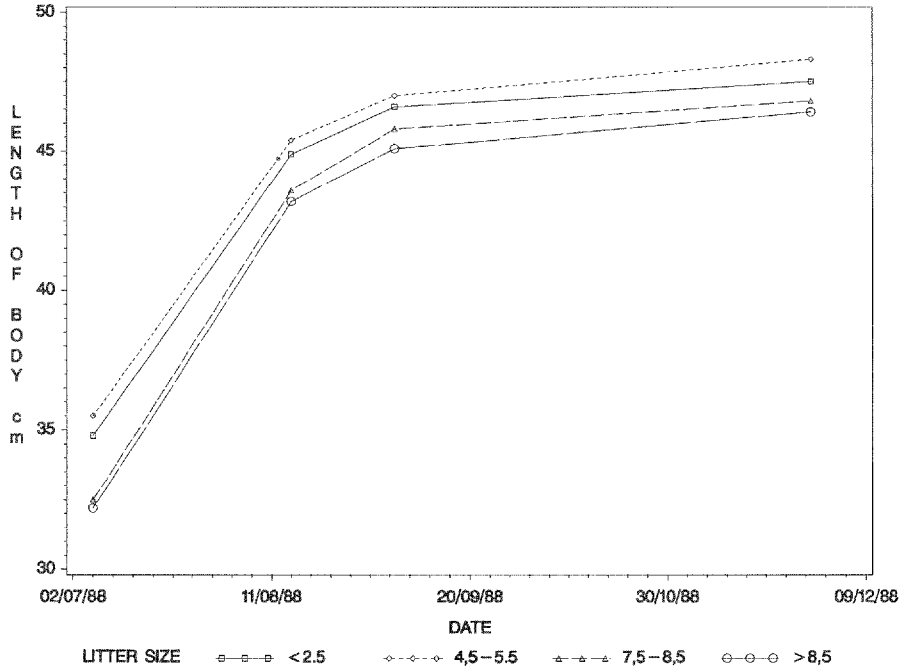

Figure 1.11 Growth in body length in relation to litter size of male mink kits from weaning till pelting.

Litter size effects development of length until August 15th, and after that the effect is not significant.

Kits from large litters partly catch up in length by the time of weaning, whereas kits from very small litters only keep up - without getting bigger than the others. At pelting the longest animals come from litters of 2.5-7.5 kits.

Weight development in relation to litter size is illustrated in figure 1.12.

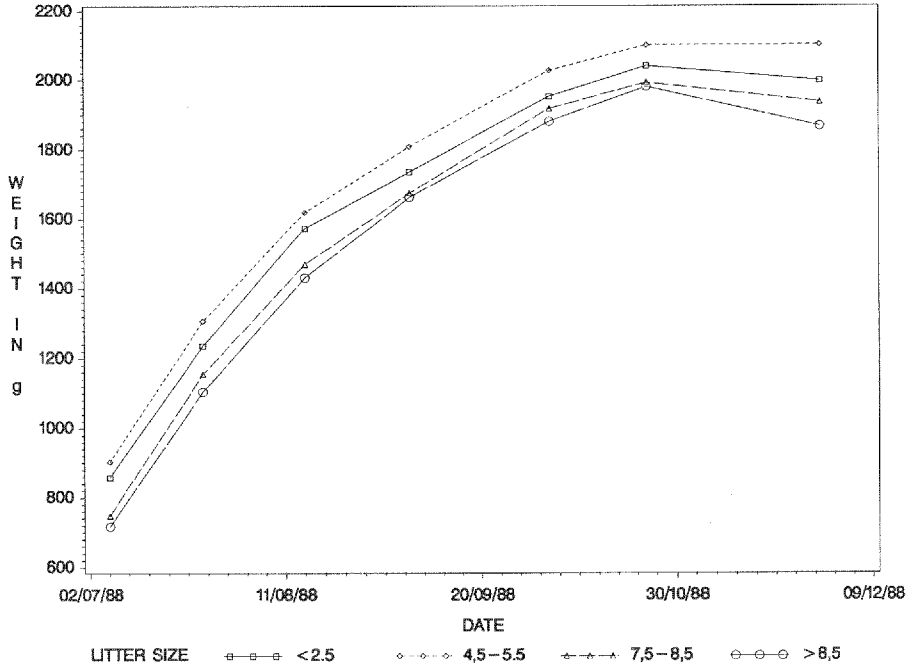

Figure 1.12 Weight gain in relation to litter size of male mink kits from weaning till pelting.

Kit weight is influenced by litter size until August 15th, and after that there is no significant difference. Even though kits from large litters gain weight faster than the others after weaning, they do not reach the medium-size litters of 2.5-6.5 kits which are the heaviest at pelting.

1.10.4 Conclusion

The conclusion is that the effect of time of birth is balanced out at the age of 16-20 weeks both with regard to development of length and weight.

Kits from large litters have some compensatory weight after wean-

ing but they do not catch up on weight or length before pelting. Very small litters do not develop as well as larger litters, and the best development of weight and length is seen in litters of 3 to 7 kits.

The results of investigations of weight and length were presented at the Annual Meeting of the Natl. Inst. of Anim. Sci. (Møller, 1988a; Hansen, 1989).

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2. BEHAVIOUR AND ENVIRONMENT OF MINK

by Steffen W. Hansen

Summary

In the project section behaviour and environment, investigations have been carried out regarding the development, weaning age, cage environment, activity and temperament of mink kits.

The ontogenetic investigations have dated important periods in the development of mink kits. Thereby possible correlations between the development stage of mink kits and the suboptimal adaptation to the production environment have been determined.

Weaning age, varying from 6 to 12 weeks, when the kits are taken away from the female and placed in pairs, male and female, is of no importance to their later reproduction capacity. The examinations of the optimum weaning age have included the welfare of the female as well as of the kits.

Individual parameters regarding the physical cage environment have shown that the nest box is important to the welfare of farmed mink and to the fur quality. A 4-time increase of the cage size area as compared to the conventional cage size did not result in a better welfare.

The installation of water trays for lactating females had no improving effect on the welfare of the females or the kits, neither on the ontogenetic development of the kits. The water trays do, however, probably have an occupational function. It was found that a place of refuge for lactating females reduced the frequency of stereotypic behaviour - a behaviour which is very energy-consuming.

The activity pattern of lactating females has been described. As for other periods of the production cycle of farmed mink, also the activity of lactating females to a considerable extent seems to be controlled by the feeding routine.

A simple and fast test (the stick test) of the temperament of mink has been developed. The test is a

practical tool for identification of farmed mink with reduced adaptability to the production environment, i.e. animals with a timid temperament.

Behavioural, physiological, haematological and physical-chemical variables are included in this project section for evaluation of the welfare of farmed mink.

Sammendrag

Under delprojektet adfærd og miljø er der gennemført undersøgelser vedrørende minkhvalpenes udvikling, fravænningsalder, burmiljø, aktivitet og temperament.

De ontogenetiske undersøgelser har tidsfæstet vigtige perioder i minkhvalpenes udvikling. Dermed er mulige sammenhænge mellem minkhvalpenes udviklingsstadie og suboptimal tilpasning til produktionsmiljøet påvist.

Fravænningsalderen, varierende fra 6 til 12 uger, ved hvilken hvalpene fjernes fra tæven og placeres parvis han og tæve, har ikke betydning for deres senere reproduktionsevne. Undersøgelserne af den optimale fravænningsalder har inddraget både tævens og hvalpenes trivsel.

Af enkeltparametre fra det fysiske burmiljø er redekassen fundet at være væsentlig for farmminkens velfærd og skindkvalitet. En 4 gange forøgelse af burstørrelsens areal i forhold til den konventionelle burstørrelse bidrog ikke til øget velfærd.

Installering af vandbakker til diegivende tæver har ikke haft nogen forbedrende effekt hverken på tævens eller hvalpenes trivsel (jævnfør hvalpenes ontogenetiske udvikling). Derimod har vandbakkerne en sandsynlig beskæftigelsesmæssig funktion. Det blev fundet, at en retræteplads for diegivende tæver reducerede frekvensen af stereotyp adfærd, en adfærd som er meget energiforbrugende.

Aktivitetsmønstret hos diegivende tæver er beskrevet. Som i andre perioder af farmminkens produktionscyklus synes også diegivende tævers aktivitet i væsentlig grad at være styret af fodringsrutinen.

En simpel og hurtig test af minks temperament (pindetest) er blevet udviklet. Testen er et praktisk værktøj til identifikation af farmmink med nedsat tilpasning til produktionsmiljøet, dvs. dyr med frygtsomt temperament.

Som grundlag for vurderinger af farmminks velfærd i dette delprojekt er inddraget adfærdsmæssige, fysiologiske, hæmatologiske og fysisk-kemiske variabler.

2.1 Introduction

When this project was started, existing knowledge as regards the basic behaviour of fur animals was very limited. The importance of cage environment and management to animal behaviour and the final production result was therefore poorly illustrated.

The purpose of this part of the project - "behaviour and environment" - was therefore to obtain more knowledge of the basic behaviour of farm produced fur animals under production conditions and to illustrate the influence of management and various environmental factors on animal behaviour, physiological stress level and production result.

2.2 Research topics

This part of the project was carried out by basic recording of the temperament and physiological stress level of mink on 20 selected farms.

Based on this information important management areas such as: weaning routines, feeding and watering, the mutual contact between animals as well as the daily contact between animals and human beings were investigated experimentally on the research farm.

Important parameters from the cage environment supposed to be of importance to the behaviour of farm mink, such as cage size, access to nest box and various sorts of enrichment of the cage environment, were investigated experimentally on the research farm.

With the project three final theses have been prepared in cooperation with the Institute of Population Biology at the University of Copenhagen.

Subjects investigated in "Behaviour and Environment":

2.3 The importance of cage size and nest boxes.

2.4 Activity pattern of lactating females and the importance of water trays and resting place.

2.5 Effect of water trays for farmed mink.

2.6 Correlation between behavioural response of stick test and level of eosinophil leucocytes of mink females.

2.7 Importance of time of weaning on mating success and temperament of mink.

2.8 Theses from the University of Copenhagen, Inst. of Population Biology:

- Importance of early weaning and handling to later timidity of mink.
- Ontogenesis in mink kits.
- Development of behaviour in juvenile mink held in cages and the importance of stress to behaviour, physiology and pelt quality.

2.3 Importance of cage sizes and nest boxes

2.3.1 Experiment 1

The cage sizes used for mink on farms are very uniform. The cages are of 3 types: 6-room, 8-room, and top nester cages with a floor area of 0.27m², 0.21m², and 0.18m², respectively.

To investigate the effect of these cage types on the behaviour of the animals, 18 adult male mink and 36 kits of both sexes were used. The behaviour of the animals: passivity, activity, stereotypies and as regards the kits also "play fighting" and "climbing in wire netting" were recorded in three periods of 5 days with 10 observations per day.

No difference in behaviour was found between mink in the three types of cages. Large individual differences were found as regards the frequency of stereotypic behaviour, and females show stereotypic behaviour more often than male mink.

The results were published in a Short Comm. from the Natl. Inst. of Anim. Sci. (Jonasen, 1987).

2.3.2 Experiment 2

To investigate whether extreme sizes of cages were of importance to the behaviour, physiology and productivity of farmed mink, 228 mink were kept in pairs (male + female) in cages with areas varying from 1.056 m² to 0.105m². The importance of nest boxes to the mink was investigated by depriving a group of mink in standard cages of nest boxes. The cage sizes examined were of the following dimensions:

- type 1 (110 L x 96 W x 76 H (cm)) = 1.056m² + nest box
- type 2 (90 L x 30 W x 45 H (cm)) = 0.270m² + nest box
- type 3 (35 L x 30 W x 45 H (cm)) = 0.105m² + nest box
- type 4 (70 L x 15 W x 45 H (cm)) = 0.105m² - nest box
- type 5 (90 L x 30 W x 45 H (cm)) = 0.270m² - nest box

The animals were observed regularly one day per week from June 28th to October 4th, 1987. The observations were made from 7.00 to 9.30 a.m. and from 11.00 a.m. to 2.00 p.m. which are the most active

periods during the day for farmed mink (Dodd, 1985). Each cage was observed for 2 minutes and any occurrence of predefined elements of behaviour were recorded for both sexes. The following elements of behaviour were recorded:

1. How often and for how long was the nest box used.
2. How often and for how long were animals lying out in the cage.
3. How often did they attack, defend, run away, push or pull each other around by the scruff (agonistic behaviour).
4. How often did they start playing, licking themselves, climbing in the wire netting, sniffing.
5. How often did the animals leave scent marks either by rubbing their chin/belly against the bottom (belly crawl) or by dragging the anal region over the bottom (anal drag).
6. How often did the animals perform "abnormal" behaviour: stereotypes, rollings and biting in the netting.

The weight development of the animals was recorded by 5 weighings throughout the experimental period. In August, October, and November blood samples were taken from all animals by toe nail cutting to determine individual eosinophil leucocyte level. In October and November blood samples were also taken for differential leucocyte counts. When the animals were killed, 20 ml blood was taken by heart puncture for examination of haematological and physical-chemical variables. The haematological variables were as follows: Haemoglobin, haematocrit, mean cell volume, mean cell haemoglobin, erythrocytes, leucocytes, thrombocytes, eosinophil granolocytes. The physical-chemical variables were as follows: alanin aminotransferase, aspartat aminotransferase, creatin kinase, total plasma protein, plasma ascorbic acid and adrenal ascorbic acid. After pelting all skins were examined for bites on the inside of the leather, and skin quality evaluated.

To relate the physiological parameters recorded to corresponding parameters of experimentally stressed mink, where the physiological stress response is known from literature (Heller & Jeppesen, 1985), 8 females of the same age kept under normal farm conditions were exposed daily to one hour of immobilization in a mink trap for 8 days. After they had been stressed, these 8 females were - together with 40 females from the various cage sizes - tested as regards function of adrenal cortex.

2.3.3 Results and discussion

2.3.3.1 Behaviour

Cage sizes were of no importance to the activity level of mink kits, but kits in cages without nest boxes had a higher activity level. The frequency of the use of nest boxes was significantly lower in cage type 1 compared to cage types 2, 3, and 4, but there was no difference in the duration of their stay in the box. In cage type 3 the dimensioning prevented the animals from performing normal marking behaviour (belly crawling).

The higher frequency of climbing behaviour in cage types 1 and 4 was due to the fact that male mink were here forced to climb in the wire netting to reach their feed, and in cage type 4 that the narrow physical environment forced the animals to pass each other by climbing over each other. The cage types investigated did not influence the frequency of agonistic behaviour but there were differences in relation to sex.

"Abnormal" behaviour such as "biting in wire netting" and stereotypic behaviour occurred as frequently in all cage types, but the type of stereotypic behaviour varied between cage sizes.

2.3.3.2 Haematology

The haematological values measured were all within the normal area for mink of the age in question. Mink in the large cages had a significantly higher haemoglobin concentration than mink in the smaller cages, but the other haematological values showed no effect of cage size.

2.3.3.3 Physical-chemical parameters

The physical-chemical examinations expressing metabolic activity showed that mink in the large cages had higher values of alanin aminotransferase and aspartat aminotransferase than mink in the smaller cages. These parameters tended to show increased values in mink without nest boxes. The other physical-chemical variables showed no effect of cage size.

2.3.3.4 Eosinophil leucocytes

The number of eosinophil leucocytes in male mink in the various cage sizes was constant and uniform throughout the experimental period.

In August, the eosinophil level of mink females was uniform and at the same level as that of male mink, but then the level dropped throughout the experimental period. At blood sampling in October, females in cage type 1 had a significantly higher eosinophil level than mink females in cage types 3 and 4. At the last blood sampling in November there was no difference between females in the various cage sizes, but the level was significantly lower than in the males.

The eosinophil level of male mink without nest boxes dropped significantly throughout the experimental period, while in male mink with nest boxes it remained high.

The results have shown that the cage sizes investigated do not influence the level of eosinophil leucocytes in mink. For mink with access to nest box males had a highher level of eosinophil leucocytes than females. These sexual differences were surprising and contrary to previous results (Jeppesen & Heller, 1985; Browerman et al., 1974). For mink without access to nest box there were no sexual differences but males without nest box had a lower level of eosinophil leucocytes than males with nest box. This makes it probable that other factors influence the level of eosinophil leucocytes. A possible explanation could be that an increased activity level reduces the eosinophil level. The behavioural investigation partly supports this assertion.

2.3.3.5 Differential leucocyte count

No difference was found in relative occurrence of leucocytes (differential leucocyte count) between mink in the various cage sizes. Gross & Siegel (1983) found that an increase in the heterophil leucocyte:lymphocyte ratio (HL-ratio) was a reliable measure of stress in chickens. Kristensen & Jeppesen (1988) confirmed this result in foxes.

The result of this experiment showed that the cage sizes examined have no influence on the stress level of mink. In relation to mink with nest boxes, mink without nest boxes had a significantly higher HL-ratio indicating that mink without nest boxes were more stressed than mink with nest boxes.

2.3.3.6 Adrenal cortex functional test

The results of the adrenal cortex functional test (AC-FT) showed no difference in cortisol release in females from the various cage sizes, which again indicates that cage size has no influence on the

physiological stress response of mink. Mink females without nest boxes reacted by releasing more cortisol than mink females which had been experimentally stressed through immobilization in a mink trap. In general, differential leucocyte counting after AC-FT showed an increase in the HL-ratio of mink females. Mink females without nest boxes had a higher HL-ratio than mink females which had been immobilized. The results show that experimental stress through immobilization and environmental stress through deprivation of nest boxes cause different physiological reactions. Mink without nest boxes could be long-term stressed with a degree of adrenal hypertrophia and reacted to AC-FT by an increased release of cortisol. Mink females that have been immobilized react to AC-FT (acute stress) by a slight increase of cortisol and HL-ratio, as if the preceding immobilization could be "regarded" as a number of repeated acute stress influences.

2.3.3.7 Production results

Examination of the mink skins showed that cage size does not affect the number of bite marks recorded on the inside of the leather which supports the results showing that agonistic behaviour was not influenced by cage size. Females had more bite marks than males.

The final grading of the skins showed no effect of cage size, but mink without access to nest boxes had a significantly poorer pelt quality than mink with nest boxes.

2.3.4 Conclusion

A 4-time increase of the area of existing cage sizes has no effect on the well-being of farmed mink. Depending on dimensions, cage sizes with an area of 0.105m² prevent certain behaviour patterns from being expressed. Deprivation of mink from the use of nest boxes caused physiological changes indicating long-term stress.

The results have been presented and published at the 4th Int. Congr. in Fur Animal Prod. and in SCIENTIFUR (Hansen, 1988; Hansen & Brandt, 1989).

2.4 Activity pattern of lactating females and the importance of water trays and resting place

2.4.1 Introduction

In the lactation period females have a considerable need for water. Part of the water is used for milk production. The increased metabolism results in a larger need for liquid for heat regulation and for

segregation of breakdown products. Besides these special conditions during the nursing period, the need of liquid and feed depends on the activity level of the female.

Patterns of movement without any function and stereotypic pendling up and down the wall of the cage at a high speed are supposed to be extremely energy demanding.

These patterns of movement often occur in connection with the feeding situation. It may also be expected that at the end of the lactation period the presence of the kits motivates the females partly to be with the kits and partly to try to get away from the kits which may increase the stereotypic activity.

The purpose of this project was to illustrate the pattern of activity of the females during the last 4 weeks of the lactation period, which must be considered the most stressing period for the female. There was also a wish to investigate the importance of water trays as an extra supply of liquid and as a place of refuge for the female. Also the importance of wire netting cylinders as a place of refuge for the female was examined.

2.4.2 Materials and methods

Fiftynine mink females of the colour type pastel were included in the experiment. Since weaning, the mink had been living in conventional mink cages. Immediately before whelping, water trays (30 cm L x 20 cm W x 2 cm H) were placed in 16 of the cages approximately 15 cm below the cage ceiling. In 20 other cages wire netting cylinders (diam. 10 cm, length 30 cm) were suspended from the cage ceiling. No changes of the cages were made for the remaining 17 females.

The water trays were filled with water daily at 10.30 a.m. Apart from that, the animals were taken care of according to normal farm routine. The animals were fed at 1.00 p.m., and the following morning at 8-9 a.m. the feed remains were redistributed.

Individual scanning observations were carried out on 5 consecutive days (Monday-Friday) in each of the weeks 22, 23, 24, and 25. The observations started when the kits were approximately 4 weeks old (on May 25th, 1987), and ended when they were approximately 8 weeks old (on June 22nd, 1987).

During a Scan sampling the observer passed the cages 4 times per hour and recorded the ongoing activity of the mink.

The following elements of behaviour were recorded:

1) Mink in nest box.
2) Mink lying in entrance to nest.
3) Mink lying on the wire netting floor of the cage.
4) Mink non-specifically active out in cage.
4a) Mink pendling out in cage.
4b) Mink performing stereotypic behaviour out in cage.
4c) Mink on water tray.
4d) Mink in wire netting cylinder.
9) Kits in entrance to nest.
10) One or more kits out in cage.

Totally 347 scannings were made, distributed on the day hours from 8 a.m. to 3 p.m., corresponding to approximately 43 observations per hour per 4 weeks.

The results of the behavioural observations have been calculated as number of observations where the position or behaviour in question was observed in per cent of total number of observations in the period in question, i.e. the per cent frequency of the position/behaviour.

The elements of behaviour "pendling", "stereotype", "in wire netting cylinder", and "on water tray" have been calculated as per cent of observations where the female has been active out in the cage.

The females were weighed on May 1st, and both females and kits were weighed, when the kits were 3 weeks old, and when the kits were weaned at the age of 7-8 weeks.

When calculating the weight development of the females throughout the experimental period, corrections have been made for time of birth according to the following formula: corrected weight = measured weight + (average number of days after birth - actual number of days after birth) · weight change per day.

2.4.3 Results and discussion

All three experimental groups increased their active behaviour out in the cage as a function of time. This increase corresponded to a decrease in the use of nest box, lying in nest box opening, and lying out in cage, respectively.

The reason for the increase in activity may be a rising temperature in the nest box due to crowding, and that the female in periods tries to get away from the kits. The importance of the increasing temperature to the use of nest boxes by the females is supported by the fact that in the afternoon the females were more out in the cage.

Investigations regarding the optimal age of weaning have shown that when the kits are weaned at the age of 8 or 10 weeks, the physiological stress level of the females is reduced. If the kits are weaned at the age of 6 weeks, no immediate reduction of the stress level is seen, but the weaning in itself seems to be stressing for the female at the time (Jeppesen, Heller & Houbak, 1988). At the end of the experimental period, the presence of the kits also causes increasing stress for the female which may result in increased activity by the females. Visual contact between neighbouring females has previously been shown to increase activity of lactating females (Hoffmeyer & Møller, 1987).

When the general activity increased, the elements of behaviour, pendling and stereotypic behaviour, amounted to an increassing part of this activity. When the kits were approximately 8 weeks old, the stereotypic behaviour occurred at the highest frequency.

Stereotypic behaviour in domestic animals is often seen as an indicator of poor adaptation to housing conditions. It has, however, not been possible to prove a significant correlation between frequency of stereotypic behaviour and physiological stress. This has given rise to the assumption that the performance of stereotypic behaviour can be stress-reducing in itself (Wiepkema, 1985).

Under production conditions, domestic animals are given limited space and their activity will therefore often be of a stereotypic nature, especially their running up and down in the cage (pendling). Later investigations (Hansen, 1990) have shown that the level of eosinophil leucocytes of the female is three times higher immediately before the time of birth than when the kits are weaned at the age of 8 weeks. Compared to the low activity level of pregnant females, it is likely that there is a negative correlation between activity and level of eosinophil leucocytes of females. This assumption is supported by results obtained with mink kept in conventional mink cages with and without nest box, respectively. Mink deprived of the use of nest boxes were more active and had a significantly lower eosinophil level (Hansen, 1989).

A comparison among the three experimental groups showed no difference as regards activity out in the cage. The recording of this element of behaviour includes, besides pendling and stereotypes, also position on water tray and in wire netting cylinder. The difference found between female groups in the first two weeks as regards the elements of behaviour

"lying out in cage" and "active out in cage" is probably due to the fact that females used the water trays partly as resting place and partly for active "bathing". They were therefore quickly empty of water and could again be used as resting place. Females with wire netting cylinders used these as resting place.

In the last two weeks the "bathing possibility" was not used, and because of water in the trays the females were prevented from using the trays as resting place. Females with wire netting cylinders continued to use these as an alternative to "lying out in cage".

Females with wire netting cylinder differed from the other two experimental groups by a significantly lower frequency of the elements of behaviour "pendling" and "stereotypes". But there was no difference between groups as regards "activity/inactivity" and "out in cage". It is therefore reasonable to assume that the use of wire netting cylinders to some extent reduces the performance of the behavioural elements "pendling" and "stereotypies".

A possible explanation may be that the narrow wire netting cylinder surrounding the body of the mink is regarded by the mink as a "safe" place, and at the same time the animal is placed up high in comparison with the neighbouring animals. This tendency to seek a high position as a resting place is also known from other carnivores (Hansen, 1985).

At the end of the experimental period the use of the netting cylinder by the females decreased probably due to the fact that the wire netting cylinders were no longer considered "safe", as the kits could now reach the female in the cylinder.

Throughout the experimental period the use of water trays by the females was very limited. That the females used the water trays at the beginning, and later on almost stopped using them, may be due to the fact that at the beginning the water trays may have stimulated the animals due to their novelty value.

At the beginning of the experimental period females with access to water trays differed from the other females by using the nest box more and by being less out in the cage.

The interpretation of the result may be that females with water trays are cooled off when using the water trays and therefore do not have to get away from the nest box, or that the kits are cooled off by the wet pelt of the female and therefore need for the female to be

with the kits for a longer time. Observations of mink having access to swimming in pools showed that the animals often concluded their "bathing session" by rubbing their pelt against a moisture-absorbing material, like for in stance sawdust. The lack of this possibility may have a reducing effect on the use of water trays. It is, however, more likely that the female has used the material in the nest for this sort of comfort-behaviour.

2.4.4 Conclusion

As regards the female's use of water trays none of the results found caused the assumption that lactating females have a need to "bathe". As it was not observed that mink kits lick liquid from the female's wet pelt, this possibility for offering extra liquid to the kits is probably non-existant, or at least of no importance.

An identical weight development of females and kits, respectively, in all three experimental groups (despite the low number of experimental animals) supports this conclusion.

The activity in the day hours seems to be controlled by the feeding hours at 8-9 a.m. and approx at 1 p.m. which has also been proved by previous investigations (Jonge et al., 1985). Between the two feeding sessions, the animals were most passive and placed either in the nest box or in the nest box entrance.

The activity of the females just before feeding was considerably higher at the end of the experimental period than in the beginning. The asynchronous presence of kits and females, respectively, out in the cage was significant in the morning hours. After feeding at 1 p.m., the females maintained a relatively high level of activity out in the cages, corresponding with a decrease in number of females in nest boxes and nest box entrances.

In the lactating period, where an effort is made to keep the female in a good body condition, it seems inappropriate to interrupt the resting period of the female by feeding when the temperature is normally highest. Ad libitum feeding with dry feed may prevent the increase in activity in the middle of the day and instead cause the mink to be active at a time more natural to them. Whether a change like that will balance out the advantages of feeding with fresh feed with a relatively high water content, has not been investigated.

The use of wire netting cylinders by mink females reduced stereotypic activity.

It is likely that installation of wire

netting cylinders in production cages with only one animal reduces stereotypic activity and thus reduces feed costs. Previous investigations (Jonge et al., 1985) have shown that on average females spend approximately 15% of their active time on stereotypic behaviour and that approximately half of the mink population spends more than 25% of their active time on this energy-consuming activity.

The results have been published in SCIENTIFUR (Hansen, 1990b).

2.5 Effect of water trays for farmed mink

2.5.1 Introduction

Under farm conditions, the need of mink for liquid is fulfilled by various sorts of water nipples. In the lactation period, dehydration of females may occur with fatal consequences. For kits a suboptimal water intake has been proved until weaning, and various auxiliary measures are used in this period (Møller & Lohi, 1988).

It has been proved that in the transition period (approximately 30 days old) between suckling and until they have learned to operate the water nipple, kits lick the female in the corner of her mouth, probably to get liquid.

By installing water trays in the cages, the kits will have an extra possibility of licking water from the pelt of the female, and thus the well-being of both female and kits should be optimized.

The aim of this project was to examine the effect on mink behaviour, physiological stress response and production variables of giving the animals access to water trays throughout a whole production year.

2.5.2 Materials and methods

After weaning at the age of 8 weeks, 156 mink kits of pastel type were in 1987 placed in pairs (male + female) in conventional mink cages (90 cm L x 30 cm W x 45 cm H) with and without water trays (30 cm L x 20 cm W x 2 cm H), respectively. The animals were fed daily at 0.15 p.m. and otherwise tended to according to normal farm routine.

The water trays were filled with water at 10 a.m. Monday-Friday. The kits were weighed once a month.

In November, the pelt of all mink was evaluated at normal live animal grading. The males were then pelted and the pelt graded subjectively. The females stayed in their cages. In the gestation period

(April) the females were observed at Scan-sampling every 20 minutes in 3 intervals at 8-9, 10-11 p.m. and 12-1 p.m.

To express the gain of the kits, they were weighed at the age of 4 and 8 weeks, respectively. The females were weighed immediately before weaning of the kits approximately at the age of 8 weeks.

Reproduction result expressed as living, dead and weaned kits was recorded. Immediately before whelping, half of the females with and without water trays, respectively, were tested for number of eosinophil leucocytes in the blood.

This procedure was repeated immediately before weaning of the kits.

2.5.3 Results and discussion

2.5.3.1 Weight development.

Mink with water trays had a lower gain than mink without water trays. Table 2.1 shows the weight development. An analysis of variance shows that the difference is significant for females ($p<0.05$) and highly significant for males ($p<0.001$). This result may be due to a higher activity level of these animals, better possibilities of movement and consequently a larger energy consumption.

Table 2.1 **Weight development of mink males and females with and without water trays (+/- watertr.) in the period from July 9th - October 28th, 1987.**

	Weight of male kits - g. ± SD		Weight of female kits - g. ± SD	
	-water tr.	+water tr.	-water tr.	+water tr.
09/07-87	868 ± 115	872 ± 103	642 ± 66	634 ± 77
05/08-87	1354 ± 123	1322 ± 125	841 ± 72	830 ± 94
10/09-87	1735 ± 160	1646 ± 171	948 ± 92	920 ± 114
30/09-87	2038 ± 202	1946 ± 218	1068 ± 126	1031 ± 125
28/10-87	2218 ± 212	2126 ± 221	1158 ± 136	1111 ± 138
Gain 09/07-28/10	1350 ± 190	1256 ± 173	516 ± 117	475 ± 106

Weight development of kits produced by females with and without water trays, respectively, appears from table 2.2.

Table 2.2 shows that there is no difference in kit weight at the age of 4 weeks, but at the age of 8 weeks kits without water trays have a higher weight than kits with water trays. A t-test shows that the difference is significant for male ($p<0.05$) as well as for female kits ($p<0.05$). There is no difference in the weight of mink females immediately before weaning of the kits.

Therefore, the use of water trays by the females has no positive effect on weight gain of the kits, while the kits are exclusively sucking. After the age of 4 weeks, when the kits are moving actively around in the cage, the negative effect of water trays must be caused by the fact that kits with water trays have a higher energy consumption for activity.

The possibility of the kits to supplement their water intake by licking water from the female's pelt in the nursing period did not reflect in kit gain, nor in female weight at weaning, or in relative survival of the kits (table 2.3) despite high temperatures in May and June in 1988.

Table 2.2 **Weight in grams ± SD of females with and without water trays and the weight in grams of their kits at the age of 4 and 8 weeks.**

	Weight	
	Kit age 4 weeks	Kit age 8 weeks
- water trays		
male kits	216 ± 47	681 ± 130
female kits	194 ± 51	514 ± 75
females		816 ± 126
+ water trays		
male kits	232 ± 126	627 ± 183
female kits	204 ± 92	568 ± 120
females		851 ± 100

Table 2.3 shows that there is a difference between the 2 groups with regard to litter size at birth as well as kit mortality. None of the differences are, however, statistically significant (Median-test).

Table 2.3 **Reproduction result ± SD for females placed in cages with and without water trays after weaning at the age of 8 weeks. Barren females and not-mated females have been excluded from the test material.**

	No. of kits - water trays		No. of kits + water trays	
	mean	SD	mean	SD
Kits, total no. born	5.91	2.28	5.20	2.06
Kits, stillborn	0.34	0.60	0.28	0.54
Kits, living	5.56	2.08	4.92	2.16
Kits, 3 days old	5.06	1.88	4.68	2.10
Kits, 7 days old	4.97	1.84	4.68	2.10
Kits, 21 days old	4.75	2.14	4.60	2.08
Kit loss, 0-3 weeks	1.16	1.57	0.60	1.00
Kit loss, 0-8 weeks	1.31	1.69	0.80	1.32

2.5.3.2 Skin qualities

The results of pelt grading of live animals appear from table 2.4.

Table 2.4 shows that mink without water trays have better pelts as regards clarity and quality than mink with access to water trays until November. The score at live animal grading was tested with Nonparametric test (Wilcoxon), $p<0.0001$.

Table 2.4 **Average scores for skin properties ± SD at live animal grading divided in males and females with and without water trays (Scores: 5 = best, 1 = worst).**

	Score for males		Score for females	
	-water tr.	+water tr.	-water tr.	+water tr.
Clarity	3.9 ± 0.7	3.4 ± 0.7	3.8 ± 0.5	3.4 ± 0.6
Quality	3.8 ± 0.9	3.0 ± 0.8	3.7 ± 0.8	3.4 ± 0.8
Density	4.0 ± 0.7	3.6 ± 0.8	3.5 ± 0.6	3.6 ± 0.9
Size	4.1 ± 0.5	3.9 ± 0.7	3.9 ± 0.7	3.7 ± 0.6

To investigate the effect of access to water trays on dried skins, male mink were pelted and the pelts treated traditionally. The results of skin grading appear from table 2.5.

Table 2.5 shows that males without water trays have clearer colour of fur than males with water trays. The difference was highly significant ($p<0.001$) in a nonparametric Wilcoxon test.

Table 2.5 **Average scores for skin properties and size in cm from male mink with and without water trays ± SD. Maximum scores are shown.**

	- Water trays			+ Water trays		
	Score	SD	Max.	Score	SD	Max.
Clarity	3.35	± 0.95	4	1.93	± 0.73	4
Quality	9.21	± 3.06	13	9.41	± 2.91	13
Colour	2.42	± 0.98	4	2.11	± 0.80	4
Size, cm	75.07	± 3.20	80	73.70	± 2.57	79

Both at live animal grading and skin grading the trait "clarity" was poorer in mink with access to water trays throughout the year than in the control group.

The reason may be a poorer micro climate in the nest box due to high-

er humidity, but insufficient cleaning of the trays may also have contributed. Defecation on the water trays was not a frequent occurrence, but the mink often drags the feed into the water tray, whereupon the feed is eaten.

The negative production results of installing water trays in mink cages must be seen on the basis that the animals have had water trays all the way until the time of pelting.

The use of water trays in limited periods of the production cycle may have an occupational and aggression-moderating influence on mink without negative effects on the final product. This has not been illustrated in this investigation and can therefore not be excluded.

2.5.3.3 Behaviour

Behavioural observations were made at the end of the gestation period, where the mink are relatively inactive. The behavioural elements, where differences between mink with and without water trays have been found, are "lying out in cage", "standing at cage door", and "total activity". The behavioural frequencies recorded are stated in table 2.6. The behavioural condition "on water tray" records that the mink is on the water tray, but no distinction was made whether the mink was resting or was active on the water tray.

When the water trays were filled with water at 10 a.m., the mink "bathed" which primarily consisted of scraping movements with their forefeet which were, later in the bathing session, followed by body rollings on the water tray. The bathing activities caused the trays to be emptied of water quickly, whereupon the mink used them as resting place and to eat their feed from.

The use of water trays as resting place may explain the difference observed in the behaviour "lying out in cage" between the two groups and indicates that mink prefer lying "up" instead of down on the wire bottom.

The increased frequency of the behaviour "standing at cage door" in mink with water trays, could be interpreted as increased curiosity, as this behaviour especially occurs in the period when water is given.

Table 2.6 **Average frequency of behaviour ± SD of mink with and without water trays and per cent distribution of these forms of behaviour.**

Forms of behaviour	+ Water trays mean	SD	%	- Water trays mean	SD	%	P-value (Wilcoxon)
In nest box	26.58	4.9	73.8	28.4	4.84	78.9	0.11
Active out not specific.	2.23	1.5	6.2	2.19	2.0	6.1	0.673
Marking beh.	0.33	0.4	0.6	0.33	0.6	0.9	0.549
Climbing in netting	0.32	0.6	0.9	0.31	0.7	0.9	0.748
Eating feed	0.55	0.7	1.5	0.64	0.7	0.8	0.557
Pendling	0.52	1.3	1.4	0.69	1.9	1.9	0.635
Active tot.	8.10	4.3	22.5	4.83	3.9	13.4	0.002
On water tray	2.77	2.1	7.7				
at 8-9 a.m.	0.58	0.7					
at 10-11 a.m.	1.26	0.9					
at 12-1 p.m.	0.94	1.2					
Lying out in cage	0.97	1.2	2.7	2.17	2.3	6.0	0.006
at 8-9 a.m.	0.26	0.5		0.61	1.1		
at 10-11 a.m.	0.13	0.4		0.22	0.4		
at 12-1 p.m.	0.58	0.9		1.33	1.3		
Standing at cage door	0.77	1.0	2.2	0.42	1.0	1.2	0.018
at 8-9 a.m.	0.13	0.3		0.14	0.4		
at 10-11 a.m.	0.65	1.0		0.28	0.7		
at 12-1 p.m.	0.00	0.0		0.00	0.0		
Stereotypic beh.	0.71	1.9	2.0	0.22	0.5	0.6	0.304
at 8-9 a.m.	0.19	0.8		0.06	0.2		
at 10-11 a.m.	0.42	1.1		0.06	0.2		
at 12-1 p.m.	0.10	0.3		0.11	0.4		
Lying out, licking their pelt	0.35	0.6	1.0	0.58	0.8	1.6	0.299
at 8-9 a.m.	0.13	0.4		0.19	0.5		
at 10-11 a.m.	0.03	0.2		0.06	0.2		
at 12-1 p.m.	0.19	0.4		0.33	0.7		

There is no difference between groups as regards "stereotypic" and "pendling" behaviour, which may be due to the large individual differences between these two forms of behaviour. Stereotypic behaviour is performed in 2% of the observations by mink with water trays and most frequently in the period when water is given, and in 0.6% by mink without water trays.

The form of behaviour "lying out and licking pelt" occurs most frequently in the periods 8-9 a.m. and 12-1 p.m. which are the periods of feeding. The first period when old feed is redistributed and the second period when new feed is given. The grooming behaviour, therefore, seems to be related more to the feeding situation than to the "bathing situation".

The use of water trays by mink does not in itself express a behavioural need, but the results show that mink like to use the water bath, and that they may have expectations of the recurring "pastime" offered by the bathing possibility.

If the bathing activity was a need of farmed mink, it could be expected that deprivation of this need would cause changes in the physiological stress level.

Table 2.6 shows that in approximately 75% of the observations the females are staying in their nest boxes. Females without water trays are lying more out on the bottom of the cages and are standing less at the cage door than females with water trays. Total activity is highest in cages with water trays. The other forms of behaviour recorded show no significant differences.

2.5.3.4 Level of eosinophil leucocytes

The level of eosinophil leucocytes that is known to reflect the physiological stress situation of mink under experimental conditions was therefore examined in half the females randomly selected from each of the two groups.

The eosinophil leucocyte level in females before whelping (25/04-88) and before weaning of kits (16/06-88), respectively, is shown in table 2.7.

Table 2.7 **Number of eosinophil leucocytes per mm^3 ± SD of females with and without water trays.**

	+ water trays			- water trays			
	N	Mean	SD	N	Mean	SD	(t-test)
25/04-88	18	604	250	20	585	289	ns
16/06-88	19	229	197	19	181	174	ns

The result showed that there was no difference in number of eosinophil leucocytes between the two groups. It has thus not been possible to prove that water trays have a stress reducing effect on farmed mink. The level of eosinophil leucocytes was significantly lower immediately before weaning of kits than immediately before time of birth. This is remarkable, as it should be assumed that mink females - immediately before weaning of the kits - are stressed owing to lack of opportunity to get away from the kits.

The results have been published at the Annual Report of the Danish Fur Breeders Association (Hansen, 1990a).

2.6 Correlation between behavioural response at stick test and eosinophil leucocyte level of mink females

2.6.1 Introduction

Experimentally induced stress at repeated immobilization of mink females in mink traps (Heller & Jeppesen, 1985), social stress (Heller, Jeppesen, 1986) and stress in connection with weaning (Jeppesen et al., 1988) cause an increase in the amount of circulating eosinophil leucocytes in the blood of mink. As regards behaviour, long-term stress results in a reduced exploratory (investigating) behaviour, reduced ag-gression and an increase in the motivation to run away (Heller & Jeppesen, 1985).

Through a deliberate selection towards aggressive and quiet mink, respectively, the behavioural response of the animals to human beings has been changed. These behavioural changes coincide with changes in the hypothalamus-hypophysis-adrenal cortex system (Namenko & Belyaev, 1980). Production animals with reduced fear of human beings have also been proved to have increased productivity (Seabrook, 1972).

The aim of the following investigation was to illustrate the correlation between behavioural response of farmed mink at the stick test and the physiological stress level expressed as number of circulating eosinophil leucocytes in the blood.

For quantification of behavioural response of mink the following test has been developed: The farmed mink was let out in the wire netting cage, and access to the nest box was blocked. The observer, placed in front of the cage, inserts in the wire netting of the cage door a tongue spatula and keeps it there for 30 seconds. If the mink approaches the "stick", sniffs at it and perhaps "chews at the stick", the behaviour is described as explorative. If the mink jumps at the stick and with considerable intensity bites constantly onto the stick, the reaction is described as "aggressive". If the mink moves away from the stick (quickly or slowly), the reaction is described as flight. A relatively rare reaction is seen when the animal stays in the same position (freezing). The reaction is then described as undetermined.

Besides the immediate reaction of the animal, also the period of latency for contact with the stick is recorded.

In September, the behaviour of 1128 female mink on 22 farms was tested at a stick test. Based on the response of the females, 195 were randomly selected for blood sampling as explorative, 218 as timid and 15 as aggressive female mink. The difference in number of eosinophil leucocytes between farms and between behaviour tested as well as the distribution of behavioural response was examined.

2.6.2 Results and discussion

The level of eosinophil leucocytes in the blood from females in the three behavioural categories is shown in table 2.8.

Table 2.8 **Number of eosinophil leucocytes per mm³ in the blood of females in the three behavioural groups.**

Behavioural response	Number	Mean	SD
"aggressive"	15	324	± 246
frightened	218	347	± 240
explorative	195	327	± 226
Totally	248	337	± 234

Results from the blood tests showed that there was no significant difference in number of eosinophil leucocytes in females responding either exploratively, frightfully or aggressively, respectively (table 2.8)

On the other hand a large variation in eosinophil level and behavioural response was found between farms. An analysis of variance confirmed that the variation in eosinophil level resulted from the difference between the individual farms ($p<0.001$), but the behavioural reaction to the stick test did not contribute significantly to this variation. The difference recorded between farms with regard to eosinophil leucocyte level and behaviour of mink has been illustrated in figures 2.1 and 2.2.

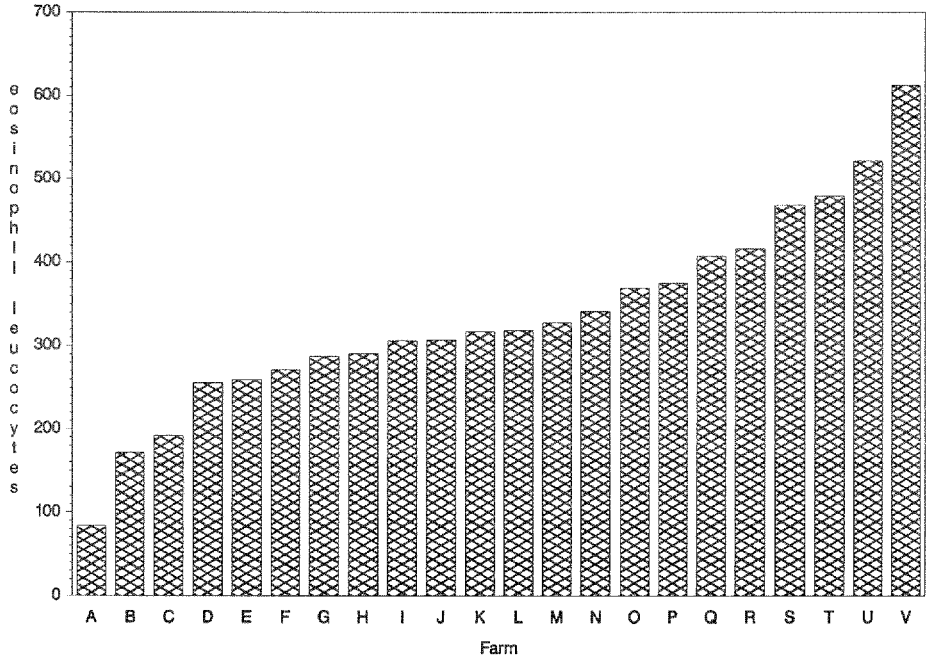

Figure 2.1 Average number of eosinophil leucocytes per mm³ per farm.

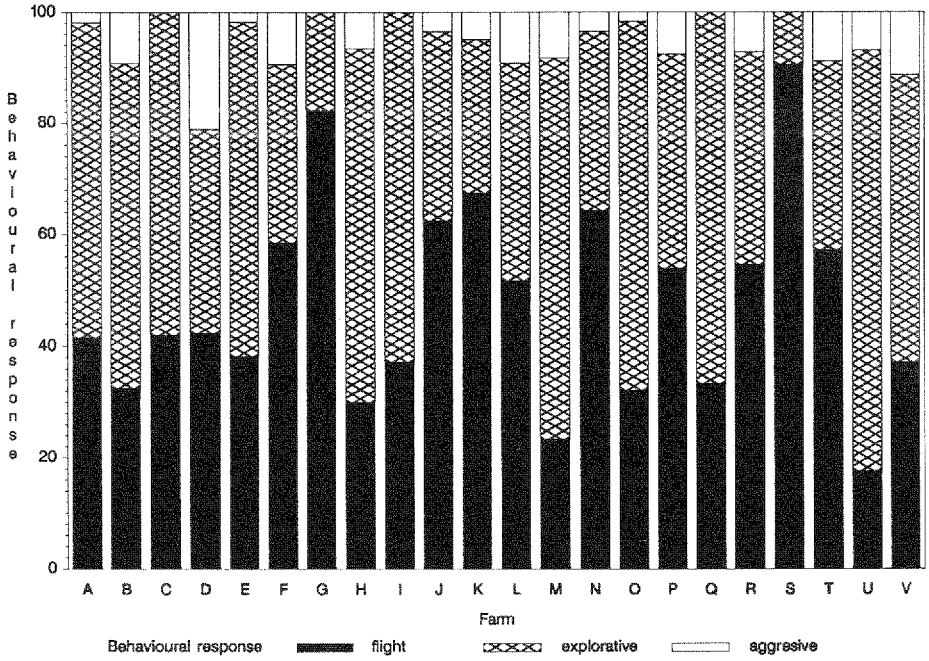

Figure 2.2. Per cent distribution of behavioural response per farm.

That the behavioural test does not reflect the physiological status of the animals, may be due to the fact that the two parameters express different sides of the adaptation of the mink to farm environment.

A high level of eosinophil leucocytes is an expression of stressing factors over a long period and depends on the total sum of stress factors. The behavioural response of the mink to the stick test is to a higher degree than the eosinophil level a result of the adaptation to and experience of the animals with human beings. Whether it is possible genetically to influence the temperament of mink, is being examined in a new project brought about by these investigations.

The results have been published in Short Comm. from the Natl. Inst. of Anim. Sci. (Hansen & Møller, 1988).

2.7 The importance of weaning time on mating success and temperament of mink

It has been proved that mink kits weaned at the age of 6 weeks and placed individually have a significantly reduced reproduction behaviour as adult mink compared to mink weaned at the age of 6 weeks and kept in groups (Bassett, 1959).

The purpose of this investigation was to describe mating success when mink were placed in pairs (male + female) after having been weaned at the age of 6, 8 and 12 weeks, respectively, and the importance of weaning time to temperament of the kits towards human beings.

One hundred pastel mink kits were divided in 3 groups and weaned at the age of 6, 8 and 12 weeks respectively, and placed in pairs in conventional mink cages. The mink were cared for according to normal farm routine. As a measure of mating willingness the average periods of latency for first neck bites and intromission, respectively, were recorded as well as per cent mating success in the weaning groups after 2 mating attempts. The results are shown in table 2.9.

Table 2.9 shows that mink males weaned at the age of 6 weeks have a shorter period of latency for first neck bite than males weaned at the age of 8 or 12 weeks (χ^2-test: $p<0.05$). No difference was found between weaning groups with regard to period of latency for intromission and per cent successful matings.

Table 2.9 **Period of latency for neckbite and intromission and per cent mating success in mink weaned at the age of 6, 8 or 12 weeks.**

Weaning age	No. of obs.	Period of latency (sec) neck bite	Period of latency (min) intromission	Mating success per cent
6 weeks	36	22 ± 34	16.3 ± 6.6	30.6
8 weeks	49	57 ± 123	19.4 ± 5.0	49.0
12 weeks	25	63 ± 122	21.6 ± 10.0	40.0

The result shows that the physical contact between kits placed in pairs in conventional cages is sufficient to secure normal reproduction behaviour, irrespective of weaning age.

The results have been presented at the Annual Meeting of the Natl. Inst. of Anim. Sci. (Hansen, 1987a).

To measure the effect of weaning time on later temperament of mink, the mink were stick tested in the period from January 6th to March 6th.

Mink that stayed in the nest box during the test or that could not be classified in one of the three categories have been left out of the table. The results are shown in table 2.10.

Table 2.10 The proportion of mink weaned at the age of 6, 8 or 12 weeks which were at the stick test classified as either aggressive, frightened or explorative.

	Jan. 6th	Jan. 22nd	Feb. 3rd	Feb. 27th	Mar. 6th
Aggressive					
6 weeks	30	52	70	67	60
8 weeks	20	39	47	51	55
12 weeks	0	10	25	40	35
Frightened					
6 weeks	7	7	0	0	0
8 weeks	12	8	6	6	0
12 weeks	20	5	0	1	0
Explorative					
6 weeks	36	17	10	20	20
8 weeks	41	27	20	20	16
12 weeks	65	65	55	40	45

It appears from the table that the proportion of animals reacting aggressively increases with time. The increase in aggression may perhaps be due to the coming mating period in March or to the feeding routine in the winter months where the feed ration of the mink is decreased. Mink weaned at the age of 12 weeks react less aggressively and more exploratively than mink weaned at the age of 6 or 8 weeks, respectively.

The results were published at the Annual Meeting of the Natl. Inst. of Anim. Sci. (Hansen, 1987b).

On basis of the immediate reactions at weaning (moving in and out of the nest box by the female, calling sounds from the kits and changes in number of eosinophil leucocytes), Houbak and Jeppesen (1988) showed that mink kits weaned individually consider weaning as a much more stressing factor than kits weaned in groups. Kits weaned at the age of 6 weeks are more stressed than kits weaned later. From the female's point of view, weaning after a kit age of 6 weeks and before they are 10 weeks old must be regarded as optimal. Naturally, reservations must be made for large litters and nursing disease.

2.8 Theses from the University of Copenhagen, Inst. of Population Biology, connected with this project

2.8.1 The importance of early weaning and handling to later timidity of mink (Birgitte Damsgaard, 1987)

One hundred mink kits were weaned at the age of 40, 47 and 54 days. For five days after weaning, half of the kits from each weaned team were subjected to short handling three times daily.

When the kits were 15 and 22 weeks old, respectively, their reaction to human beings was tested by letting a person put his hand with a leather glove into the cage. At the age of 15 weeks, the reaction of the mink to an unknown object was tested.

The result has shown:

- that the handling practised had no influence on the timidity of the mink,
- that the sensitive period of the mink to primary socialization ends between 40 and 47 days after birth, and that kits weaned before this time will be less frightened of human beings,

- that the reduced fear is especially valid in connection with human beings, and that it is not a generally reduced fear, as there was no difference in the reaction of the animals towards an unknown object.

2.8.2 The ontogeny of mink kits (Birthe Jonasen, 1987)

The ontogeny of mink kits has been investigated in the following manner:

A) Testing of 116 kits at the age from 1 to 35 days with regard to locomotive skills, development of the sensory apparatus, utterances of sound as well as requirement for contact.

B) Scanning observations of 54 litters of mink kits with regard to activity, locomotive skills, development of sight, saliva licking and play fighting as a function of age.

C) Comparison between two litters of mink kits raised under seminatural condition and two litters raised under normal farm condition.

The investigations have given a good description of the ontogenetic development of mink kits under conventional farm condition.

It has not been possible to show any statistical difference in the ontogenetic development between mink kits raised under normal farm conditions and mink raised in seminatural conditions.

2.8.3 The development of behaviour of juvenile mink in cages and the importance of stress to behaviour, physiology and fur quality (Frank Dodd, 1985)

The development of behaviour was investigated in juvenile caged mink which had, until the age of 9 weeks, been living in different environments, but were then placed on the same farm.

The development showed that as a result of their natural maturing, the animals showed a reduction in play behaviour corresponding with an increase in agonistic behaviour in weeks 11 to 16 after birth. The increase in agonistic behaviour (especially in connection with the feeding situation) and the lack of escape possibilities resulted in a conflict situation which presumably induces the development of stereotypic behaviour.

The agonistic behaviour influenced marking behaviour and this proved to be dominance-dependent as it disappeared during establishment of relations of dom-

inance in week 19 and later on returned at an increased frequency in week 22.

The eosinophil level of females was higher than that of males, possibly due to the subordinate status of the females. Correlation between behaviour and eosinophil level indicates that a high motory activity may reduce the physiological stress level.

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3. MINERAL CONTENT OF FEED AND HAIR AND MICROSCOPIC STUDIES ON HAIR.

by Outi Lohi, Palle Vistisen Rasmussen and Lone Vejgaard Jensen

A special part of this project was to investigate the mineral content of mink hair and its relation to minerals in feed. The pelt material collected for mineral analyses was also used for microscopic studies on hair in order to get a detailed description of variation of hair size and shape and to study possible relationships between mineral content and hair morphology.

The following two investigations deal with these aspects.

Summary

The mineral content of mink feed was analysed in weekly samples from five feed kitchens in the period from August 15th until December. The daily fluctuation was studied within two weeks. Feed values were compared with pelt characteristics and with the mineral content in hair of scanblack male pelts from customer farms. Furthermore objective measure of fur weight and morphometric characteristics of guard hairs were compared with the mineral content in hair.

There was a clear correlation between the amount of Ca, Mg, Na and Se in feed and in hair. The levels of Zn, Cu and P in feed were not reflected in the hair. Pelt quality was not related to minerals in feed or hair but hair colour showed significant correlation to the content of Ca, Mg and P in hair. Interactions between minerals play an important role in mineral metabolism and in organ concentrations.

Great variation could be shown in guard hair types on pelts from different farm populations. Of single minerals only Ca showed correlation to morphometric characters of hair. Hair samples from the hip region expressed more clearly differences in pelt quality than samples from the center of the back.

Sammendrag

Mineralindholdet i foder blev kontrolleret med ugentlige foderprøver fra den 15. august til december. Dag til dag variationen blev undersøgt i to uger. Indholdet i foder blev sammenholdt med pelskaraktererne og med mineralindholdet i hår af scanblack skind samlet fra fodercentralernes kundefarme. Desuden blev vægten af hår per skindareal og længde og tykkelse af dækhår sammenlignet med pelsbedømmelsen og hårets mineralindhold.

Der blev fundet korrelation mellem foder og hår m.h.t. mineralerne Ca, Mg, Na og Se. Mængden af Zn, Cu og P i foder havde ikke effekt på mængden i hår.

Kvaliteten var ikke korreleret til mineralindholdet i foder eller hår, men hårfarven viste signifikant korrelation til hårets indhold af Ca, Mg og P. Det viste sig, at interaktioner mellem mineraler spiller en stor rolle i mineralmetabolismen og i lagring af mineraler i organerne.

Morfometriske undersøgelser viste store variationer i hårtype mellem forskellige dyrepopulationer. Af enkelte mineraler viste kun Ca korrelation til hårmassen og morfometriske mål af hår. Hårprøver fra hoften giver et bedre udtryk for pelskvaliteten end hårprøver fra ryglinjen.

3.1 MINERAL COMPOSITION OF MINK FEED AND MINK HAIR

By Outi Lohi and Lone Vejgaard Jensen

3.1.1 Introduction

In addition to the main nutrients, protein, fat and carbohydrates, macro- and micro-minerals are an essential part of animal diets. In mink feed the minerals derive partly from different feed ingredients and partly from special mineral supplements. Thus the feed kitchens and farmers try to secure that an adequate amount is always available for the animals.

However, very little is known about the requirements of fur animals and even less about the factors affecting availability and utilization of the minerals.

From humans and from other animal species it is known that excess of minerals in food can cause high concentration in hair and that deficiency of certain minerals can also be detected by hair analyses. Besides the intake of minerals, concentration in hair also depends on hair cycle, sample location and hair colour (Kossila et al., 1972; Berestov et al., 1985; Lohi, 1987). Therefore, in order to know when the level of minerals in the hair is an expression of deficiency in feed or health problems, it is necessary to know the normal variation in mineral concentration.

The aim of this investigation has therefore been to study the variation of mineral content in Danish mink feed in the period from August until pelting and the relation of minerals in feed to the mineral content of hair on animals fed with these diets during the main growing phase of the hair. Furthermore to study the relationship of minerals to fur quality.

3.1.2 Material and methods

The feed samples were collected from five Danish feed kitchens located one in Zealand, two in North-Jutland, one in Mid-Jutland and one in South-Jutland. From the 15th of August until pelting a small sample of the daily production was frozen six times a week. Before analysing, the six daily samples were in the laboratory mixed to form a sample representing the weekly production.

To get an impression of the day to day variation in the mineral content of the feed, the individual daily samples from two weeks (No. 41 and 44) were analysed.

Feed samples were analysed for minerals Ca, P, Mg, K, Na, Zn, Fe, Cu, Mn, Sr and Se.

The laboratory treatment of both feed and hair samples and the analytical methods are in detail described by Vejgaard Jensen et al., (1988).

In addition to the feed, animals also consume a lot of water. The total daily intake has been measured to be about 200 to 250 ml per mink (110 g/kg body weight) (Kangas, 1973). When using fresh feed, about three quarters of this comes from the feed, but the rest, 50 to 150 ml, is drinking water.

If the content of minerals in drinking water is high, it could make additional differences in mineral intake between farms. In order to control this, samples of the drinking water were collected from each farm in August 1985.

As scanblack type (dark mink) is one of the main colour types in Danish mink production, it was decided to use this colour type for hair investigations. To collect a representative range of samples, four farms were chosen among the clients of each feed kitchen and 15 scanblack male pelts were selected from their production the same year.

The sample pelts were selected to represent three quality groups (5 pelts per group):

1. Saga selected/Saga
2. Quality I
3. Pelts with fur defect metallic.

To eliminate the effect of colour intensity, the aim was to select all pelts in the same colour group. Because of the great variation in the scanblack type between different farms, this proved impossible in practice. However, within each farm the colour variation was minimized. For the mineral analyses, a 4 cm wide area was shaved from the right side of the back from 1 cm above the tail until the level of the front legs. The sample was analysed as a whole without separating guard hairs from underfur.

To get a detailed description of the quality and the colour, the pelts were graded according to the normal subjective grading system for research pelts for traits quality (ranging from 1 to 15), density of hair (points 1 to 5), colour (points 1 to 10) and fur defect metallic (points 0 to 4). The colour group was determined by the optic colour grading unit of the Danish Fur Auctions.

The thickness of the leather was measured with a micrometer screw at two places on the skin about 10 cm from the tail root, in the middle

of the back and in the middle of the shaven area. It is, however, important to notify that the leather thickness on a dry pelt is a result of both the real leather thickness and the fleshing method. The figures therefore have to be considered with some reservation.

Hair samples were analysed for minerals Ca, P, Mg, K, Na, Zn, Fe, Cu and Se.

3.1.3 Results and discussion

3.1.3.1 Mineral composition of Danish mink feed.

Larger studies on the mineral content of fur animal feed have previously been done by Kangas (1974); Nielsen (1975), and Hansen (1986). The results of this investigation are in table 3.1 compared to these previous studies. Since the 1970's, the ingredients used in fur animal diets in the Scandinavian countries have changed. Slaughter house by-products and cod have more and more been exchanged to small fish and fish silage. At the same time also the use of dry ingredients like fishmeal, soybean meal etc. has increased. Because of these changes, mink feed today has a higher dry matter, fat and energy content than in the 1970's. Thus even though the "as fed" basis concentration of minerals is more or less unaltered, the concentration per dry matter or per energy is lower than 15 years ago.

Table 3.1 Mineral content of mink feed in different surveys. Data on "as fed" basis.

Literature		(1)	(2)	(3)	(4)	(5)
Year		1969 April-June	1966-71	1984	1985 Jan.-June	1985 Aug.-Dec.
Country		DK	SF	DK	DK	DK
Ca	g/kg	11.0 (6.6-15.0)	7.3	(6-8)	12.9 (11.7-13.3)	7.6 (5.5-10.3)
P	g/kg	7.3 (5.4-9.0)	4.4	(0.4-0.6)	7.5 (7.2-8.1)	5.1 (4.3-6.1)
Mg	g/kg	0.5 (0.4-0.6)	0.5	(0.4-0.6)	0.5 (0.5-0.6)	0.6 (0.5-0.6)
Na	g/kg	1.9 (1.5-3.2)		(1.2-1.4)	1.5 (1.3-1.6)	1.3 (0.9-1.6)
K	g/kg	2.6 (2.4-3.0)		(2.0-3.0)	2.9 (2.8-3.1)	2.9 (2.7-3.0)
Fe	mg/kg	202 (106-350)	70	(75-226)	235 (154-279)	185 (99-227)
Cu	mg/kg	11 (4-23)	3.2	(5-20)	10 (9-11)	8 (6-10)
Zn	mg/kg		17	(25-69)	37 (26-42)	43 (29-65)
Se	mg/kg			(0.27-0.36)		0.3 (0.25-0.38)

(1) Nielsen 1975, (2) Kangas 1974, (3) Glem-Hansen 1984,
(4) Hansen 1986, (5) Present investigation.

However, compared to the Scandinavian minimum requirements (Glem-Hansen, 1984) both Ca and P well exceed the required amount. The Ca:P ratio is within the accepted range (1.0 - 1.7). Fe, Cu and Zn also well exceed the requested level.

The mean values per feed kitchen for the total investigation period are given in table 3.2, and the variation from August until pelting time in table 3.3.

Table 3.2 **Mineral content of mink feed. Feed kitchen means for the period of 16 weeks. Data on dry weight basis.**

Feed kitchen	Ca g/kg	P g/kg	Mg g/kg	Na g/kg	K g/kg	Fe mg/kg	Zn mg/kg
101	17.8	12.8	1.4	2.9	7.1	433	73
214	18.6	12.0	1.4	3.7	7.0	571	89
215	25.9	15.4	1.6	4.0	7.6	568	101
301	14.0	11.1	1.5	2.5	7.6	271	167
401	19.4	12.7	1.5	3.3	7.1	521	111

Feed kitchen	Cu mg/kg	Se mg/kg	Mn mg/kg	Sr mg/kg	Ca/P ratio	Dry matter
101	16.0	0.94	61	55	1.4	39.9
214	24.1	0.73	86	54	1.5	38.6
215	24.0	0.82	105	73	1.7	39.9
301	18.6	0.65	63	54	1.2	38.9
401	19.1	0.69	106	52	1.5	39.9

The greatest variation between feed kitchens and between periods is found in Fe, Zn, Mn and Ca contents whereas P, Mg and K show little variation. In general, feed kitchen No. 215 has a high level of all minerals. Through the whole period feed kitchen No. 301 is low in Ca, P, Na and Fe but has fed high levels of Zn compared to other kitchens. Feed kitchens 215 and 401 have higher levels of Mn than other feed kitchens.

The changes in mineral concentration during the investigation period are illustrated in figures 3.1.a - 3.1.k (appendix).

Table 3.3 Mineral content of mink feed on dry weight basis. (Part 1.)

	Weeks:	34 - 37	38 - 41	42 - 45	46 - 49
	F.K.	$\bar{x} \pm$ SD	$\bar{x} \pm$ SD	$\bar{x} \pm$ SD	$\bar{x} \pm$ SD
Ca:	101	18.0 ± 1.0	17.7 ± 1.0	15.7 ± 1.1	20.0 ± 2.9
	214	17.4 ± 0.9	16.6 ± 1.9	20.1 ± 0.6	20.0 ± 1.4
g/kg	215	24.2 ± 1.0	24.7 ± 0.8	26.4 ± 0.7	28.4 ± 3.9
	301	15.2 ± 1.6	15.1 ± 1.3	13.1 ± 2.5	12.6 ± 2.5
	401	19.9 ± 0.8	20.1 ± 1.2	18.5 ± 0.8	19.1 ± 0.7
P:	101	12.9 ± 0.5	12.7 ± 0.3	12.0 ± 0.8	13.6 ± 1.3
	214	11.5 ± 0.5	11.0 ± 0.7	12.8 ± 0.7	12.9 ± 0.8
g/kg	215	14.6 ± 0.3	15.0 ± 0.5	15.6 ± 0.5	16.5 ± 2.0
	301	12.0 ± 0.9	11.6 ± 0.7	10.8 ± 1.5	10.2 ± 1.4
	401	13.4 ± 0.5	12.9 ± 0.7	12.1 ± 0.2	12.5 ± 0.4
Mg:	101	1.4 ± 0.03	1.4 ± 0.01	1.3 ± 0.09	1.4 ± 0.02
	214	1.4 ± 0.02	1.4 ± 0.04	1.4 ± 0.07	1.5 ± 0.03
g/kg	215	1.7 ± 0.02	1.7 ± 0.06	1.6 ± 0.10	1.6 ± 0.07
	301	1.5 ± 0.03	1.5 ± 0.07	1.6 ± 0.13	1.5 ± 0.04
	401	1.5 ± 0.05	1.5 ± 0.04	1.4 ± 0.04	1.5 ± 0.07
Na:	101	2.8 ± 0.1	3.2 ± 0.2	2.9 ± 0.2	2.7 ± 0.4
	214	3.6 ± 0.1	3.6 ± 0.2	3.7 ± 0.1	3.9 ± 0.1
g/kg	215	4.4 ± 0.1	4.1 ± 0.2	3.8 ± 0.2	3.7 ± 0.2
	301	2.3 ± 0.4	2.3 ± 0.1	2.1 ± 0.1	3.2 ± 0.8
	401	3.5 ± 0.1	3.5 ± 0.2	3.2 ± 0.6	3.1 ± 0.4
K:	101	7.0 ± 0.3	7.3 ± 0.4	6.8 ± 0.3	7.1 ± 0.3
	214	7.5 ± 0.1	6.8 ± 0.4	6.5 ± 0.2	7.0 ± 0.3
g/kg	215	7.8 ± 0.1	8.1 ± 0.3	7.3 ± 0.4	7.2 ± 0.3
	301	7.7 ± 0.1	7.7 ± 0.3	7.7 ± 0.3	7.5 ± 0.6
	401	7.1 ± 0.1	7.0 ± 0.1	6.7 ± 0.7	7.5 ± 0.6
Fe:	101	455 ± 15	453 ± 34	397 ± 25	429 ± 51
	214	558 ± 33	533 ± 20	567 ± 88	624 ± 46
mg/kg	215	663 ± 68	597 ± 50	516 ± 21	495 ± 20
	301	274 ± 31	272 ± 11	254 ± 27	281 ± 26
	401	626 ± 175	512 ± 26	505 ± 69	441 ± 112

Table 3.3 **Mineral content of mink feed on dry weight basis. (Part 2.)**

	Weeks:	34 - 37	38 - 41	42 - 45	46 - 49
	F.K.	$\bar{x} \pm$ SD	$\bar{x} \pm$ SD	$\bar{x} \pm$ SD	$\bar{x} \pm$ SD
Zn:	101	79 ± 2	79 ± 5	66 ± 4	70 ± 6
	214	88 ± 3	91 ± 4	93 ± 3	87 ± 5
mg/kg	215	103 ± 3	111 ± 7	99 ± 6	92 ± 8
	301	173 ± 18	141 ± 51	196 ± 2	156 ± 61
	401	139 ± 13	119 ± 11	98 ± 5	94 ± 7
Cu:	101	16 ± 4	15 ± 0.8	17 ± 2	16 ± 2
	214	27 ± 2	24 ± 3.5	23 ± 4	23 ± 1
mg/kg	215	39 ± 12	29 ± 2.6	11 ± 3	17 ± 6
	301	18 ± 10	15 ± 5.7	19 ± 2	23 ± 2
	401	23 ± 6	19 ± 0.8	12 ± 4	23 ± 4
Mn:	101	62 ± 1	61 ± 2	58 ± 3	64 ± 6
	214	83 ± 1	82 ± 3	88 ± 8	92 ± 6
mg/kg	215	109 ± 1	113 ± 15	104 ± 11	96 ± 7
	301	61 ± 5	64 ± 2	61 ± 1	68 ± 4
	401	110 ± 5	108 ± 9	102 ± 5	102 ± 8
Sr:	101	58 ± 4	54 ± 4	48 ± 5	60 ± 8
	214	47 ± 4	46 ± 9	63 ± 2	62 ± 6
mg/kg	215	67 ± 4	67 ± 2	76 ± 4	83 ± 13
	301	58 ± 5	58 ± 6	49 ± 12	49 ± 13
	401	57 ± 3	53 ± 3	49 ± 3	51 ± 4
Se:	101	1.0 ± 0.06	1.0 ± 0.03	0.9 ± 0.03	0.9 ± 0.11
	214	0.7 ± 0.05	0.7 ± 0.01	0.8 ± 0.02	0.7 ± 0.06
mg/kg	215	0.9 ± 0.02	0.9 ± 0.08	0.8 ± 0.09	0.7 ± 0.08
	301	0.7 ± 0.04	0.6 ± 0.05	0.6 ± 0.05	0.7 ± 0.04
	401	0.8 ± 0.19	0.7 ± 0.03	0.6 ± 0.04	0.6 ± 0.09

The daily changes in mineral content were controlled with samples from weeks 41 and 44. Occasionally the variation from day to day is large. To a certain degree such short term changes are a result of the natural variation in fresh feed ingredients. However, in extreme cases they are obviously due to careless dosing of mineral supplements. This experiment showed that large variation can occur at short term. Mineral investigations on fresh feed should therefore never be based on single day feed samples. An illustration of the range of variation is in regard to Se content given in figure 3.2.

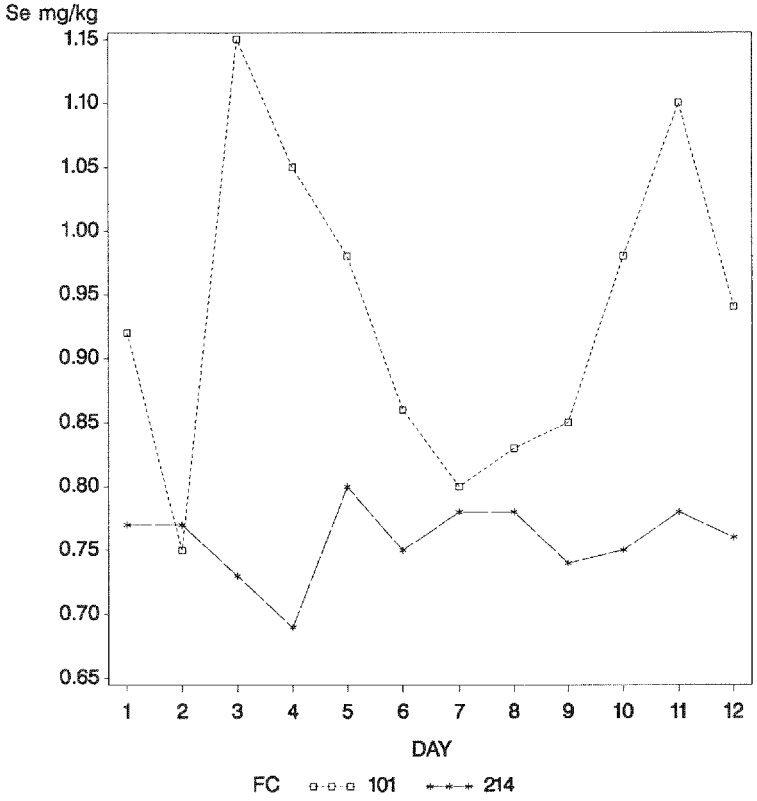

Figure 3.2 **Day to day fluctuation in Se content of feed on two feed kitchens (dry matter basis).**

Additional variation in the total amount of minerals the animals have consumed during the investigation period is caused by the differences in feed consumption. Calculations based on 10 farms, where data of the daily delivered amount of feed were available, are shown in table 3.4.

After passing through the alimentary tract, minerals either end in different tissues or are excreted in urine and faeces. In this investigation only the content in hair was analysed, and therefore the material does not allow conclusions about the effects in other tissues or changes in excretion.

3.1.3.2 Minerals in drinking water.

All farms on Zealand (feed kitchen 101), three farms from Northeast-Jutland (feed kitchen 214) and one farm in South-Jutland (feed kitchen 401) were connected to the municipal water system. All others had own water supply.

The results of the water analyses are presented in table 3.5.

In general, mineral contents were very low. On two farms sodium content was exceptionally high and three farms had high amounts of potassium in the water. The reason for these variations is unknown.

A local area in South-Jutland and a single farm in Mid-Jutland have noticeable amounts of manganese. Calcium levels vary from 8 to 173 mg per liter and magnesium from 2.6 to 26 mg per liter. High magnesium is more typical of farms from Zealand (feed kitchen 101). Calcium is high both on Zealand and in Northwest-Jutland (feed kitchen 215).

Even with the highest contents found in water samples, the intake of minerals through water is, however, below 6 percent of amounts obtained in feed.

The only exceptions are the two farms with high sodium content and the one with high level of manganese. In these cases sodium in water can amount to as much as 20-25 percent and manganese as much as 10 percent of the total intake. However, sodium content in hair samples from these two farms was not abnormal. Manganese was not determined in the hair samples.

Table 3.4 Variation in mineral consumption between 10 farms based on farmers' information about feed consumption.

Calculated total amount of minerals per animal in period August-December.

Feed kitchen	Farm	Ca g	P g	Mg g	Na g	K g	Fe mg
101	133	93	68	7.4	16.2	38.0	2.3
101	134	91	66	7.2	15.8	37.1	2.3
214	61	92	60	7.2	18.6	35.4	2.7
301	10	66	52	7.0	9.9	34.7	1.1
301	27	68	53	7.2	10.2	35.7	1.1
301	31	70	55	7.4	10.5	36.7	1.2
301	52	65	51	6.9	9.8	34.3	1.1
301	173	69	55	7.4	10.4	36.5	1.2
401	150	109	72	8.2	19.3	38.3	3.1
401	155	110	72	8.3	19.5	38.7	3.1

Feed kitchen	Farm	Zn mg	Cu mg	Mn mg	Sr mg	Se mg
101	133	401	82	322	289	5.1
101	134	393	81	315	282	4.9
214	61	462	123	427	262	3.8
301	10	751	74	282	254	2.9
301	27	772	76	289	260	3.0
301	31	791	78	298	268	3.0
301	52	747	74	278	251	2.8
301	173	789	78	296	266	3.0
401	150	662	100	596	293	3.8
401	155	669	101	602	296	3.9

Table 3.5 **Mineral content of drinking water (mg/liter) on farms included in the investigation.**

Farm	Feed kitchen	mg/liter Ca	Mg	Na	K	Fe	Zn	Cu	Mn	B
132	101	100	16.9	24	3.0	0.012	0.007	<0.005	<0.002	0.02
133	101	68	26.0	26	4.2	0.003	0.160	0.036	<0.002	0.08
134	101	8	13.0	170	16.1	0.003	0.008	0.016	<0.002	0.36
138	101	29	7.7	200	4.8	0.020	0.030	0.007	<0.002	0.47
272	101	90	22.0	18	4.6	0.028	0.160	0.032	<0.002	0.14
29	214	49	5.9	19	1.1	0.005	0.015	<0.005	<0.002	<0.02
61	214	34	4.9	16	1.6	0.007	0.210	0.013	<0.002	<0.02
198	214	34	4.3	14	1.2	0.012	0.015	<0.005	<0.002	<0.02
299	214	35	5.3	18	1.8	0.031	0.011	<0.005	<0.002	<0.02
35	215	62	4.9	22	1.2	0.005	0.150	<0.005	0.063	<0.02
51	215	93	6.8	22	2.3	0.054	0.080	<0.005	<0.002	<0.02
276	215	118	7.4	29	11.9	0.080	0.041	<0.005	0.056	0.02
326	215	55	3.9	20	1.6	<0.002	0.320	<0.005	<0.002	<0.02
10	301	37	3.6	14	0.7	0.002	0.062	<0.005	<0.002	<0.02
27	301	30	13.0	16	2.3	0.010	0.130	<0.005	0.044	<0.02
31	301	13	4.6	26	15.7	0.016	0.017	0.008	0.039	<0.02
52	301	38	4.6	13	1.7	0.15*	0.014	<0.005	0.52	<0.02
173	301	58	2.6	16	1.0	0.004	0.086	0.260	<0.002	<0.02
150	401	40	2.9	14	6.6	0.056	0.220	0.012	0.22	<0.02
151	401	57	12.2	13	4.8	0.002	0.016	<0.005	<0.002	0.06
152	401	54	4.8	11	3.5	0.022	0.023	0.005	0.250	<0.02
155	401	33	6.5	11	3.1	0.190	0.460	0.020	0.390	<0.02
156	401	173	8.0	14	1.1	0.028	0.091	<0.005	2.100	<0.02
362	401	42	6.9	12	4.3	0.090	0.018	<0.005	0.100	0.04

* In water phase. After dissolving the sediment with HCl = 10 mg/liter.

3.1.3.3 Minerals in hair and correlation between feed and hair.

The average amounts of minerals in feed and hair are shown in table 3.6. The only mineral, where the concentration in hair is higher than in the feed, is Zn. The other minerals, which to a high degree seem to concentrate into hair, are Cu, Na and Se.

Correlation and regression analyses were used to test the role of feed for the mineral content of the hair. The statistics are run both based on mineral content per dry matter and per metabolisable energy. In the total material correla-

tions were significant for Ca, Mg, Na and Se which agrees well with other investigations (Kossila et al., 1972; Hornshaw et al., 1985).

Table 3.6 Mean content of minerals in feed and hair samples.

Mineral	Minerals in feed N = 80 mg/kg dry matter	Minerals in hair Scanblack male pelts N = 300 mg/kg dry hair
Calcium	19130	1301
Phosphorus	12810	433
Magnesium	1480	235
Sodium	3280	747
Potassium	7260	554
Zink	108	249
Iron	475	17
Copper	20	8
Selenium	1	1
Sum	44564	3545

In regard to Zn the earlier investigations have given contradictory results. In our investigation the levels of Zn, Cu and P in feed were not reflected in hair. Slightly elevated potassium levels were found in hair samples from feed kitchens 215 and 301, which also had high levels in feed. However, higher level in hair also applies to kitchen 214 even though the content of K in feed was low. Neither did we find the interaction between Zn in feed and K or Mg in hair reported by Combs et al., (1982) and Hornshaw et al., (1985). However, this only confirms, as other researchers have also pointed out, that metabolism of Zn and especially its interactions with other minerals need further studies under controlled conditions.

The iron content of feed seems to have an effect on the amount of Fe in hair even though a clear correlation could not be detected. The results also raise the question of interaction between Zn and Fe as reported by Hornshaw et al. (1985). The "normal" content of Fe in hair samples from feed kitchen 301, even though the content in feed was low compared to other kitchens, could be explained by a favourable effect of a high Zn

level on Fe metabolism.

Even though the amount of minerals in feed has a significant effect on the content in hair in all cases, mineral levels in feed only explain a small part of the variation in hair. In many cases a considerable part of the variation seems to be due to some pelt characteristics and intake of other minerals.

The mean content of minerals in hair per farm is presented in table 3.7.

Correlations between the content of different minerals in feed and hair point out possible interactions between several minerals. The cases where mineral levels in feed show significant correlation to the level of different minerals in hair or have relation to hair colour are presented in table 3.8.

Table 3.7 Mineral content of hair from scanblack pelts. Average of 15 pelts per farm. F1 = hair colour.

F.K./ Farm	F1	mg/kg hair								
		Ca	P	Mg	Na	K	Fe	Zn	Cu	Se
101:										
132	8.93	1420	478	295	744	557	23	204	8.9	1.3
133	1.20	941	360	161	688	433	22	234	8.3	1.1
134	3.93	1357	430	211	578	393	16	282	8.1	1.3
138	4.73	1327	459	240	555	386	19	214	8.0	1.2
214:										
29	3.93	1225	430	225	725	722	33	301	9.0	1.1
61	6.53	1339	455	218	692	996	16	285	8.0	1.0
198	4.67	1248	398	232	899	738	14	323	8.8	1.1
299	6.40	1285	479	227	783	543	13	250	8.4	0.9
215:										
35	4.73	1386	428	263	918	588	15	225	8.6	1.0
51	8.07	1582	468	283	728	433	21	207	8.6	1.0
276	8.67	1569	445	309	1105	606	20	214	8.6	1.0
326	7.87	1333	442	284	985	621	13	253	8.5	1.0
301:										
10	4.20	1205	433	210	801	626	14	269	8.7	1.0
27	5.80	1325	437	241	709	553	13	283	7.5	1.2
31	6.27	1389	449	246	729	547	13	223	8.5	1.0
173	1.87	929	383	160	605	429	13	271	8.1	1.0
401:										
150	4.13	1223	435	197	782	486	15	254	8.1	1.0
152	8.33	1423	395	235	396	353	15	225	9.5	1.1
155	4.80	1235	425	224	691	547	19	220	8.9	1.1
156	5.33	1287	423	238	835	530	19	236	8.7	1.0

Table 3.8 Minerals in mink hair affected by mineral intake and/or hair colour.

Correlation coefficients and significance

Minerals in hair	Mineral intake in feed: Ca	P	Mg	Na	K	Fe
Ca	0.41***	0.41***	0.37***	0.34***	0.38***	0.28***
P	0.13*	0.12*		0.13*		
Mg	0.46***	0.47***	0.43***	0.37***	0.44***	0.29***
Na	0.40***	0.38***	0.45***	0.37***	0.45***	0.26***
K			0.13*	0.23***	0.12*	0.20***
Fe				0.13*		0.18**
Zn						
Cu	0.18**	0.17**	0.17**	0.21***	0.16**	0.23***
Se			-0.33***	-0.19**	-0.32***	

	Mineral intake in feed: Zn	Cu	Se	Mn	Sr	Hair colour+
Ca		0.29***	0.24***	0.30***	0.42***	0.70***
P		0.15**	0.12*		0.15*	0.46***
Mg		0.32***	0.30***	0.30***	0.51***	0.70***
Na		0.42***	0.13*	0.29***	0.47***	0.21***
K		0.38***				0.14*
Fe	-0.30***		0.25***			
Zn						
Cu		0.16**		0.25***		
Se	-0.36***	-0.35***	0.33***	-0.27***	-0.15**	

* = $p<0.05$

** = $p<0.01$

*** = $p<0.001$

+ Objective measure of colour.

3.1.3.4 Minerals in hair and grading characteristics.

No difference in hair minerals could be detected between the three quality groups of skins within feed kitchens. Nor were correlations between hair quality and minerals in hair significant even though a more detailed graduation of quality was included in calculations.

More interesting is, however, the relationship between hair colour and minerals in hair. Variation based on the colour type has been reported on several animal species (Kossila et al.,1972; Kornegay et al., 1981; Hornshaw et al., 1985).

Whether the differences are due to the type or the amount of melanin, has not been analysed.

In this study only scanblack type was included and thus the variation in melanin type was excluded. However, even within the same colour type a relationship between the amount of melanin and the concentration of some minerals is obvious (table 3.7). In some cases this correlation overrides the effect of feeding level (table 3.8).

A highly significant correlation ($p<0.001$) was found between the hair colour and the amount of Ca and Mg in the hair. These correlations were strong, $r=0.70$ in both cases (table 3.8).

At the same time the amounts of Ca and Mg depend on their level in the feed. Altogether 56 % and 50 % of the variation can be explained when both hair colour and mineral content of feed are included in the model (R-square values in table 3.9).

An interesting thing about Se is, that, while correlation to Se amount in feed is positive ($r=0.33$), many other minerals seem to have a negative effect on Se concentration in hair (table 3.8).

In the case of P the correlation with colour is low ($r=0.46$), yet significant ($p<0.001$) except in the samples from two feed kitchens. The divergence in these two cases could be explained by interaction between P and high level of Mn. However, a multiple regression analysis does not support this theory.

3.1.3.5 Correlation between minerals in the hair.

Many investigations point out interaction between minerals. In the hair material of this investigation a positive correlation could be found between Ca, Mg and P levels

as one group and between Mg, Na and K levels as an other group. Levels of other minerals did not show correlation with each other.

Table 3.9 Relationship between mineral content of hair and mineral intake and hair colour. Regression equations with highest R-squares.

Mineral in hair	Minerals in feed and pelt characteristics affecting the content in hair	Significance	R^2
Ca	Ca	***	0.14
Ca	Ca + hair colour	***	0.56
Mg	Mg	***	0.05
Mg	Mg + hair colour	***	0.50
Mg	Mg + Zn + hair colour	***	0.55
Na	Na	***	0.11
Na	Na + hair colour	***	0.13
Na	Na + Cu + hair colour	***	0.22
Se	Se	***	0.13
Se	Se + hair colour	***	0.14
Se	Se + Cu + hair colour	***	0.20
Se	Se + Fe + hair colour	***	0.26
Se	Se + P + hair colour	***	0.20
Se	Se + Ca + hair colour	***	0.20
Se	Se + Na + hair colour	***	0.31
Se	Se + Mg + hair colour	***	0.27
Se	Se + Sr + hair colour	***	0.24

*** = $p<0.001$

3.1.4 Conclusions

The present investigation confirms that there are many factors connected with and influencing the metabolism of minerals.

In regard to hair samples, it is important to standardize the material in regard to the colour phase, colour intensity and sample area.

Interactions between minerals obviously have a vital importance in mineral metabolism and therefore more work should be done in future to investigate the interactions.

If the animals are not suffering from deficiency, the mineral content of the hair is not correlated to hair quality.

The mineral levels normally found in Scandinavian mink feed must be considered adequate.

3.2. MORPHOLOGICAL TYPES OF HAIR, VARIATION AND RELATION TO THE MINERAL CONTENT OF HAIR

by Outi Lohi and Palle Vistisen Rasmussen

3.2.1 Introduction

In Scandinavian auction houses pelts from different farms are mixed, but as a result of skilfull grading, auction lots with even quality and colour are achieved. To some degree even special characteristics as for example hair length are taken into account.

However, lots made from the production of a single farm, in other words pelts from animals with the same genetic background, are often more even in regard to the type of hair.

The variation in hair type is assumed mainly to be due to genetics, but in connection to some feeding trials also feed effects are reported.

The aim of this study was to describe objectively the variation in hair type of scanblack mink between some Danish farms and to examine if any relationship between hair type and minerals in hair could be detected.

3.2.2 Material and methods

The variation between populations in hair types became quite obvious in the pelt material from 20 farms, which was in 1985 collected for mineral analyses.

This material, excluding the pelts with the fur defect metallic, was used in this study. Thus the total material was 200 pelts, 100 representing good quality (SS/Saga), and 100 skins of the lower category, quality I.

The following characteristics were examined on this material:

1. Weight of guard hairs and wool hairs per skin area.
2. Length of guard hairs and wool hairs.
3. Shape and area of guard hair cross sections.

The procedure for sampling and results are in detail described by Rasmussen (1987, 1988).

3.2.3 Results and discussion

Even in visual inspection of pelts, clear differences could be seen in the type of hair in pelts from different farms. It was also quite obvious that in some cases the variation within farm population was greater than in others. The morphological studies confirm these findings.

3.2.3.1 Hair length and hair shape.

Hair length and shape of guard hairs were studied on 100 pelts representing good quality (5 pelts per each farm).

The average wool hair length was 15.0 mm and variation between farms from 14.4 to 15.7 mm. Within population variation, expressed as standard deviation of the mean, varied from 0.3 to 1.4 mm. The corresponding figures for guard hair length were: average length 22.1 mm, variation between farms from 21.0 to 24.1 mm and within populations sd varying from 0.3 to 2.4 mm. The variation between farms is illustrated in figure 3.3.

The correlation between wool length and guard hair length was significant ($r=0.7$, $p<0.001$).

The relationship between wool length and guard hair length was also described as wool length in percentage of guard hair length. On average this relationship was 68.0 percent, variation between farms ranging from 64.8 to 72.0 percent with a slight tendency towards the pelts with long guard hair also being "long nap"-type. Opposite to this, the wool length has no correlation to wool hair/-guard hair relationship.

Microscopic study of size and shape of guard hair cross sections was based on guard hairs, which were longer than wool hairs. The average sample size was 31 guard hairs per skin. With 5 skins per farm, the differences between farms are thus based on exact measurements on 100 to 200 guard hairs per farm.

The thickness of guard hairs is considered as a negative characteristic because pelts with thin guard hairs often feel more silky. The thickness and shape of guard hairs were defined by measuring the two axes of the elliptical cross sections from the thickest point of the lancet part. The longer axis (a) varied from 118 µm to 131 µm and was in average 1.74 times the length of the shorter axis (b).

The relation a/b describes the shape of hair cross sections. The mean of a/b values per farm varied from 1.64 to 1.81.

None of these three characteristics (a, b and a/b) was significantly correlated to guard hair length. However, it is interesting to note that the correlations of measures a and b with hair length within both sample areas were negative, which shows that long hairs by no means need to be thick.

The total area of lancet cross sections was calculated by using the following form: $a/2 \bullet b/2 \bullet \pi$.

Because of the elliptical form of lancet, the measure of the shorter axis (b) is more decisive for the shape and the total area of cross sections than axis a. The distribution of guard hairs in groups based on cross section area and cross section form is illustrated in figures 3.4 and 3.5.

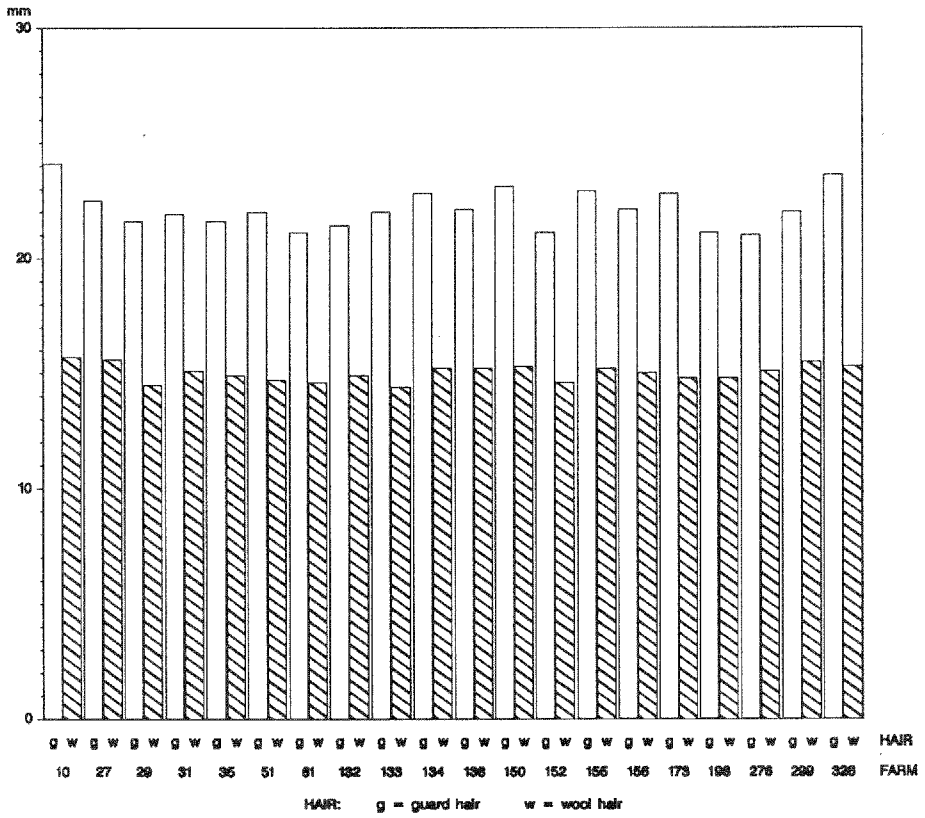

Figure 3.3 Hair length in scanblack males from 20 farms

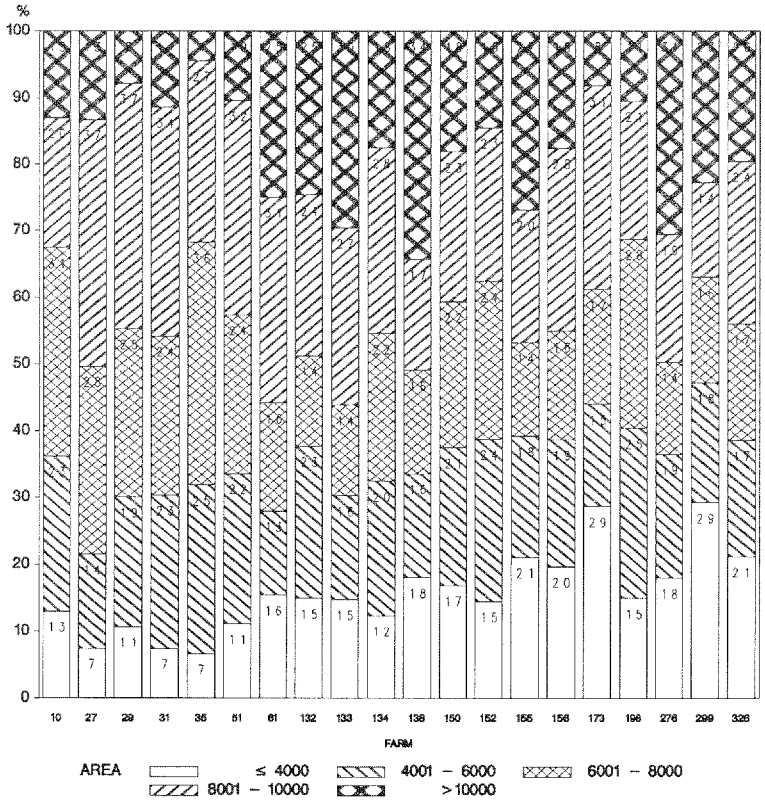

Figure 3.4 Distribution of guard hairs based on lancet cross section area (μm^2)

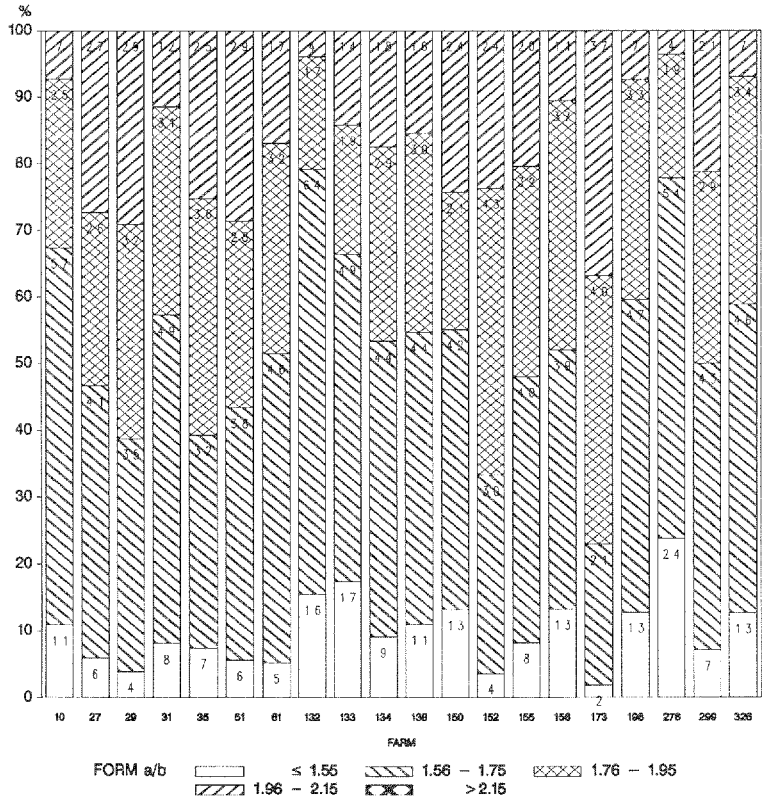

Figure 3.5 Distribution of guard hairs based on lancet cross section form (a/b)

In samples from some farms guard hairs with cross section area over 10000 μm^2 represent more than 30 percent of all guard hairs, whereas on some farms the corresponding figure can be under 10 percent.

3.2.3.2 Weight of hair per skin area.

The mean weight of furs from a skin area of 1.3 cm^2 was 55 mg. Long and intermediary guard hairs corresponded to 28 percent of the weight and wool hairs including

very short guard hairs to 72 percent (table 3.10).

The weight of fur per cm^2 of skin depends on the number of hairs, hair type (= cross section area) and hair length. Correlations between weight and morphological traits are significant in most cases, yet always stronger on the samples taken from the hips than on samples taken from the middle of the back (table 3.11). The area of guard hair cross sections is positively correlated to weight of hairs per cm^2 but the shape of guard hair cross sections (a/b) shows no relation to the weight. (tables 3.10 and 3.11).

Table 3.10 Weigth of fur per area of dried pelt.

Farm	Fur weight mg / sample area (1.3 cm^2)		
	wool hairs $\bar{x} \pm sd$	guard hairs $\bar{x} \pm sd$	total $\bar{x} \pm sd$
10	38.6 ± 7.1	14.7 ± 3.4	53.4 ± 10.4
27	42.4 ± 5.2	16.7 ± 1.5	59.1 ± 5.3
29	35.9 ± 4.5	13.7 ± 1.6	49.6 ± 5.1
31	40.0 ± 5.6	14.3 ± 2.5	54.2 ± 7.8
35	40.0 ± 3.9	14.6 ± 0.9	54.5 ± 4.3
51	39.2 ± 4.9	16.0 ± 1.9	55.2 ± 6.0
61	46.0 ± 4.8	15.1 ± 2.0	61.1 ± 6.4
132	41.9 ± 3.3	15.5 ± 2.5	57.3 ± 3.8
133	38.4 ± 1.9	15.1 ± 1.7	53.5 ± 3.2
134	39.4 ± 7.6	16.0 ± 0.9	55.4 ± 8.4
138	45.0 ± 4.0	16.5 ± 2.2	61.6 ± 5.9
150	38.3 ± 3.5	15.0 ± 1.3	53.2 ± 4.6
152	40.1 ± 6.8	13.6 ± 2.4	53.8 ± 8.8
155	44.7 ± 1.0	17.2 ± 2.8	62.0 ± 2.8
156	33.9 ± 4.6	14.2 ± 2.6	48.1 ± 6.7
173	35.7 ± 2.8	14.1 ± 1.3	49.8 ± 3.9
198	36.1 ± 4.0	13.0 ± 2.6	49.2 ± 5.6
276	45.8 ± 1.7	16.7 ± 1.5	62.4 ± 2.4
299	39.6 ± 3.6	14.9 ± 1.2	54.5 ± 3.4
326	40.5 ± 6.0	18.6 ± 2.2	59.1 ± 7.5
Mean	40.1 ± 5.4	15.3 ± 2.3	55.4 ± 6.9

Table 3.11 Correlation between some physical characteristics of hair.

Correlation between:			Correlation coefficients + significance Samples from the back skin: Midline	On the hips
Guard h. length	-	Guard h. weight	0.38 ***	0.46 ***
Wool h. length	-	Wool h. weight	0.36 ***	0.57 ***
Guard h. length	-	Total weight	0.29 ***	0.40 ***
Wool h. length	-	Total weight	0.35 ***	0.57 ***
Guard hair cross sections:				
Long axis (a)	-	Total weight	0.24 *	0.27 ***
Short axis (b)	-	Total weight	ns	0.38 ***
Cross sect. area	-	Total weight	0.23 *	0.39 ***
Cross sect. form	-	Total weight	ns	ns

* = $p<0.05$
** = $p<0.01$
*** = $p<0.001$

3.2.3.3 Morphological parameters and mineral content of hair.

Single minerals show very little relation to morphological parameters. However, correlation is shown between the weight of wool hairs or hairs in total per area and calcium content of the hair. Values of correlation coefficient are low, from 0.16 to 0.22, yet significant ($p<0.05$).

An interesting result was that there seems to be correlation between mineral content of hair and the form of guard hair cross sections ($r=0.27$, $p<0.01$).

3.2.3.4 Morphological parameters and pelt grading results.

The amount of hair per area, measured as the weight, could also be reflected to the subjective grading of general quality and fur density. Correlations are presented in table 3.12. The fact that correlations are higher, when samples from the hip region are used, shows the importance of this area in the grading process.

Other grading results did not show correlation with morphological parameters.

Table 3.12 Correlations between subjective grading of general fur quality and hair density with morphological parameters.

	Subjective grading	
	General quality	Fur density
Samples from the middle line of the back:		
Guard hair weight	0.21 **	0.23 ***
Wool hair weight	0.24 ***	0.38 ***
Total hair weight	0.25 ***	0.37 ***
Samples from hip area:		
Guard hair weight	0.56 ***	0.23 ***
Wool hair weight	0.58 ***	0.65 ***
Total hair weight	0.62 ***	0.37 ***

** = $p<0.01$
*** = $p<0.001$

3.2.4 Conclusions

Large variations in hair length, thickness of guard hairs and form of lancet cross sections are found in animals from different farms.

The differences in hair density observed by grading were objectively shown in the weight of hair per area.

High mineral content seems to be more characteristic of a hair type with round cross section shape than with more oval ones. This relationship would be an interesting subject for further studies.

Objective morphological measurements confirm the importance of the hip region in the general impression of quality.

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Appendix

The variation of mineral content in weekly samples from five feed kitchens in the period from August 15th until pelting time is for minerals Ca, P, Mg, Na, K, Fe, Zn, Cu, Mn, Sr and Se illustrated in the following figures 3.1.a - 3.1.k.

Mineral contents are given per kilogram feed dry matter.

FC = feed kitchen.

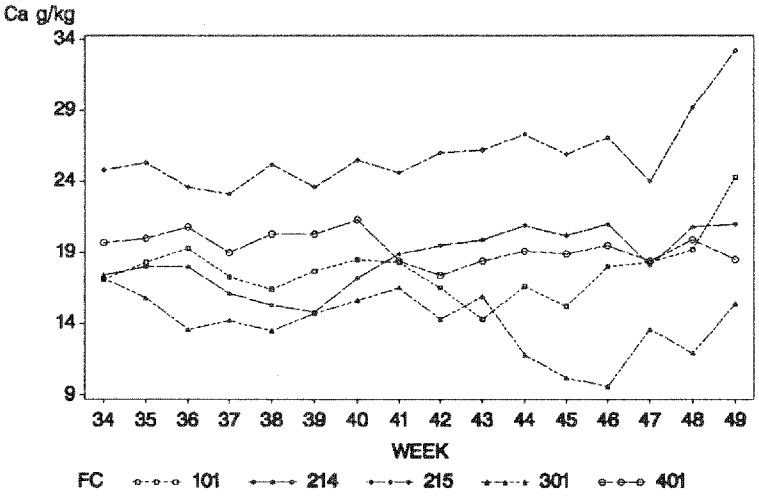

Figure 3.1.1 Calcium (Ca) grams in kilogram feed dry matter

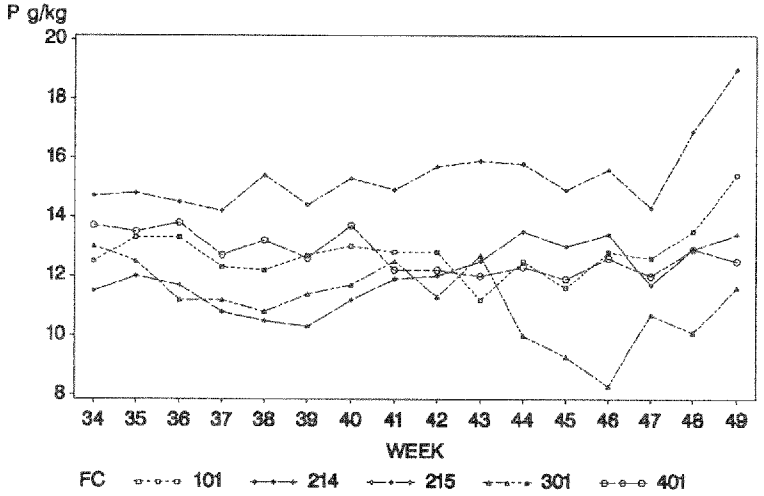

Figure 3.1.b **Phosphorus (P)**
grams in kilogram feed dry matter

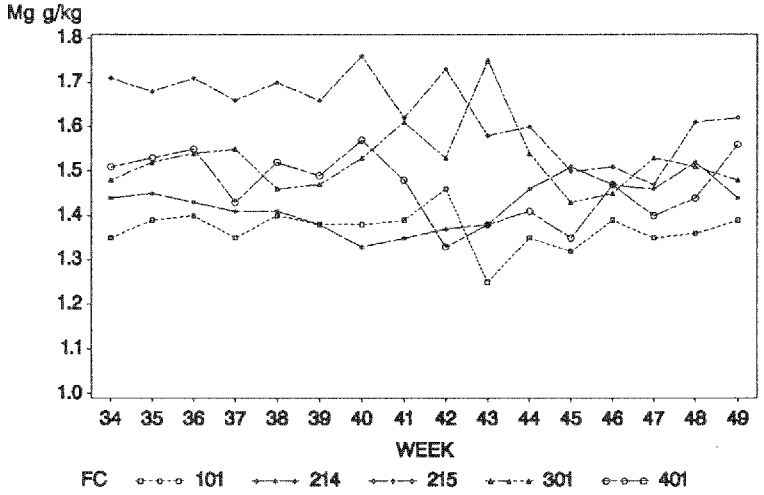

Figure 3.1.c **Magnesium (Mg)**
grams in kilogram feed dry matter

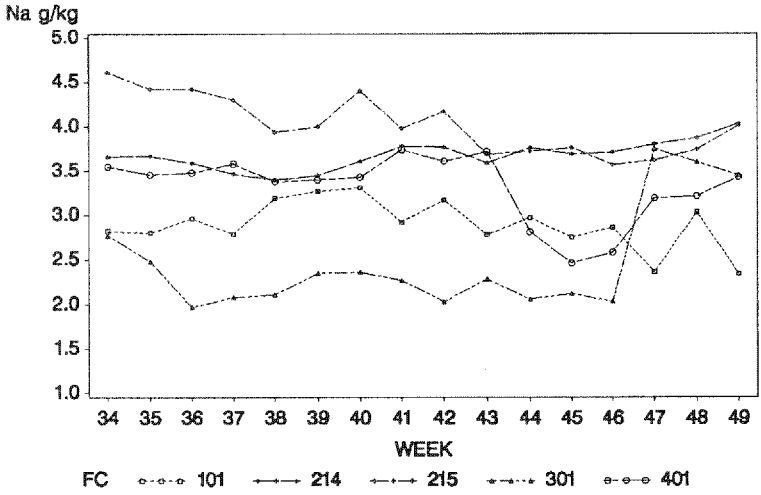

Figure 3.1.d **Sodium (Na)**
grams in kilogram feed dry matter

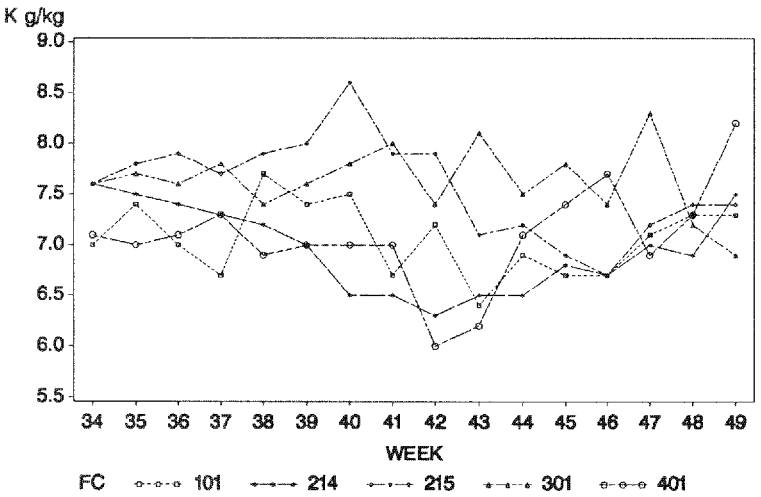

Figure 3.1.e **Potassium (K)**
grams in kilogram feed dry matter

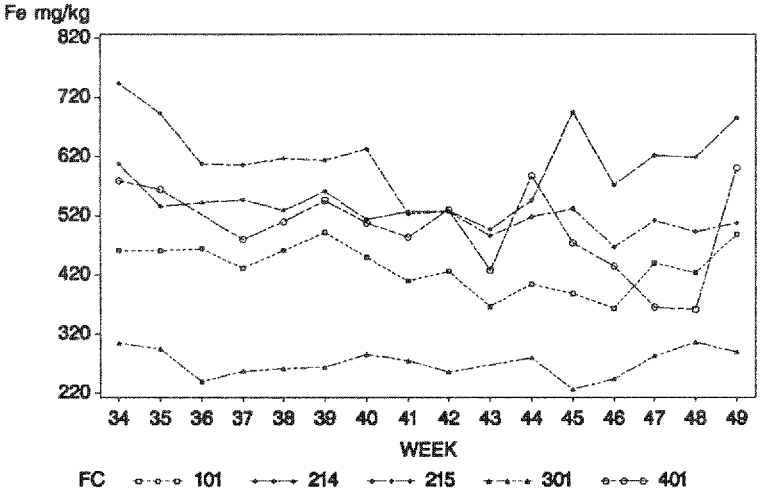

Figure 3.1.f **Iron (Fe)**
milligrams in kilogram feed dry matter

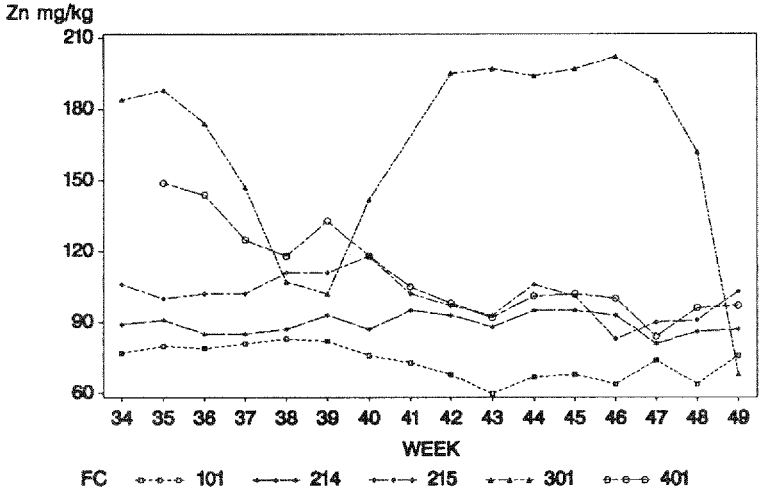

Figure 3.1.g **Zink (Zn)**
milligrams in kilogram feed dry matter

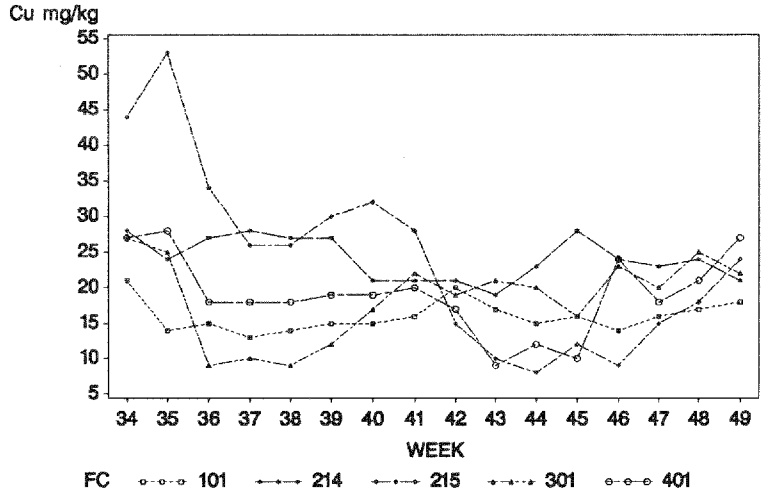

Figure 3.1.h **Copper (Cu)**
milligrams in kilogram feed dry matter

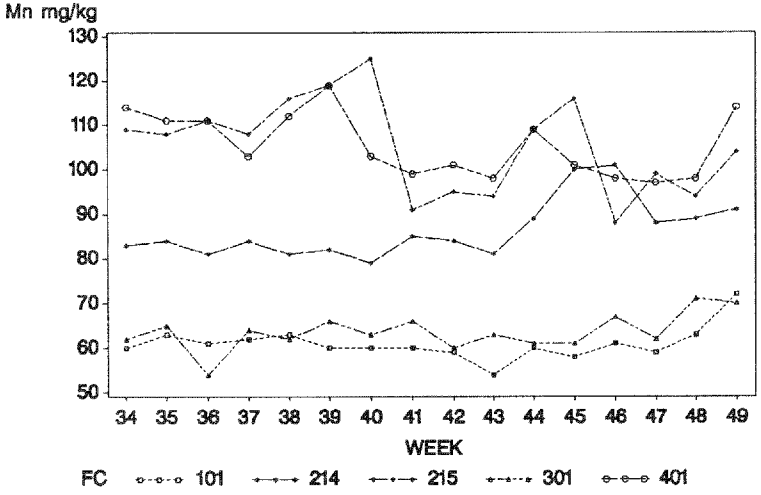

Figure 3.1.i **Manganese (Mn)**
milligrams in kilogram feed dry matter

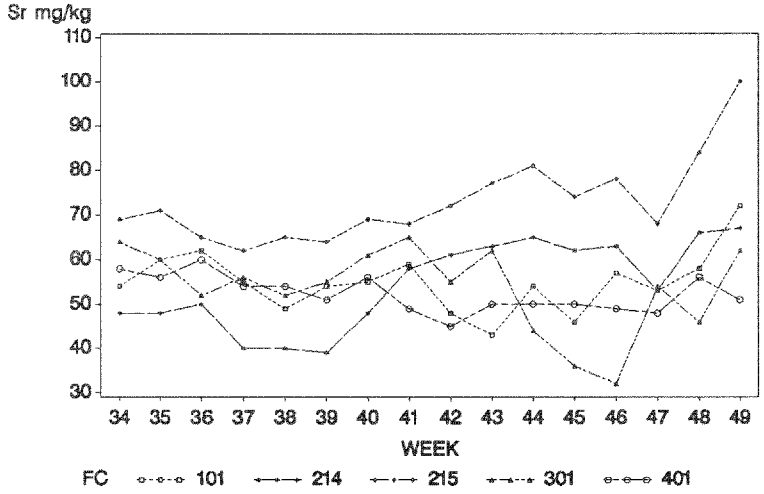

Figure 3.1.j **Strontium (Sr) milligrams in kilogram feed dry matter**

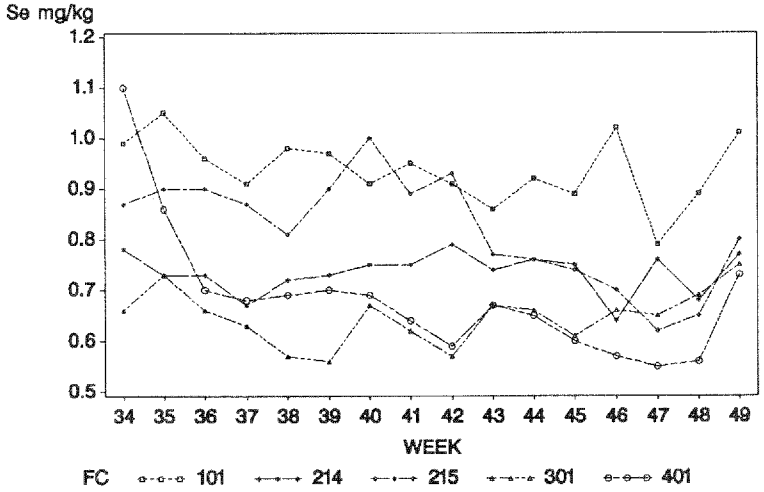

Figure 3.1.k **Selenium (Se) milligrams in kilogram feed dry matter**

4. PHYSIOLOGICAL, GENETIC AND ENVIRONMENTAL VARIATIONS IN HAEMATOLOGICAL AND CLINICO-CHEMICAL PARAMETERS IN MINK;

- THEIR APPLICATION IN HEALTH SURVEILLANCE IN MINK POPULATIONS.

by Asbjørn Brandt

Summary

The investigations were divided into 3 areas:

1) Initially a comprehensive study of variance concerning the effect of physiological, genetic and environmental factors on haematological and clinico-chemical variables in mink was done.

   Thus reference values were determined for a major part of haematological and clinico-chemical blood parameters in mink. Age variation was shown to be particularly important. Interesting representatives of this were the number of erythrocytes, the mean erythrocyte volume, haemoglobin and haematocrit, mean cell haemoglobin concentration and the plasma activity of alkaline phosphatase.

   Heritability of the measured parameters has been analysed provisionally and significant environmental and genetic variations in both haematological and clinico-chemical variables were found. Together with mink production data, further analysis of these data will be performed as an intergral part of a PhD thesis.

   Different analytical methods were developed and applied to mink parallel to these analytical investigations. The most significant methods developed were:

   a) Plasma protein agarose gel electrophoresis with densitometry/quantification of plasma protein classes (alpha-1-2, betha-1-2 and immunoglobulins).

   b) Quantification of lipoproteins (HDL, LDL, VLDL and chylomicrones).

c) Isolation and quantification of creatinekinase isoenzymes MB and MM).

d) Rocket immunoelectrophoresis and quantification of immunoglobulin A, G and M in plasma and milk.

Plasma and milk immunoglobulin G and A concentrations were found to vary dramatically during the lactation. This was also the case for the concurrent change in the mink kit plasma values and the variation found between litters and farms. The significance of these new findings in neonatal kit disease resistance and mortality was discussed.

2) In the work concerning the applicability of clinical pathology, clinical biochemistry and haematology in health surveillance in mink populations it was demonstrated that haematological and clinico-chemical analyses are of great diagnostic and prognostic value and useful in monitoring the nutritional and disease status in mink populations.

Thus clinical and haematological profiles were suggested for the nutritional myocardial degeneration syndrome and nursing disease.

Certain clinico-pathological parameters were evaluated as disease predictors in mink populations on problem farms and in outbreaks and cases of greasy mink kits (early and late type), nursing disease, cystitis, nephritis, urolithiasis, and sudden death syndromes.

Based on this part of the project a general mink Health/production-Check-Profile (HCP) was suggested. It should include parameters of haematology (erythrocyte and leucocyte indices), enzymology (ASAT, ALAT, alkaline phosphatase and CK) and urology (density, blood and pH).

The general HCP could be applicable in a health surveillance programme for screening a given population for sub-clinical symptoms concerning both environmental/nutritional, hereditary or systemic diseases in mink.

3) In the experimental part of the study significant effects of dietary iron, copper and zinc on haematological and enzymological development, on fur growth and on mineral balance and turnover were demonstrated.

It was suggested that the present investigations should be followed by a statistical analysis of haematological and clinico-chemical blood data together with environmental and pelt data. This should include a test of the predictive value and validity of the proposed mink HCP in a larger scale and evaluated as a service function for the mink farmer.

Sammendrag

Undersøgelserne blev opdelt i 3 områder:

1) Indledningsvis blev der foretaget en analyse af effekten af fysiologiske, genetiske og miljø faktorer på haematologiske og klinisk-kemiske variabler hos mink.

   Der blev bestemt reference intervaller for et større antal haematologiske og kliniskkemiske blodparametre hos mink. Aldersvariationen viste sig at være særdeles vigtig. Som interessante eksempler herpå kan nævnes antallet af erytrocytter, erytrocytternes gennemsnitsvolumen, hæmoglobin koncentration, hæmatocrit og erytrocyt hæmoglobinkoncentration samt basisk fosfataseaktivitet.

   Der blev fundet væsentlige miljømæssige og genetiske variationer i materialet, og heritabiliteten blev beregnet for hæmatologiske og kliniskkemiske variabler. Sammen med andre data fra produktionen vil en videre analyse af det store materiale indgå i en kommende licentiatafhandling.

   Sideløbende med dette analysearbejde blev der arbejdet med analysemetodeudvikling og applikation. De vigtigste metoder, der blev udviklet var, a) plasmaprotein ved agarosegelelektroforese og densitometri/ kvantificering af plasmaproteinklasserne (alpha-1,-2, betha-1-2 og immunoglobuliner), b) kvantificering af lipoproteiner (HDL, LDL, VLDL og chylomikroner), c) isolering og kvantificering af kreatinfosfokinase isoenzymer (MB og MM), d) raket-immunoelektroforese og kvantificering af immunoglobulin A, G og M i plasma og mælk.

   Under laktationen viste det sig, at mængderne af plasma- og mælkeimmunoglobulinerne G og A varierede dramatisk. Dette var også tilfældet for den synkrone ændring i minkhvalpenes plasmakoncentrationer samt kuld- og farmvariation af disse. Betydningen af nævnte nye fund i forbindelse med neonatal hvalpesygdomsresistens samt hvalpesygdomsmortalitet blev diskuteret.

2) Arbejdet omfattede en applicering af klinisk-patologi, klinisk-biokemi og hæmatologi som værktøj i sundhedsovervågning i minkpopulationer. Det blev demonstreret, at hæmatologiske og klinisk-kemiske analyser har en høj grad af

diagnostisk og prognostisk værdi samt er anvendelige, når den ernærings- og sygdomsmæssige status skal måles/-overvåges i en given minkpopulation.

Hæmatologiske og kliniskkemiske analyseprofiler blev foreslået for syndromet ernæringsbetinget myokardiel degeneration og diegivningssyge.

Visse klinisk-patologiske parametre blev vurderet med henblik på egnethed som 'sygdomsprediktorer' i minkpopulationer, på problem minkfarme og under sygdomsudbrud så som fedtede halpe (sen og tidlig form), diegivningssyge, blærebetændelse/blæresten, nyrebetændelse og velfærdssyge.

På baggrund af denne del af projektet blev general minksundheds/produktions-Check-Profil (HCP) foreslået. Den skulle indeholde hæmatologi- (erytrocyt- og leukocytvariabler), enzymologi- (ASAT, ALAT, basisk fosfatase og kreatin-fosfokinase) og urologiparametre (vægtfylde, blod og pH).

Den generelle HCP kunne anvendes i forbindelse med et sundhedsovervågningsprogram, hvori opgaven var at afsøge en given population for subkliniske symptomer på miljø-/ernærings betingede, arvelige eller systemiske sygdomme hos mink.

3) I den eksperimentelle del af undersøgelsen fandtes en signifikant effekt af diætetisk jern, kobber og zink på hæmatologiske, enzymologiske variabler samt pelsudviklingen og mineral balancen.

   På lignende måde fandtes signifikante effekter af diætetisk dl-alpha-tocopherylacetat, natriumselenit og polyumættet fedt.

   En videreførelse af projektet blev foreslået at kunne være en statistisk bearbejdning af de hæmatologiske og klinisk-kemiske bloddata sammen med miljø- og pelsdata. Dette arbejde skulle også inkludere beregning af den prædiktive værdi og validiteten af den foreslåede mink-HCP i større skala, herunder vurdere mink-HCP som et servicetilbud til minkfarmere.

4.1 Introduction

Haematological and clinico-chemical analyses are of great diagnostic and prognostic value and useful in monitoring the nutritional status in mink, but so far the general use of these analyses has been limited due to the lack of applicable reference values for the populations at stake.

Several efforts have been made to determine the normal variation of haematological and clinico-chemical blood values in mink (Jørgensen & Christensen, 1966; Skrede, 1970; Rotenberg & Jørgensen, 1971; Poulsen & Jørgensen, 1977; Zeissler et al., 1980 & 1981), the organ distribution of various enzymes (Juokslahti et al., 1980), and the effect of sedation and anaesthesia on haematological indices and the concentration of blood plasma enzymes (Jepsen et al., 1981).

The literature lacks, however, a more comprehensive study where significant bias such as plasmacytosis is eliminated and where the reference population, environment and analytical method are specified.

Field observations have shown that in situations of stress there can be great differences in the haematological and clinico-chemical blood values in mink from different farms. The animals react differently to changes in environment, feed and disease pattern.

The objective of this part of the project was to determine physiological, genetic and environmental variations in haematological and clinico-chemical parameters in mink and seek application of these in health surveillance in mink populations.

The investigations were divided into 3 areas:

1) Haematology, enzymology, vitaminology and metabolites:
   a) method development
   b) reference value determinations
   c) heritability studies in mink.

2) Clinico-pathological and epidemiological investigations on "problem mink farms" with:

   Greasy mink kits (early and late type), nursing disease, cystitis, nephritis, urolithiasis, sudden death syndromes (e.g. myocardial degeneration). Evaluation of certain clinico-pathological parameters as (subclinical) disease predictors in populations (epidemiological screening for diseases).

3) Experimental investigations concerning:
   a) minerals: iron, copper and zinc turnover with emphasis on haematological and enzymological age development, mineral balance and fur development.
   b) diseases: nursing disease, nutritional myopathy and wet belly.

In tables 4.1, 4.2 and 4.3 the total number of studies, animals, sexes, genotypes, farms, samples and references concerning the 3 investigation areas mentioned are summarized.

4.2 Materials and methods

4.2.1 Animals, blood samples and analyses

The mink used for the different parts were from Trollesminde (Experimental Farm), or were bought from project farms and housed at Trollesminde.

The mink for the heritability studies 1.c were both project farm mink and family members of these placed at Favrholm Experimental Farm in order to eliminate as many environmental factors as possible.

All animals were randomly assigned to the study lots among normal plasmacytosis-free Danish domestic mink.

The mink kits were caged conventionally with males and females in pairs.

The animals in all studies except those concerning a specific disease complex were fed conventional Danish feed kitchen diets unless otherwise stated in the text.

Feed and water were provided ad libitum throughout the experimental periods.

Blood samples were collected in the morning on fasting (minimum 8 h) animals, except for suckling mink kits.

In studies where more than 1 ml blood was needed, samples were taken by vena jugularis puncture under anaesthesia: 25 mg Althesin ®/kg body weight intraperitoneally.

Where less blood was needed or in studies of haematological variables blood samples were taken from v. jugularis until 34 days of age and thereafter from v. cephalica.

Twenty µl capillary blood was transferred by a capillary blood pipette to the dilution medium (Dilued TM I.T. Barker), for electronical counting of the number of

erythrocytes (Linson 431 A, Cap. Curr. = 700, Discr. = 75, window = 471, dia. = 80). Haemoglobin (Hb (mmol/l)) was determined by the cyanomethaemoglobin method. The haematocrit (HCT (%)) was measured electronically (Linson 432 A). Mean corpuscular volume (MCV (fl)) and mean corpuscular haemoglobin (MCH (fmol)) were measured electronically (Linson 432 A) and mean corpuscular haemoglobin concentration (MCHC (mmol/l)) was calculated.

All results were the means of double tests.

Leucocyte differential counts were determined on 2 May-Grünwald-Giemsa stained smears from each animal. The results were the means of 100 white blood cell counts on each slide.

The animals were of optimal health as judged by the weight development and the continuous clinical surveillance.

Approved laboratory methods were used when applicable or with necessary modifications to fit sample material from mink - for method summarization see appendix table 4.8.

All analyses were made with plasma, but might as well be made with serum.

4.2.2 Haematology and clinico-chemical investigations

4.2.2.1 Method development study

Methods applied are summarized in appendix table 4.8.

Sample material used was both fresh and frozen blood, plasma, urine and organs from more or less all studies mentioned in tables 4.1, 4.2 and 4.3.

4.2.2.2 Reference value determination studies

The distribution and number of animals, sexes, genotypes, samples and references concerning method development studies of haematological, enzymological, vitaminological and metabolite blood variables are shown in table 4.1, and the methods are shown in Appendix table 4.8.

Table 4.1. The distribution and number of animals, sexes, genotypes, samples and references concerning the haematological and clinico-chemical area.

Study	Age (days)	Mink	Sex	Number of types	Farm	Samples	Reference
1.a.1	0-42	56	2	2	1	13	Brandt, 1988d, e
1.a.2	0-42	40	2	2	1	9	Brandt, 1988e
1.b.1	34-adult	160	2	2	2	17	Brandt, not publ.
1.b.2	adult	40	2	2	1	12	Brandt, not publ.
1.b.3	34-adult	40	2	2	1	12	Brandt, not publ.
1.b.4	0-42	58	2	2	1	13	Brandt, 1988e Brandt, not publ.
1.b.5	adult	12	1	1	1	12	Hau et al., 1988
1.c.1	adult	110	1	1	10	1	Brandt & Lohi, 1986
1.c.2	34-adult	86	1	1	8	1	Lohi & Brandt, 1988
1.c.3	adult	929	1	1	8	1	Lohi & Brandt, 1988

- Age variation, growth period (study 1.b.1)

Blood samples were taken 16 times from 7 days after birth until pelting, from a group of 160 normal mink kits of equal age. At weaning 55 days after birth the kits were assigned to a randomized factorial survey: 2 (male and female) · 2

(pastel and standard) · 2 (feed kitchen dietary formula).

- Age and cyclic variation (study 1.b.2)

Blood samples were collected by v. cephalica puncture from 20 male and 20 female normal pastel mink once a month for a year, starting at 4 months of age.

Plasma calcium and magnesium were determined by atomic absorption spectrometry; inorganic phosphorus by the molybdate/-vanadate method; sodium and potassium by flame emission spectrometry; chloride by electrometric titration.

Total heparinized plasma alkaline phosphatase activity was determined by the method of Bessey et al. (1946) as modified by Hausanen et al. (1967).

4.2.2.3 Investigations on heritability (study 1.c)

The heritability of haematological and enzyme values was studied on three sets of material. A material of 442 blood samples was in 1983 collected from animals in progeny groups representing 10 different farm populations and 30 breeding males. Each male was represented by offspring from 4 litters, 2 males + 2 females each. Thus the material allowed comparison between progeny groups and farm populations.

The effect of environment was studied by comparing animals raised on the progeny test farm from July 1st until pelting to their full sibs raised on the farms of origin (Lohi et al., 1986).

In 1984 the material consisted of 90 progeny groups deriving from 16 different farms. The animals were again submitted to same management and feeding conditions from July 1st until pelting. Due to outbreak of 'sudden death syndrome' the material offered a possibility to compare progeny groups and farm populations under stress conditions.

In 1985 all male kits after 8 breeding males from the research farm population were sampled.

4.2.3 Clinico-pathological and epidemiological investigations.

- Wet belly syndrome (study 2.b).

In July, September and October spontaneous urine samples were collected from 600 male mink kits from a problem farm. The fre-

quency and severity of Wet Belly Syndrome (WBS) defined as an peripraeputial hair and skin discolouration, alopecia and atopia were registrated. Fresh urine samples were tested using Boehringer Mannheim BM9 stick with qualitative and semi quantitative reaction fields for nitrite, leucocytes, pH, protein, glucose, ketones, urobilinogen/bilirubin and blood.

At pelting microbiological and histopathological samples were taken from the bladder, liver and kidney and P-Urea, P-CK and P-AP were analysed in the blood plasma by methods mentioned in Appendix table 4.8.

- Myocardial degeneration (study 2.c).

Total plasma creatine kinase (P-CK, EC 2.7.3.2) activity was determined using the BM optimized method applied to the LKB 8500 Reaction Rate Analyzer. The mink were euthanized by exsanguation and autopsied. Tissue from the left cardial ventricle was fixed in 4% neutral formaldehyde solution. The selected myocardium samples were embedded in paraffin, cut and stained with haematoxylin and eosin and van Gieson's connective tissue stain. The degree of myocardial degeneration (MCD) was evaluated and slight, medium and severe were catagorized as histological MCD.

The sensitivity, specificity and predictive value of the CK-test were calculated.

- Nursing disease (study 2.d).

Lactating mink females with number of kits exceeding 6 were weighed and blood samples collected on 27.05 and 17.06. Samples were analysed for haematocrit, P-sodium and P-potassium. Mink showing severe signs of nursing disease were euthanized by exsanguation. The blood was submitted to an extensive haematological and clinico-chemical screening. Samples of liver and kidney were embedded in paraffin, cut and stained with haematoxylin and eosin and van Gieson's connective tissue stain and evaluated histopathologically.

- Milk examinations (study 2.e).

Milk was collected by digital milking from mink females with greasy kits from different feed kitchens (FK). The milk was analysed for the content of fatty acids by gas chromatography. Total saturated and monounsaturated, omega-3-poly unsaturated, and omega-6-poly unsaturated fatty acids were calculated.

4.2.4 Experimental investigations

- Mineral supply (study 3.a.1).

Equivalent ferro-EDTA, amino-acid chelated iron, iron sulphate and ferri glutamate (50ppm) were supplemented to an ordinary fish offal based mink diet. The total dietary iron, zinc and copper content was determined by atomic absorption spectrophotometry.

Ceruloplasmin in plasma was determined by rocket immuno electrophoresis using crossreacting rabbit antibodies against rat ceruloplasmin. Total and latent iron binding capacity was determined in the blood plasma by BM-methods. Digestibility trials were performed for iron, zinc, and copper. At pelting the mink were euthanized by thiobarbital-Na. The blood was submitted to extensive haematological and clinico-chemical screening. Samples of the right liver lobe were analysed for total iron, zinc, and copper content by atomic absorption spectrophotometry.

- Effect of chelators (study 3.a.2).

The effect of dietary Vantosil ® was monitored in a factorial experiment with 3 levels of Vantosil, 3 levels of copper, and 2 levels of zinc.

- Effect of glycosinolates (study 3.a.3).

The effect of increasing dietary rape glycosinolate concentrations in mink kits was studied.

Thyroxine (T3 and T4, Labovet) was determined using Radio Immuno Assay. The mink were euthanized by exsanguation. The blood was submitted to extensive haematological and clinico-chemical screening. Samples of thyroidea were embedded in paraffin, cut and stained with haematoxylin and eosin and van Gieson's connective tissue stain and evaluated histopathologically.

- Effect of other dietary factors (study 3.b).

The effect of dietary factors on the development of nursing disease symptoms and performance was studied in lactating mink.

Acid base was determined using anaerobically arterilized capillary blood. The acid-base data were examined at 37°C according to the micro-method (Siggård/Andersen) using Blood Micro Equipment 22-Radiometer.

Mink showing severe signs of nursing disease were treated as described in study 2.d.

4.2.5 Data analysis

The mean values and standard deviation of the variables were calculated. Analysis of variance for the treatments and the interactions between treatments were carried out by means of the GLM procedure (Helwig & Council, 1979). Significance of differences between mean values was tested using the Student's t-test.

By regression analysis the character of the relationship between dependent and independent variables was investigated (Draper & Smith, 1966).

All analyses were performed using the SAS software for Personal Computers.

Table 4.2 **The distribution and number of animals, sexes, genotypes, samples and references concerning the clinico-pathological and epidemiological areas.**

Study	Age (days)	Mink	Sex	Number of types	Farms	Samples	Reference
2.a.	adult	118	1	1	10	1	Brandt & Lohi, 1986
2.b.	90-adult	600	1	6	2	3	Brandt & Clausen, 1988
2.c.	adult	384	2	2	1	1	Brandt, 1986c
2.d.	adult	800	2	1	2	2	Brandt & Henriksen, 1987b
2.e.	adult	42	1	2	8	2	Brandt, 1987b

Table 4.3 **The distribution and number of animals, sexes, genotypes, samples and references concerning the experimental investigation area.**

Study	Age (days)	Mink	Sex	Number of types	Farms	Samples	Reference
3.a.1	adult	600	1	1	6	1	Brandt & Lohi, 1986; Brandt & Mejborn, 1987
3.a.2	34-adult	130	2	1	1	1	Mejborn & Brandt, 1988
3.a.3	34-adult	48	1	1	1	2	Brandt & Henriksen, 1987c
3.b.	adult	64	1	1	1	5	Brandt et al., 1986

4.3 Results and discussion.

4.3.1 Haematology and clinico-chemical investigations

4.3.1.1 Method development

In general, the development of laboratory methods was concentrated on comparative studies of different available commercial test kits or approved methods and applying the most suitable to mink sample material.

- Haematology and coagulation factors (study 1.a.1).

In haematology emphasis was made to master basic haematological disciplines, especially the quantification (differential count) of leucocytes in conjunction with the automated electronical counting of erythrocytes, leucocytes and platelets.

Erythrocyte size distribution was measured using different window values.

Platelet counting was sought correlated to different coagulation factors, but determining the latter required time and money exceeding our possibilities.

- Immunological methods (studies 1.a.2. & 3.a).

An intensive study was made in optimizing the agarose gel, running conditions, scanning apparatus, antibodies in the process of developing methods of quantifying plasma, milk (and urine) proteins using electrophoresis.

Thus proteins such as albumin, alpha, betha and gamma globulins, were determined in agarose gel electrophoresis (fig 4.1) and quantified (fig 4.2).

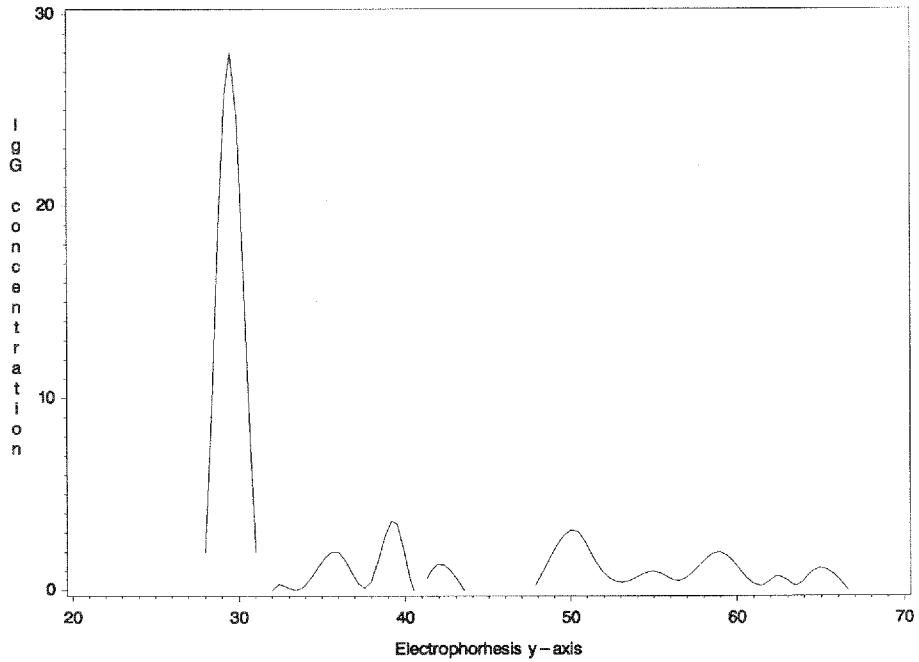

Figure 4.1 Densiometry of normal mink plasma.

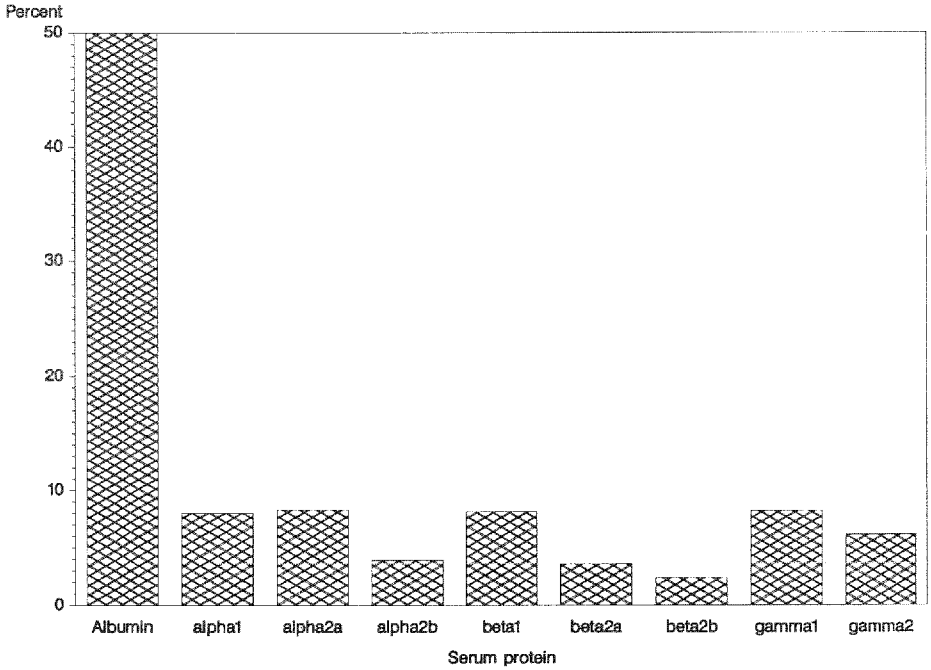

Figure 4.2 Percent distribution of plasma proteins in agarose gel electrophoresis.

In the same manner agarose gel electrophoresis and densitometry for the quantification of mink plasma lipoproteins and the creatinine phosphokinase isoenzymes were developed (fig. 4.3).

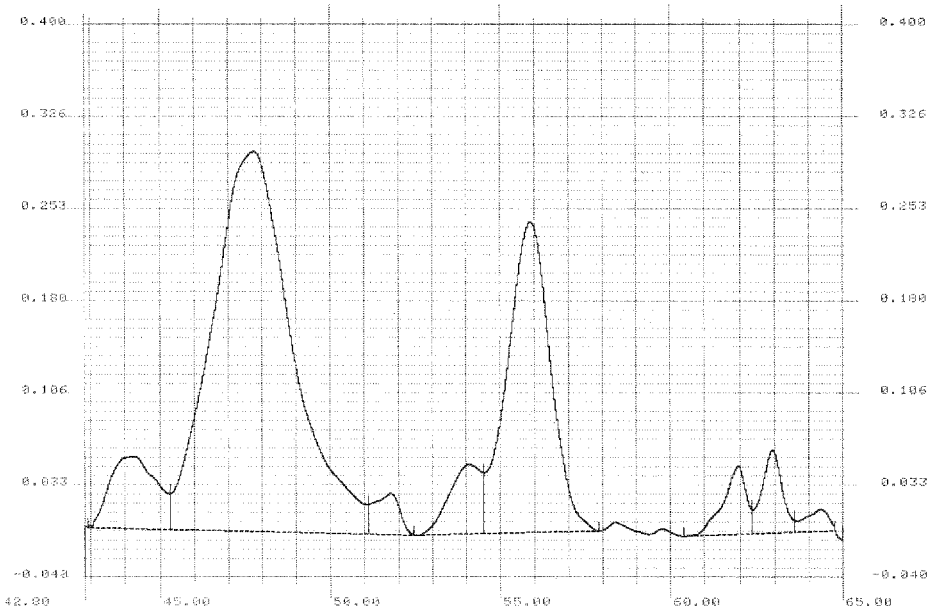

Figure 4.3 Densitometry of lipoproteins in normal mink plasma.

Rocket immunoelectrophoresis analysis of the contents of plasma and milk immunoglobulins A, G and M in mink was developed using rabbit antibodies against human proteins which cross react with mink. An example of the rocket immunoelectrophoresis analysis is shown in fig. 4.4. (Brandt, 1988d).

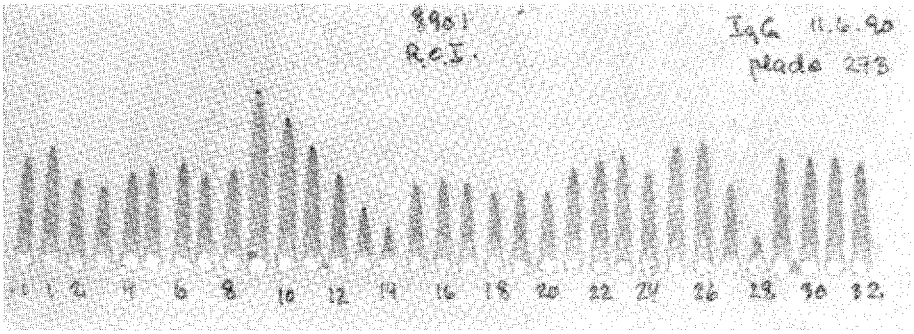

Figure 4.4 Rocket immuno electrophoresis of mink plasma (IgG).

For the milk studies a simple efficient method for digital milking of mink females was developed.

4.3.1.2 Reference value determination

- Age variation in haematological and clinico-chemical values (studies 1.b.1., 2. & 3).

Variations in haematological indices were studied in 16 blood samples from each of 160 kits from weaning until pelting showing significant age variations in haematocrit, haemoglobin, MCV, MCH, MCHC, erythrocytes, leucocytes, differential counts of leucocytes, phosphorus and alkaline phosphatase (tables 4.4 & 4.5, fig. 4.5). There were no variations in plasma conc. of calcium, magnesium, sodium, potassium and chloride (Brandt, in press).

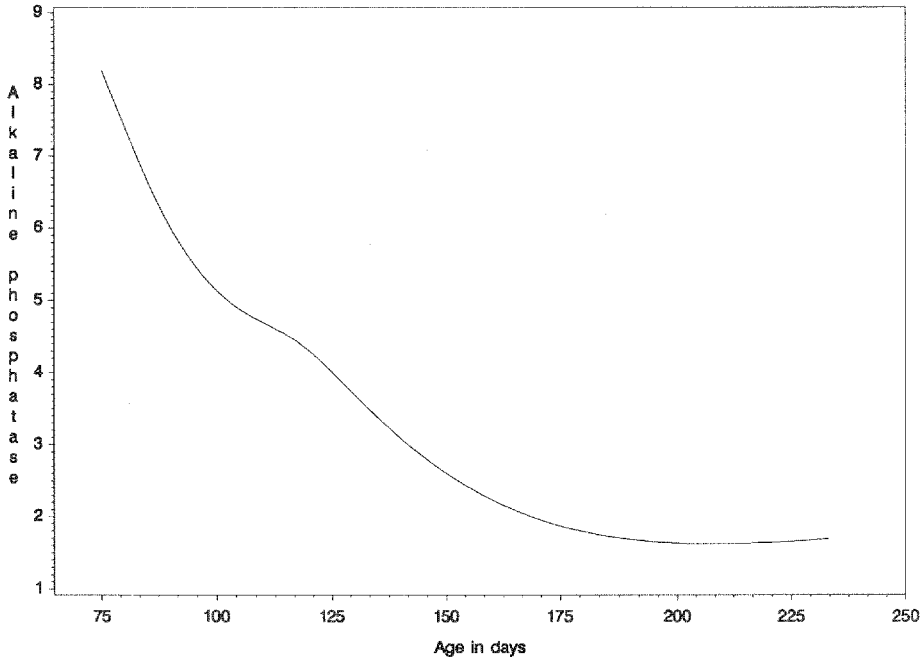

Figure 4.5 Age variation in mink plasma alkaline phosphatase mckat/liter

- Age variation in immunological variables (study 1.b.4).

Significant age variations in immunoglobulin A, G and M classes in mink female milk, plasma and kit plasma were shown. A surprisingly high milk IgA and slow development of IgG in kit plasma were found.

The data are presented graphically in fig. 4.6 & 4.7. (Brandt, 1988d, 1988e).

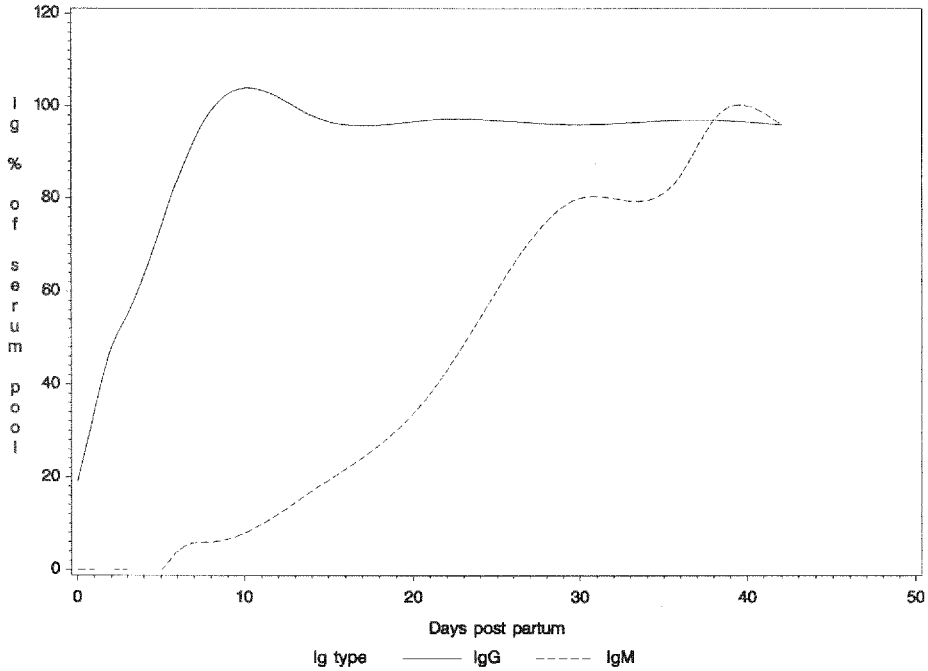

Figure 4.6 IgG and IgM concentration in plasma of mink kit.

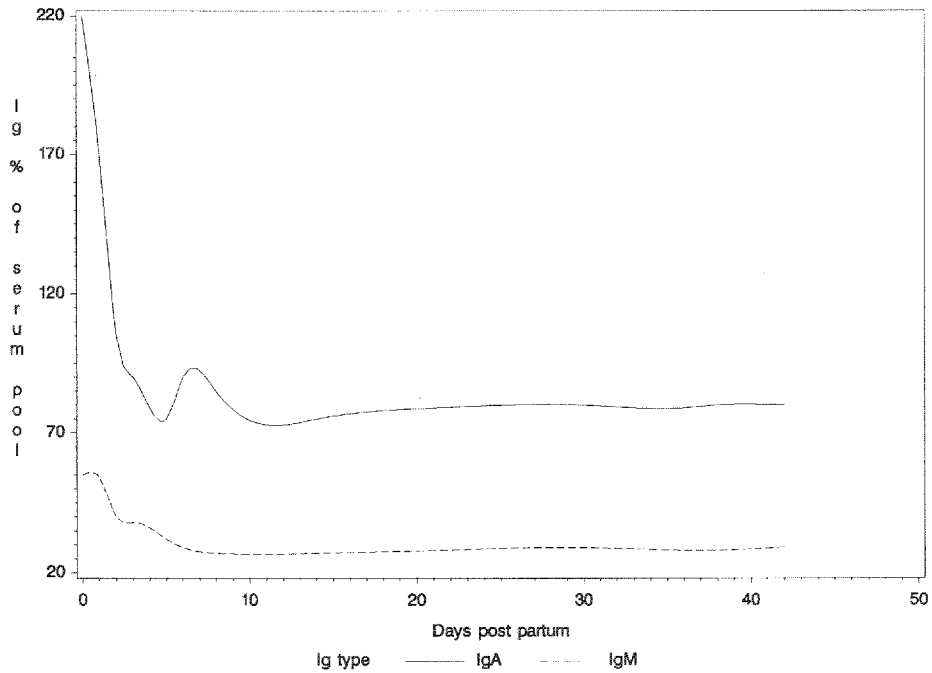

Figure 4.7 IgA and IgM concentration in colostrum of mink.

- Variation in fibrin/fibrinogen level during pregnancy and lactation (study 1.b.5).

Extremely high levels of plasma fibrin/fibrinogen were recorded in female mink during the breeding season using rocket immunoelectrophoresis. During the subsequent gestation period the levels gradually decreased to reach non-breeding levels at term. The fibrin/fibrinogen molecules were found to exist mainly as large aggregates (molwt 630kD). The molecules were found to bind heparin and concanavalin A (Hau et al., 1988).

Table 4.4 Reference values for haemoglobin (Hb), haematocrit (HCT), erythrocytes (RBC), MCV, MCH, MCHC and leucocytes (WBC) of female mink kits from 7 days of age until pelting. Mean values and standard deviations (SD). Part I.

	DAYS AFTER BIRTH					
	7	14	21	34	41	55
	n: 20	20	20	30	39	80
Hb (mmol/l)	7.98	7.3	6.67	6.13	7.25	8.08
SD	0.89	0.62	0.44	0.44	0.56	0.82
Lower-upper[1)]	6.5-10.5	6.8-9.0	5.0-7.4	5.4-7.0	6.1-8.5	6.7-9.7
HCT (%)32.94	30.1	27.49	24.90	31.89	32.74	38.91
SD	2.68	2.00	2.37	2.00	2.25	2.68
Lower-upper	27.9-39.0	25.1-38.1	22.0-31.5	20.1-28.6	27.7-36.4	27.7-38.8
RBC (1012/l),	5.29	4.11	3.10	3.62	4.40	5.29
SD	0.56	0.58	0.24	0.27	0.35	0.56
Lower-upper	4.5-6.6	4.0-6.0	2.6-3.6	3.1-4.2	3.8-5.2	4.5-6.6
MCV (fl)	72.21	69.90	69.00	69.05	68.20	62.81
SD	2.48	2.01	2.26	2.09	2.00	2.48
Lower-upper	67.0-77.8	67.1-76.0	64.0-73.0	64.1-73.0	64.0-72.1	59.0-68.8
MCH (fmol)	2.24	2.08	2.03	1.69	1.65	1.54
SD	0.16	0.10	0.13	0.10	0.09	0.07
Lower-upper	1.9-2.1	1.8-2.2	1.8-2.3	1.5-1.9	1.5-1.8	1.5-1.7
MCHC (mmol/l)	25.03	25.08	25.47	24.54	24.0	24.53
SD	1.28	1.68	1.43	1.10	0.90	0.98
Lower-upper	23.6-28.3	23.1-28.7	27.3-32.0	22.8-26.7	20.9-25.4	23.1-27.8
WBC (109/l)	6.21	6.3	7.98	8.08	11.11	10.21
SD	2.28	2.22	1.98	1.96	2.98	2.28
Lower-upper	2.8-12.4	3.3-9.8	4.6-10.9	4.6-10.9	6.1-16.6	6.8-16.4

1) Determined 90% interval: 5th to 95th percentile.

Table 4.4 **Reference values for haemoglobin (Hb), haematocrit (HCT), erythrocytes (RBC), MCV, MCH, MCHC and leucocytes (WBC) of female mink kits from 7 days of age until pelting. Mean values and standard deviations (SD). Part II.**

DAYS AFTER BIRTH						
72	90	104	120	139	167	233
80	78	78	78	78	77	38
10.14	11.33	11.91	12.15	12.75	12.87	12.69
0.83	0.74	0.83	0.64	0.90	0.98	0.48
8.6-11.6	9.9-12.4	10.5-13.3	11.0-13.3	11.3-14.1	11.4-15.2	11.5-13.4
44.28	46.66	46.13	48.56	48.73	49.8	
3.39	2.72	3.12	3.01	2.81	3.12	2.88
31.7-44.3	39.2-48.8	40.3-50.6	41.0-51.8	43.5-52.6	44.3-55.2	44.0-55.2
6.37	7.57	8.08	8.16	8.57	8.69	8.60
0.50	0.46	0.57	0.40	0.59	0.61	0.75
5.2-7.4	6.6-8.2	7.0-9.1	7.5-8.8	7.6-9.7	7.6-10.3	7.9-10.4
61.30	59.06	58.95	57.54	57.13	56.75	56.17
2.44	2.03	2.27	1.76	2.10	2.03	2.39
57.0-64.0	56.8-64.0	55.2-64.6	55.0-61.0	54.0-62.0	54.0-60.0	53.9-60.1
1.60	1.50	1.47	1.49	1.50	1.48	1.45
0.10	0.06	0.07	0.04	0.10	0.11	0.17
1.5-1.8	1.4-1.6	1.4-1.6	1.4-1.6	1.3-1.7	1.3-1.7	1.1-1.7
26.07	25.39	25.04	25.90	26.28	26.14	26.12
1.94	1.31	1.19	0.99	1.79	1.46	1.50
23.8-29.8	23.2-28.2	23.3-27.7	24.4-27.9	24.1-29.1	23.7-27.9	22.9-29.7
18.78	11.93	10.42	12.01	10.96	9.09	10.14
2.67	2.99	2.50	2.29	2.61	2.07	2.79
10.0-12.9	7.9-19.4	6.3-19.4	6.7-17.8	7.8-16.5	5.0-14.9	4.2-13.3

Table 4.5 **Haemoglobin, haematocrit, MCHC, rel. neutrophil, lymphocyte, monocyte, eosinophil and basophil counts of 20 male and 20 female pastel mink from 15/8 until 15/7 (one year). Mean values and standard deviations (SD). Part I.**

Male: n = 20 Fem.: n = 20	DATE 15/8	15/9	15/10	15/11	15/12
Hb (mmol/l), male	10.49A	10.81AB	11.12AB	11.55B	12.17C
SD	0.43	0.56	0.50	0.37	0.50
Hb (mmol/l), female	10.43A	10.87AB	10.92B	11.36B	11.67C
SD	0.30	0.37	0.37	0.37	0.37
HCT (%), male	52.0A	56.0B	57.0C	60.0D	61.0D
SD	1.8	2.3	2.2	1.9	2.7
HCT (%), female	52.0A	57.0B	57.0B	59.0C	59.0C
SD	1.6	2.2	1.7	1.9	1.9
MCHC (mmol/l), male	19.4A	19.4A	19.5A	19.4A	19.9A
SD	0.6	0.6	0.6	0.5	0.7
MCHC (mmol/l), female	19.1A	19.0A	19.2A	19.3A	19.8A
SD	0.5	0.7	0.6	0.5	0.5
rel. neutrophil count (%), male	39.2	41.5	45.3	52.6	44.2
SD	6.4	5.3	6.9	8.5	6.2
rel. neutrophil count (%), female	33.8	40.1	42.3	49.8	44.3
SD	6.7	8.5	6.9	7.7	6.5
rel. lymphocyte count (%), male	59.8	56.2	53.2	44.7	52.8
SD	6.1	5.6	6.5	8.5	6.1
rel. lymphocyte count (%), female	64.7	57.8	54.6	47.6	53.3
SD	6.4	8.7	7.1	8.2	6.8
rel. monocyte count (%), male	0.4	0.4	0.3	0.3	0.1
SD	0.7	0.6	0.4	0.4	0.3
rel. monocyte count (%), female	0.3	0.4	0.1	0.2	0.1
SD	0.5	0.5	0.3	0.4	0.3
rel. eosinophil count (%), male	0.5	1.5	2.7	1.8	2.7
SD	0.9	1.4	1.9	1.6	1.7
rel. eosinophil count (%), female	0.8	1.6	2.4	2.5	1.2
SD	1.1	1.8	2.9	3.3	1.0
rel. basophil count (%), male	0.0	0.1	0.0	0.2	0.2
SD	0.0	0.3	0.0	0.4	0.5
rel. basophil count (%), female	0.1	0.0	0.2	0.0	0.2
SD	0.2	0.0	0.3	0.0	0.4

A, B, C: Mean values with the same letters are not significantly different ($p < 0.01$)

Table 4.5 **Haemoglobin, haematocrit, MCHC, rel. neutrophil, lymphocyte, monocyte, eosinophil and basophil counts of 20 male and 20 female pastel mink from 15/8 until 15/7 (one year). Mean values and standard deviations (SD). Part II.**

Male:n = 20
Fem.:n = 20

DATE

15/1	15/2	15/3	15/4	15/5	15/6	15/7
12.05C	12.54D	12.17CD	11.12AB	11.43AB	11.86BC	11.18C
0.37	0.43	0.70	0.30	0.43	0.65	0.50
11.42C	11.49C	11.61C	10.18A	11.05B	11.67C	10.68AB
0.30	0.50	0.56	0.56	0.43	0.43	0.84
60.0D	62.0D	61.0D	56.0B	56.0B	56.0B	54.0AB
2.4	2.2	2.5	1.9	1.9	3.1	2.0
58.0C	58.0C	59.0C	51.0A	56.0B	56.0B	54.0AB
2.8	2.1	1.6	2.4	2.1	2.1	2.0
20.1A	20.1A	19.9A	9.9A	20.4A	21.2B	21.1B
0.6	0.7	0.6	0.6	0.4	0.5	0.4
19.8A	19.9A	19.8A	19.9A	19.9A	20.8B	20.7B
0.6	0.6	0.6	0.6	0.4	0.5	0.5
50.2	54.3	60.1	51.2	43.6	40.3	41.0
9.5	6.9	5.8	7.4	10.0	11.0	10.0
48.0	59.3	54.6	51.2	43.6	40.4	38.5
7.0	4.2	6.6	5.8	4.8	5.8	7.5
48.7	43.1	37.8	44.5	53.5	57.4	54.5
8.5	4.9	5.6	4.6	9.4	10.2	9.3
49.4	39.5	42.3	47.3	53.9	57.1	58.2
6.9	4.4	4.6	5.5	4.4	5.9	6.5
0.2	0.2	0.0	0.0	0.1	0.2	0.2
0.4	0.3	0.0	0.0	0.3	0.4	0.3
0.6	0.2	0.0	0.0	0.2	0.1	0.2
0.6	0.4	0.0	0.0	0.0	0.3	0.4
0.8	2.1	1.8	4.1	2.8	1.8	1.0
1.1	2.2	2.2	3.0	2.2	2.3	0.9
1.5	1.8	1.8	0.8	2.1	2.0	4.3
2.0	1.5	1.5	1.2	2.3	2.0	2.3
0.1	0.2	0.0	0.2	0.0	0.0	0.0
0.3	0.4	0.0	0.4	0.0	0.0	0.0
0.3	0.1	0.0	0.0	0.0	0.1	0.0
0.4	0.3	0.0	0.0	0.0	0.3	0.0

A, B, C: Mean values with the same letters are not significantly different ($p < 0.01$)

4.3.1.3 Investigations on heritability

In all three sample series differences between progeny groups were detected. An example of this is illustrated in figure 4.8.

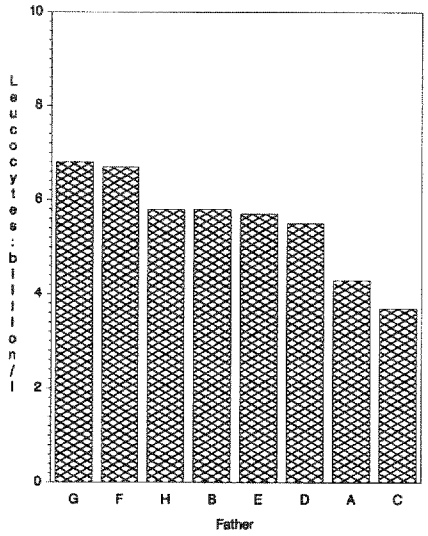

Figure 4.8 Genetic strain difference in leucocyte count.

The change of environment including management but excluding feed type and feed quality had some effect on blood count values and on the activity of the enzymes ASAT and CK.

In special stress conditions the differences between farm populations and progeny groups were emphasized. The farm of origin was largely responsible for the variation in blood count values whereas the progeny groups were responsible for the variation in ASAT and ALAT values.

In stress conditions also the early growth period from birth to weaning was shown to be of great importance for the animals ability to resist negative effects of feed and /or environment. (Brandt & Lohi, 1986; Brandt, 1986; Lohi et al., 1986; Lohi & Brandt, 1988).

The heritability of haematological and enzymological variables will be analysed on the total material of 929 blood samples as an integral part of a future PhD thesis.

4.3.2 Evaluation of clinico-pathological parameters as (subclinical) disease predictors in mink.

Clinico-pathological and epidemiological methods were applied on problem mink farms in numerous outbreaks and cases of greasy mink kits (early and late type), nursing disease, cystitis, nephritis, urolithiasis, sudden death syndromes (e.g. Myocardial degeneration).

Epidemiological principles were revised and examples of utilization of ideas of feasible implementa-

tion in fur animal production were given (Brandt, 1988c).

In another communication the sensitivity, specificity and predictive values of diagnostic tests were exemplified in mink (Brandt, 1988c; Hau et al., 1988).

Clinical and haematological profiles have been given for nutritional myocardial degeneration syndrome and nursing disease - an example is shown in table 4.7 (Brandt, 1989).

- Myocardial degeneration (study 2.a).

In this study there was a clear demonstration of suboptimal haematological indices and elevated enzymological values (e.g. creatine kinase) in otherwise healthy mink from strains of mink in which nutritional myocardial degeneration developed (Brandt & Lohi, 1986).

- Wet Belly Syndrome (study 2.b).

In this study of Wet Belly Syndrome (WBS - defined as a cronic peripraeputial hair and skin discolouration, alopecia and atopia as a result of incontinentia and cronic soaking by urine) 15 urine and 3 blood parameters were measured and correlated to the score of WBS. Thus urine protein and blood, plasma urea and creatinine were elevated in WBS mink (Brandt & Clausen, 1988).

- Validity of CK-test (study 2.c).

Total CK activity (creatine kinase) was correlated to the histopathological scores of myocardial degeneration (MCD) in mink of different families, sexes, fed different diets and kept under different environmental conditions. There was only sign of MCD when the plasma activity of CK exceeded 200 U/l. The validity of CK as a screening test parameter for MCD was fair. See table 4.6 (Brandt, 1986c).

Table 4.6 The sensitivity, specificity and predictive value of the plasma-CK-test for MCD in mink.

	Item		
a)	Number in the population	380	
b)	Histological MCD	37	
c)	Histological MCD-free	343	
d)	True positive values	11	
e)	False positive values	1	
f)	True negative values	342	
g)	False negative values	26	
h)	Total positive values (d+e)	12	
i)	Total negative values (f+g)	368	
j)	Sensitivity (dX100/d+g)	29.7	%
k)	Specificity (fX100/f+e)	99.7	%
l)	Predictive value for + test (d/h)	91.7	%
m)	Predictive value for - test (f/i)	92.9	%

- Nursing disease (study 2.d).

Nursing disease in lactating mink has a poorly defined aethiology, but distinct pathological characteristics. The disease has been associated with dietary stress, high protein, sodium excess, lactational state, selection/genetics and disturbed water balance combined with suboptimal management conditions.

In this study the following clinicopathological conditions were found to be characteristic. Weak, anorectic, low weight females showed signs of ketoacidosis with low plasma insulin, potassium and high haematokrit and carbamide combined with patho-gnomonic vacuolization of the kidney tubulicells (Brandt & Henriksen, 1987b).

- Variation of fatty acid content in mink milk (study 2.e).

Fatty acid analysis was performed on milk samples from mink females with greasy kits from different feed kitchens (FK). The percentage of total saturated and monounsaturated, omega-3-poly unsaturated, and omega-6-poly unsaturated were calculated. There

was a large individual variation which overshadowed a tendency of greasy kit milk having a larger content of omega-3 fatty acids. See table 4.7 (Brandt, 1987b).

Table 4.7. Total content of polyunsaturated fatty acids of milk (% of total milk fat) in females with and without greasy kits.

Fatty acids	Females with normal kits	Females with greasy kits
Number of samples	18	33
C18:3ω3	1.0	1.3
C20:5ω3	0.3	1.0
C22:5ω3	0.2	0.3
C22:6ω3	1.0	2.7
Total ω3	2.5	5.3
C18:2ω6	8.8	12.4
C22:5ω6	0.4	0.8
Total ω6	9.2	12.8

4.3.3 Experimental investigations

The following questions were studied under experimental conditions:

a) minerals: iron, copper and zinc turnover with emphasis on the haematological and enzymological age development, mineral balance and fur development,

b) diseases: 1. Nursing disease
2. Nutritional myocardial degeneration

- Effect of dietary microminerals (study 3.a.1)

The effect of equivalent ferro-EDTA, amino-acid chelated iron, iron sulphate and ferri glutamate dietary supplements (50ppm) on

growing mink kits was monitored from weaning until pelting using haematological indices, relevant clinico-pathological variables and pelt quality criteria.

A development of microcytic anaeemia and suppressed growth was found as a sequel of the generally low iron supplementation level. At 5.5 months of age 4 mink kits from each group were submitted to a digestibility trial for the determination of the excretion (balance) of iron, zinc and copper.

No effect of the iron source on the balances was seen. Most of the ingested minerals were excreted in the faeces.

Zinc of EDTA iron supplemented mink was excreted significantly more in the urine (Brandt & Mejborn, 1987).

The effect of dietary iron and fat sources on haematological indices was shown in 4 figures merging results from 4 related experiments (Brandt & Lohi, 1986; Brandt & Mejborn, 1987).

- Effect of chelators (study 3.a.2).

There was an anaemic effect of dietary Vantosil ® probably due to its complex binding of copper and zinc, on the haematological status in mink kits (Mejborn & Brandt, 1988).

- Effect of glucosinolates (study 3.a.3)

Glucosinolates in concentrations of 11, 46, 86 mcmol/g in 10% dietary rape of the total feed was fed mink kits from weaning until pelting. The result was a depressed growth rate, haematological indices and hyperthrophic thyroids and depressed plasma thyroxin concentrations (Brandt & Henriksen, 1987c).

- Dietary factors influencing nursing disease (study 3.b).

Having characterised the clinico-chemical and clinico-pathological characteristics of nursing disease, an experiment was conducted to illuminate some of the aethiological hypotheses:

The main effect of dietary factors (unbalanced fat, protein and carbohydrate ratios) on the development of nursing disease symptoms and performance in lactating mink.

A negative energy balance with characteristic acid-base disturbances with hypo-potassiaemia and metabolic acidosis was shown to be the most aggravating factor for the development of nursing disease (Brandt et al., 1986).

Drinking water supplemented with potassium was shown to be an effective preventive measure against nursing disease in mink (Brandt & Henriksen, 1987b).

- Nutritional myocardial degeneration (study 3.c).

Nutritional myocardial degeneration was experimentally studied in parts under the project: The Interrelationship between Dietary dl-alpha-Tocopheryl Acetate, Sodium Selenite, Antioxidants and Polyunsaturated Fatty Acids in Mink. The results are published by Brandt (1986b).

4.4 Conclusion

In the project physiological, genetic and environmental variations in haematological and clinico-chemical parameters in mink and health surveillance in mink populations were examined.

Reference values have been determined for all significant haematological and clinical parameters as shown in the Appendix table 4.8.

The determination of heritabilities of the measured parameters in the massive data collected has been provisionally analysed showing significant environmental and genetic variations in both haematological and clinico-chemical variables. The data will be analysed further together with the production data as an integral part of a PhD thesis.

The work concerning the applicability of clinical pathology, clinical biochemistry and haematology in health surveillance in mink populations may in part only serve as appetizers and pilot investigations. Based on the results a general Health/production-Check-Profile (HCP) in mink can be outlined to include parameters of: haematology (erythrocyte and leucocyte indices), enzymology (ASAT, ALAT, alkaline phospatase and CK) and urology (density, blood and pH).

The general Health/production-Check-Profile in mink will be applicable in a health surveillance programme for screening a given population for sub-clinical symptoms concerning both environmental/nutritional diseases and more specific agent, hereditary or systemic diseases in mink.

The HCP should not be confused with a profile designed for a specific diagnosis on individual mink level. This level is typically the next step in the surveillance

programme, at which special clinico-chemical analyses if necessary, together with bacteriological, virological or serological, toxicological and histopathological analyses can be used.

After this diagnostic phase, an intervention can be initiated, such as treatment regimes (vaccination, medical or dietary supplementation) or changes in the management, environment or most radically - culling.

The results presented here underline that it is of paramount importance to any use of clinico-biochemical and/or haematological parameters to register the sex and exact age of the animal.

Future activities:

a) Statistical work up of data in connection to environmental and pelt data.

b) Test of the predictive value and validity of the proposed Health/production-Check-Profiles in mink on a bigger scale to establish the relationship between the test results in:

1) lines of female/male breeders and the production results,
2) farm animals being tested within the plasmacytosis test programme conducted by the Danish Fur Breeders Association,
3) problem farms with clinical symptoms.

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APPENDIX

Table 4.8. The status of haematological, enzymological, vitaminological metabolite method development and application.

Status of analysis:

1. being applied at the department laboratory
2. applied at the department laboratory
3. reference values and ANOVA have been determined in normal adult mink in order to determine:
4. the effect of age
5. the effect of sex
6. the effect of genotype
7. the effect of environment/feed kitchen etc.
8. the effect of certain diseases

All analyses were made with plasma, but might as well be made with serum.

HAEMATOLOGY

Analysis	Status	Method
B-Haemoglobin (Fe)	2, 3, 4, 5, 6, 7, 8	Cyano methhaemoglobin
B-Erytrocytes	2, 3, 4, 5, 6, 7, 8	Elec. counting (Linson)
B-MCV	2, 3, 4, 5, 6, 7, 8	Elec. calc. (Linson)
B-MCHC	2, 3, 4, 5, 6, 7, 8	Elec. calc. (Linson)
B-HCT	2, 3, 4, 5, 6, 7, 8	Elec. calc. (Linson)
S-LIBC	2, 3, 5, 8	BM - Bathophenanthrolin
S-TIBC	2, 3, 5, 8	BM - Bathophenanthrolin
B-Thrombocytes	2, 3, 5, 8	Elec. counting (Linson)
B-Reticulocytes	2, 8	Microscopy counting
B-Leucocytes	2, 3, 4, 5, 6, 8	Elec. counting (Linson)
B-Eosinophils	2, 3, 4, 5, 6, 8	Microscopy counting
B-Differential count	2, 3, 4, 5, 6, 8	May-Grünwald-Giemsa

ENZYMOLOGY

P/S-ALAT	2, 3, 4, 5, 6, 8	BM DGKC -standard method
P/S-Albumin	2, 8	Bromcresol Green
P/S-Amylase	2, 8	BM DGKC -standard method
P/S-ASAT	2, 3, 4, 5, 6	BM DGKC -standard method
P/S-BASP	2, 3, 4, 5, 6, 8	Bassey/Hausanen
F-Chymotrypsin	2, 8	BM DGKC -standard method
P-Chymotrypsin	2, 8	BM DGKC -standard method
P/S-CK (Creatine kinase)	2, 3, 8	BM DGKC -standard method
P/S-CK-isoenzyme	(2)	Agarose Electrophoresis
P/S-LDH	2, 3, 8	BM DGKC -standard method
F-Trypsin	2, 8	BM DGKC -standard method

VITAMINS

P-Ascorbic acid	(2), 3, 8	Hansen
O-Ascorbic acid	(1), 8	Hansen
P-vitamin-K 2)	2, 3, 8	HPLC
P-α-tocoferol 2)	2, 3, 8	HPLC
O-α-tocoferol 2)	2, 3, 8	HPLC

METABOLITES

B-Base excess	2, 3, 4, 8	Siggaard/Andersen
P/S-Bilirubin	2, 3, 8	Jendrassik-Grof
P/S-Carbamide	2, 3, 4, 5, 6, 7, 8	Urease/GLDH
P/S-Cholesterol	2, 3, 4, 8	Liebermann-Burchard
P/S-Creatinine	2, 3, 4, 8	Enzymatic - Gutmann
P/S-Bile acids	1	mod. BM
P/S-Glucose	2, 8	Hexokinase
P/S-Lactate	2, 8	Noll.F
M-Lactose	2, 8	mod. BM
P/S-Triglycerides	2, 8	Enzymatic-mod. Wahlefeld

MINERALS

S-Calcium	2, 3, 4, 5, 6, 7, 8	mod. Ellmann
S-Cloride	2, 3, 4, 5, 6, 7, 8	Mercuri Thiocyanate
S-Phosphorus	2, 3, 4, 5, 6, 7, 8	Molybdate/vanadate
S-Iron	2, 3, 8	BM - Bathophenanthrolin
S-Copper	2, 8	BM - Bathocuproin
S-Potassium 1	2, 3, 4, 5, 6, 7, 8	Flame emission
S-Magnesium	2, 3, 4, 5, 6, 7, 8	Calmagite
S-Sodium 1	2, 3, 4, 5, 6, 7, 8	Flame emission

PROTEINS

P/S-Ceruloplasmin (IEA)	2, 8	Immuno Electrophoresis
P/S-Fibrinogen (IEA)	(2, 8)	Immuno Electrophoresis
P/S-b-Lipoprotein (IEA)	(2, 8)	Immuno Electrophoresis
P/S-Lipoproteins(AE)	2, 8	Agarose Electrophoresis
P/S-Haptoglobulin (IEA)	1	Immuno Electrophoresis
P/S-Protein Total	2, 8	Biuret
M-Protein Total	2, 8	Biuret
M-Proteins	2, 8	Agarose Electrophoresis
P/S-Proteins	2, 8	Agarose Electrophoresis
P/S-Transferrin	(1)	Immuno Electrophoresis
P-IgA	2, 8	Immuno Electrophoresis
P-IgG	2, 8	Immuno Electrophoresis
P-IgM	2, 8	Immuno Electrophoresis
M-IgA	2, 8	Immuno Electrophoresis
M-IgG	2, 8	Immuno Electrophoresis
M-IgM	2, 8	Immuno Electrophoresis

URINE ANALYSIS

U-pH	2, 3, 8	BM-stick
U-Spec. gravity	2, 3, 8	Refractometer
U-Albumin	2, 3, 8	BM-stick
U-Glucose	2, 3, 8	BM-stick
U-Ketones	2, 3, 8	BM-stick
U-Blood	2, 3, 8	BM-stick
U-Bilirubin	2, 3, 8	BM-stick
U-Erythrocytes	2, 3, 8	BM-stick
U-Epithlial cells	2, 3, 8	Microscopy
U-Bacteria	2, 3, 8	Microscopy
U-Protein	2, 3, 8	BM-stick
U-Proteins	(1)	Agarose Electrophoresis

B-full blood, F-faeces, M-milk, O-organ, P-plasma, S-serum, U-urine.

BM - Boehringer Mannheim.

DGKC - Deutsche Gesellschaft für Klinische Chemie

1) in collaboration with the laboratory of the Danish Fur Breeders Association.

2) in collaboration with the State Veterinary Serum Laboratory.