Landbrugsministeriet
Statens Husdyrbrugsforsøg

Research in Fur Animals at the National Institute of Animal Science

Present Status and Future Perspectives

Contribution in honour of Gunnar Jørgensen

Outi Lohi and Christian Friis Børsting (Eds.)

720 Beretning

Foulum 1993

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The National Institute of Animal Science was founded in 1883 and is a governmental research institute under the Ministry of Agriculture.

The aim of the institute is to carry out research and accumulate knowledge of importance to Danish animal husbandry and to contribute to an efficient implementation of the results to the producers.

In the research great importance is attached to the utilization of resources, environment and animal welfare and to the quality and competitiveness of the agricultural products.

The National Institute of Animal Science comprises five research departments, a Central laboratory, a department for Farm Management and Services, and a Secretariat. The research departments comprise: Dept. for Animal Physiology and Biochemistry, Dept. for Research in Cattle and Sheep, Dept. for Research in Pigs and Horses, Dept. for Research in Poultry and Rabbits, and Dept. for Research in Fur Animals.

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Report from the National Institute of Animal Science, Denmark

Outti Lohi and Christian Friis Børsting (Eds.)
Dept. for Research in Fur Animals

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Manuskriptet afleveret april 1993

Trykt i Frederiksberg Bogtrykkeri a·s 1993

Preface

It is not necessary to go back very far to obtain an overall survey of fur animal research. The research did not really start until after World War II, when the National Institute of Animal Science and the Danish Fur Breeders Association succeeded in establishing the first fur animal research farm. Despite the late beginning, much has, however, been achieved. Like the rest of the fur animal trade, research in fur animals has been characterized by "entrepreneurs", who have understood how to exploit existing possibilities to create fruitful results. Today a lot of good and sound research results exist, all contributing to Denmark's position as one of the world's leading countries in the fur animal field.

One of the entrepreneurs, who has for more than 30 years been the central figure of fur animal research, is head of department Gunnar Jørgensen. A survey of fur animal research at the National Institute of Animal Science must thus inevitably start with his work. To honour his enthusiastic work with fur animals this report is therefore dedicated to Gunnar Jørgensen at his 35th anniversary at the National Institute of Animal Science on the 30th of April 1993.

The first chapter also pays tribute to the international research cooperation, to which Gunnar Jørgensen has been seriously and actively committed. Under the leadership of Gunnar Jørgensen, the Department of Fur Animals has reached a central and fruitful position in international cooperation. In appreciation, this report is published in English. It is our hope that the numerous international contacts created by Gunnar Jørgensen all over the world will continue to contribute to the international exchange of research results.

The report is also a tribute to the cooperatin between the National Institute of Animal Science and the Danish fur breeders and their organizations. The activities of the Department for Research in Fur Animals have been implemented in a close and fruitful collaboration with the Danish Fur Breeders Association and its related organizations. The future perspectives for fur animal research outlined in this report are also a future challenge for the fur animal trade. The optimum results and improvements can only be achieved by continued close cooperation between trade and researchers.

March 1993
A. Hjortshøj Nielsen
Director, NIAS

Gunnar Jørgensen

Gunnar Jørgensen

1933 Born on the 17th of April in Ringkøbing, Denmark.
1940-47 Primary and secondary school.
1947-52 Continuation School and assistant on different Danish farms
1952-54 Military service at the 9th and 7th Regiments.
1954-55 Course in Agricultural Management at Ladelund Agricultural School.
1955-58 Agricultural student at The Royal Veterinary and Agricultural University, Denmark.
1958 M.Sc. in agriculture.
1958 Employed as a scientific assistant by the Dept. for Research in Pigs, Horses and Fur Animals, at The National Institute of Animal Science, (NIAS), Denmark.
1965 Acting Chief Scientist in the newly established Department for Research in Fur Animals under NIAS.
1970-80 Chief Scientist at NIAS, Dept. for Research in Fur Animals.
1973-74 Chairman of the Committee on Nutrient Requirements and Feed Quality under The Scandinavian Association of Agricultural Scientists, Division for Fur Animals.
1973-80 Secretary to the Research Board in Fur Animals under NIAS.
1974-87 Chairman of The Committee for Feeding under The Scandinavian Association of Agricultural Scientists, Division for Fur Animals.
1975 Initiator and chairman of the Organizing Committee of the 1st International Scientific Congress in Fur Animal Production, Helsinki, Finland (April 1976).
1976 Initiator and responsible editor for the scientific periodical "SCIENTIFUR".
1978 Appointment as the Scandinavian representative of the Subcommittee on Fur Bearer Nutrition, National Research Council, Commission on Natural Resources, US.
1980-89 Acting Head of Department of the Department for Research in Fur Animals, NIAS and appointed member of the NIAS Commission and the Commission on Fur Animals under NIAS.
1980 Chairman of the Organizing Committee of the 2nd International Scientific Congress in Fur Animal Production, Vedbæk, Denmark (April 1980).
1985 Editor of "Mink Production". Also published in Japanese and Spanish.
1987 Board member in the Division for Fur Animals, Scandinavian Association of Agricultural Scientists.
1987 Hon. consultant to the International Welfare Organization for Farmed Fur Animals.
1987 Editor of "Vakre Pelsdyr – og deres fargegenetikk" (Norwegian) ("Beautiful Fur Animals – and their colour genetics"). Also published in Danish, Swedish, English, and Japanese.
1988-91 Chairman of the NIAS Publishing Commission.
1988 Initiator and board member of the International Fur Animal Scientific Association.
1989 Appointed Head of Department at NIAS.
1991 Received Distinguished Leadership Award for service to the International Scientific Cooperation regarding fur Animals.
1992 Appointed vice-president of the International Fur Animal Scientific Association.

Publications:

142 scientific publications, 112 literary publications and congress papers.

In addition to this, several invited lectures in the US, the UK, the Netherlands, Germany, Poland, Czechoslovakia, Yugoslavia, Hungary, Greece, Korea, and Japan.

Contents

Tributes to Gunnar Jørgensen

Head of Department, M.Sc.Agr. Gunnar Jørgensen, and the Department for Research in Fur Animals

Niels Glem-Hansen

Nothing was written in the stars about Gunnar Jørgensen being the leading scientist of the world within fur animal science. Nevertheless, he turned out to be so.

For all of us who have had the privilege to know him – and that is more or less colleagues all over the world in the field of fur animal science – Gunnar has always been an inspiration with his enthusiasm and interest in all aspects concerning future scientific prospects in any possible field of fur animal science. The fur industry, which I have been representing for the last decade, has in Gunnar had a collaborator and a real head of the department with a very advanced understanding of how the scientific field can benefit the industry. However, I feel that Gunnar has not always received the credit he fully deserves for his persistent battle for the fur industry.

Gunnar, please consider this an appreciation which you deserved many years ago.

When Gunnar initiated his career in 1958, he was appointed at the Department for Research in Pigs and Horses, to which the experimental farm for fur bearing animals was attached. Even though he did not have the proper title, and probably not the salary either, he has in fact been responsible for the entire scientific work at the experimental farm already from 1960.

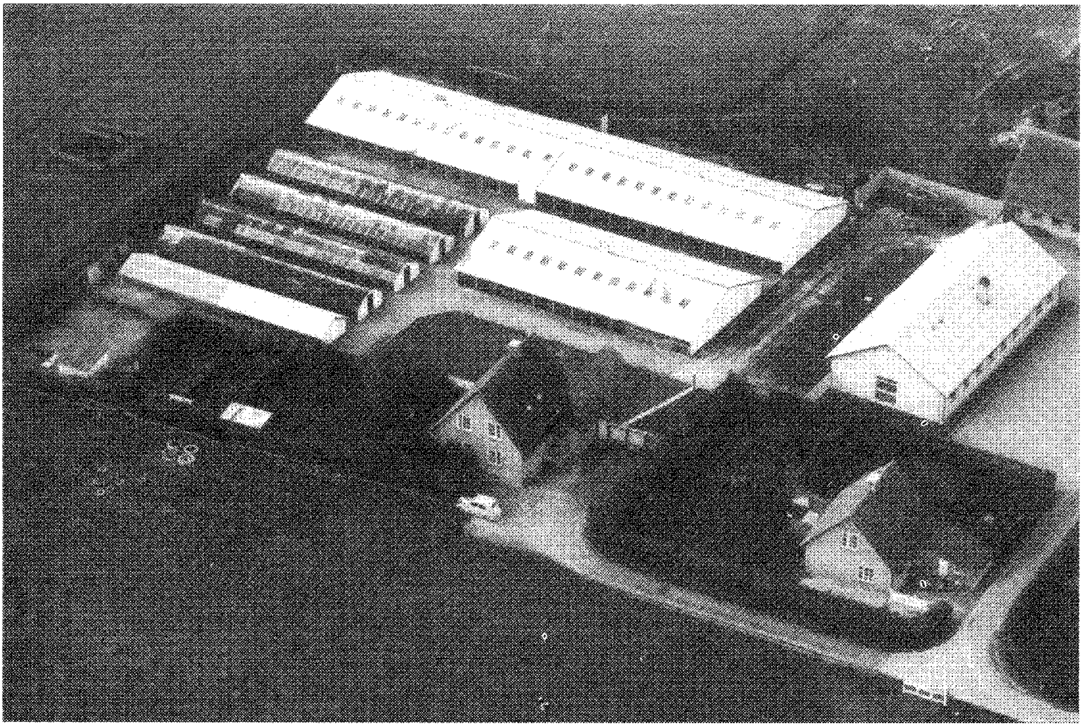
Compared to animal husbandry with other species, fur bearing animals are rather new as a subject for scientific investigations in Denmark. The first experimental farm for fur bearing animals in Denmark was established in 1947 at Trollesminde. At that time the board of the Danish Fur Breeders Association had the visions and they were not afraid to use some money to realize them, as a matter of fact they decided to empty the cash account to build the experimental farm which was then handed over to the state as a gift.

In 1963, the experimental farm was expanded and equipped with up-to-date facilities. This was again financed by the association.

During the years from about 1975 until the institute was moved to Foulum, the staff and the scientific activities were gradually increased even though it was sometimes hard to find the necessary space for the additional personnel.

Gunnar has always had more visionary ideas than could ever be carried through. He can be depressed and angry when one of his good ideas cannot be realized, but never for very long. Quite often he has also succeeded both in matters concerning his own department and in his world wide ideas. The establishment of the International Congresses in Fur Animal Science was his idea, SCIENTIFUR is a product of his imagination, and so is the International Fur Animal Scientific Association (IFASA). He was the prime mover when these – for the fur animal scientists – important activities were to be established, and if it had not been for him, none of them would ever have succeeded.

Officially, Gunnar became the head of the department in 1989. During the years from 1958, when he was the single employee working with fur bearing animals, to 1989 he managed to turn the department, which predominantly carried out feeding trials with practical aspects, into a department covering practically all scientific disciplines of interest to fur production. During the same period, the research at the department was directed towards more basic re-



The new-built Research Centre Foulum. The fur farm is seen at the top to the right.

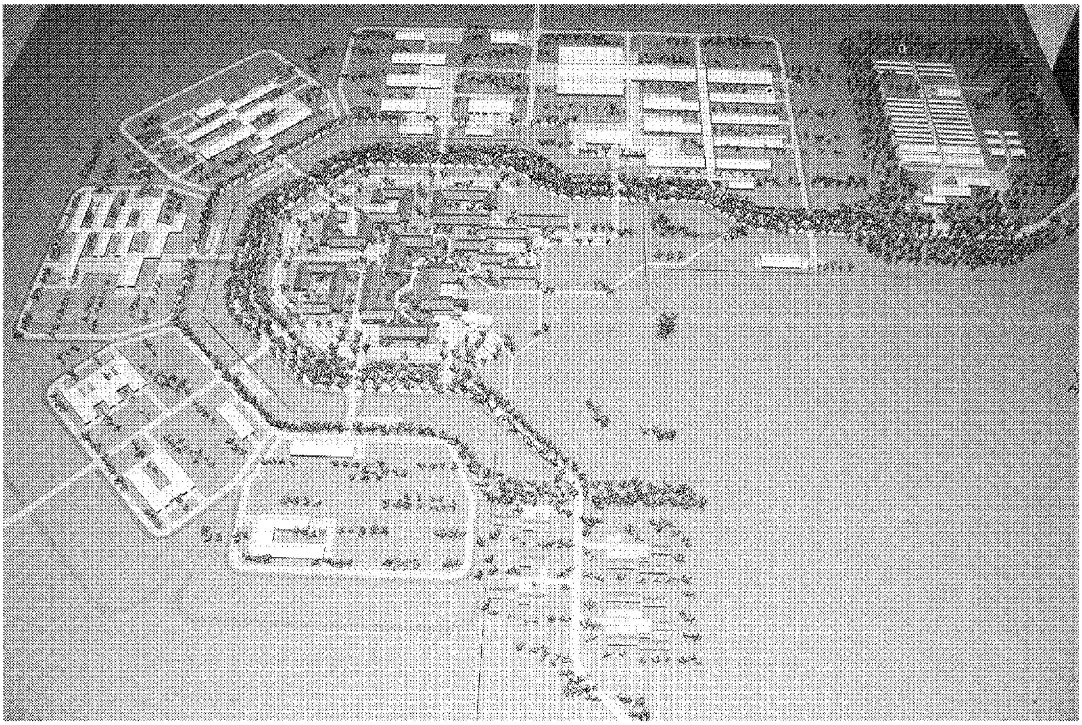
search, but still with the aim of solving the most urgent problems for the fur industry in the best possible way.

This has been done in a way, which can well be compared with international institutions, as can be read from an international evaluation of Danish agricultural research, which was carried out in 1992. From the panel's report I would like to cite the following:

"The Panel was impressed with the work of this Department, in nutrition, reproduction, applied physiology and behaviour, and genetics and breeding.

In the latter, and in particular with mink, Denmark is a world leader; the work on colour traits, which are determined by single genes, is of general importance, since little work has been done on colour genetics with other species, yet there are many homologies in inheritance across species. The studies on the economic value and genetic parameters of main pelt traits have been most important for fur animal scientists and breeders, and will be reinforced by new genetic information on reproduction, kit survival, and lactation, and on various aspects of hair 'quality'. The Department would need considerably more staff to tackle all these topics fully; the Panel concluded that this need should, wherever possible, be dealt with by cooperation, as in the present extensive collaboration between this Department and the Department of Animal Physiology and Biochemistry, as well as with other Nordic countries, rather than by expansion of the present staff and facilities. It should, however, be noted that the present method of allocating 'basic' funding at the National Institute of Animal Science has created particular problems in this Department because of the large year-to-year fluctuations in the values of fur products. Despite the high quality of the research, there have been relatively few international publications from the Department – at least in part because of the relatively low priority that is given to fur production in the principal 'publishing countries'."

This description was more or less the best



The fur farm at Trollesminde.

characteristic of a department the panel could rise to in the report. However, before we get too excited about these praising words, we should keep in mind what Gunnar has so often said, when the grants for scientific work have been discussed: "We are by far the largest producer of mink and, therefore, we should not expect anybody else to do the scientific work for us".

In 1972 the Danish government decided to move the National Institute of Animal Science from Trollesminde at Hillerød to Foulum close to Viborg in Jutland. When Gunnar and I (as his collaborator at the time) first planned the requirements for a department for fur bearing animals, we thought we asked for as much as we could expect to get, when we planned a department with 4 scientists. However, from 1972, when the planning was initiated, to 1987, when the Department for Fur Animals as the last department should realize the establishment of their research facilities, the yearly production of mink skins in Denmark had increased from 3 to 10 mill.

I think everybody knows how difficult it is to expand a governmental project once you have described the requirements. In this situation, the fur industry was fortunate enough to have a person like Gunnar to take care of its interests. I venture to assert that nobody but Gunnar would have had both the visions and the courage to fight for what he has got today, namely a staff of 9 scientists, a technical staff and facilities to carry out up-to-date research work on an international level. At present four sections are established

1. Section for genetics and reproduction, working with both quantitative and single gene traits. Genetic aspects concerning pelt characteristics as well as the reproduction capacity and how to take these factors into consideration in the breeding programmes have been given high priority. During the last few years this section has initiated the establishment of a gene bank for colour types in mink and foxes, the so called 'Zoo' of the department.

2. Section for applied physiology, nutrition and feedstuff evaluation is probably the most developed section, since this scientific field has been cultivated most intensively over the last fifty years. The most obvious advantage for the fur industry from this section has been the results from digestibility trials and experiments for determination of the requirements for important nutrients. However, the more basic studies to determine biochemical pathways and processes will be the basis for further progress in the future.
3. Section for hair and skin is of a rather new date. This is surprising, since hair and skin are what the fur farmers produce for their living. Of course, we have always evaluated the characteristics of the pelt, but until recently this was done entirely on a subjective basis. The most important aim of the section is concentrated on developing objective methods for measuring certain characteristics of importance and compare them to the subjective evaluation.
   Morphological and biochemical methods are important tools when defects in hair and skin are to be studied, whether the defect is of genetic, nutritional, behavioural or any other origin.
4. Section for ethology, management and environment is also rather new. Until recently, animal behaviour was not given much attention in research as well as in practice. To be honest it was the massive attack from the anti-fur people which enhanced the interest in this field from the fur industry. Anyway, whatever the reason, ethology has been accepted as an important scientific field also in fur bearing animals.

Due to the serious economic situation for the fur industry during the last four years the Danish Fur Breeders Association decided in 1992 to reduce the research work within the framework of the association dramatically and to cut down the support to the governmental research institutions. The consequences of this decision are a loss of expertise in the association. Especially scientists/advisers who have a great experience in interpreting the users' need for research and transmitting scientific results to the users will be missed. This situation emphasizes the importance of ensuring the activities and fully utilizing the facilities established at the National Institute of Animal Science. Gunnar has built up the frames for future research in fur animals for the industry to use.

We are many colleagues and friends (that is more or less synonymous when it concerns Gunnar) who owe you a lot. I think it counts especially for 'newcomers' into the crowd of scientists. Gunnar has always been very open to 'newcomers' which has made it easier for them to join the 'club'.

Finally, I want to wish you a happy birthday when you reach the age of sixty on the 17th of April and to congratulate you with your 35th anniversary on the 30th of April. I also wish you a happy retirement when you settle down in Norway. I am sure your retirement will not be too quiet, since you will still be engaged as the editor of SCIENTIFUR and will probably also be active in IFASA.

N. Glem-Hansen

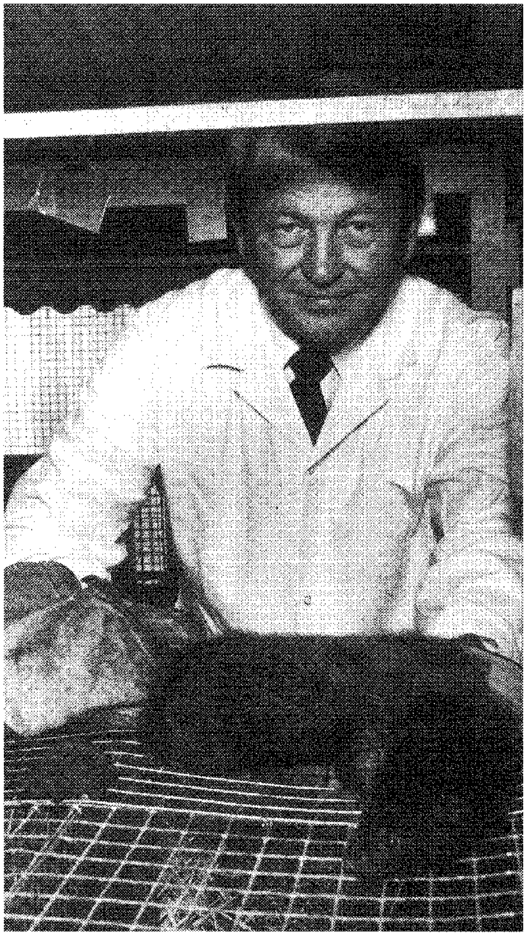
Gunnar and the Scandinavian Association of Agricultural Scientists

Åke Qvist

A young, jovial Danish agronomist with a twinkle in his eye and often taking a stimulating suck at his old pipe – this is how I first met Gunnar Jørgensen, researcher and chief scientist at the fur animal research farm in Hillerød. This was some time at the end of the 1950s. Little did I know that Gunnar was to become a colleague and a friend, and that in the decades to come we were to meet several times a year. It was in the fur animal trade and especially in the research activities that we shared the same interests and daily tasks.

It was only natural that the young fur animal research was attached to the Scandinavian Association of Agricultural Scientists (SAAS), an umbrella organization and symbol of cooperation within Nordic agricultural research. This happened already in 1947, just after the war. Within the framework of SAAS the researchers and scientists dealing with fur animals would meet regularly within the Division for Fur Animals.

As chief scientist Gunnar attended wholeheartedly to questions and problems in connection with fur animal feeding and was at an early stage the obvious representative and spokesman of this section within the Division for Fur Animals. He has been a member of the Committee for Feeding from 1973 to 1987 and a member of the board of the Division for Fur Animals since 1987.



Gunnar Jørgensen at work at the experimental station.

As the fur animal trade expanded in the Nordic countries, new as well as more feedstuffs were required. The standing question was: "What is good for fur animals and in which quantities?" Experiment and research activities were faced with ever growing demands for solutions, and both producers and researchers found the Scandinavian cooperation utterly important. The name Gunnar Jørgensen was often mentioned with respect to research as well as cooperation (not forgetting Hans Rimeslåtten, Gustav Åhman, Jouni Kangas, Jaakko Mäkelä, Anders Skrede, among others).

During his many years as research leader and scientist and as an extremely active member of the SAAS, Gunnar has often presented papers, reports and lectures on many different topics within the feeding of fur animals (especially of mink). He has almost always been present at SAAS congresses, annual meetings and smaller group meetings concerning feeding. He has participated in most of the SAAS congresses since the Oslo congress in 1959 (nine all in all) and in all division meetings from Vaasa 1961 to Espoo in Finland 1991 (22 altogether). To this can be added 5 international congresses on fur animal research from Helsinki 1976 to Oslo 1992.

Also at congresses and other international events outside the SAAS, Gunnar has been a good representative for the fur animal division. As an example he gave a lecture at the EAAP congress in Warsaw in 1975 (at which occasion many participants, at any rate from the Nordic countries, for the first time experienced the problems which could arise during visits to countries within the Eastern bloc).

Within the Division for Fur Animals we – his colleagues, advisers and farm representatives – have profited from his experience and advice in questions regarding dry feed as well as various fresh feeds, their protein content, energy value and digestibility. The vitamin requirements of fur animals has also been an interesting matter of discussion. In studies on feeding requirements and determination of nutritive values of various raw materials, Gunnar has in a very meritorious way 'had his finger in the pie'!

In connection with meetings within the Division for Fur Animals as well as other meetings, social gatherings have always been a part of the programme. At such gatherings, discussions regarding the fur animal trade were often prevailing, but also other, lighter topics of conversation were allowed and welcomed. At these enjoyable occasions Gunnar has always been welcome at the table.

That research and the Division for Fur Animals within the SAAS have always been dear to Gunnar, has also been expressed in the discussions regarding the economic resources of the Division for Fur Animals. More than anyone else he has often asked the question: "Why don't the fur breeder organizations transfer a part of all the resources they allocate to the SAGA organization for sales promotion purposes to product development instead?"

Numerous are the memoirs, articles and publications which, furnished with Gunnar Jørgensen's signature, have been distributed via the Division for Fur Animals to researchers and others connected with the fur trade. Many are also the friends and places in the Nordic countries that Gunnar has visited during his long career as a researcher. Even in the archipelago of Tammisaari on a rock at the beach you can find a greeting, scratched into stone – in real viking style – by Gunnar himself: "Greetings from Hilleroed".

I wish you a long and happy retirement.

Åke Qvist

Åke Qvist

Gunnar Jørgensen – The International Link

Einar J. Einarsson

In August 1992 the Vth International Scientific Congress in Fur Animal Production was arranged in Oslo, Norway. Besides being the Vth International Congress, it was the first one to be organized by the International Fur Animal Scientific Association (IFASA).

IFASA was established during the previous International Congress in Toronto, Canada, in 1988. As always, work like that is depending on enthusiastic people, and once again Gunnar Jørgensen was one of the key persons. He has always been able to see the importance of cooperation across borders.

Gunnar's long experience within the scientific and practical fields of the fur industry and all his personal contacts made it possible to rapidly build up the new organization with a net of contacts all over the world. This has been very valuable for IFASA during the time between the IVth and the Vth congress. Gunnar Jørgensen was elected as a member of the first board of IFASA, and during the first four years he acted in a working group together with the President. During that period he built up the secretariat of IFASA in Foulum, Denmark.

The Ist International Scientific Congress in Fur Animal Production was arranged in Helsinki, Finland, in April 1976. It was also at that time, Gunnar's idea of an English journal became a reality, and SCIENTIFUR was born. At the IInd International Congress in April 1980 in Vedbæk, Denmark, Gunnar himself was the organizer. In April 1984 the International Congress was held in Paris, France, and Gunnar was now an important consultant for the organizers. At the IVth International Congress in Toronto, Canada, in August 1988, Gunnar Jørgensen was one of the enthusiastic persons behind the foundation of IFASA. In August 1992 Oslo, Norway hosted the Congress, and now Gunnar was elected Vice President of the Board. Nearly one thousand people have participated in these five Congresses, representing more than twenty countries.

Gunnar Jørgensen has been strongly involved in all the five International Scientific Congresses in Fur Animal Production arranged over the past 16 years. We know that Gunnar will also be present at the VIth Congress in Poland in 1996, at the time celebrating his 20th anniversary of the International Congresses.

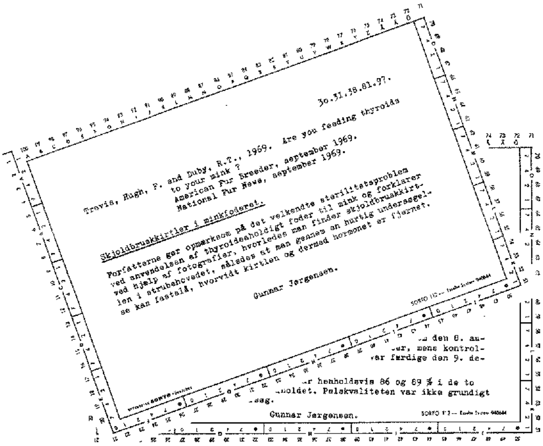
Gunnar Jørgensen has played an important role within the Division for Fur Animals of the Scandinavian Association of Agricultural Scientists, he is the father of SCIENTIFUR (now published by IFASA), an important founder and now the Vice President of IFASA, editor of books etc.

Gunnar is not only one of the grand old men of international fur animal science, Gunnar Jørgensen is the grand old man!

Einar J. Einarsson

From ‘Sorto’ to ‘Scientifur’

Outi Lohi



Esselte Sorto needle cards.

An important task in all scientific work is to follow up the research in other institutes and countries – a time consuming task even with modern technologies. The Scandinavian scientists therefore realized fairly early that this was a field where cooperation would save time and one person could work for all. In 1963 the Division board therefore decided to establish a recording of fur animal literature and a literature service.

The idea was to try to record all relevant literature beginning in 1950 and in the future to combine a short abstract with each recording. They also had found a – for its time – very advanced technology for the system, the Esselte Sorto needle cards. Both the references and the abstracts were to be written on special cards – one per card – which after a punching system would allow the user with a long needle to select out the records representing a special research area, subject or initial of the author.

The first editor for this literature service was Sven Sanne from the Swedish University of Agriculture in Uppsala and due to his serious contribution the service got a good start. A lot of reference cards were distributed already in 1964 and in February 1965 he sent out 380 abstracts of scientific reports on fur animals. Later on, both the editor responsibility and the task for writing abstracts were divided and circulated among advisers and researchers in the Scandinavian countries.

However, the later development was not as successful as the beginning. Even though the file of cards slowly increased all the time, it became obvious that it was hard to find volunteers to do the work and to have them cover the increasing amount of literature sources. Until 1975, altogether approximately 1000 reference cards were published. Meanwhile many scientists had started collecting their own reference files and used quite a lot of time in libraries. For example Gunnar Jørgensen had in connection with his research by then collected about 2000 articles on fur animals.

As a man of cooperation he therefore, at the First International Scientific Congress in Fur Animal Production in Helsinki 1976, launched his idea about an international information newsletter with abstracts of current research reports to be issued four times a year. The reactions from the audience were positive. The idea was greeted with great acceptance, yet many had doubts about how to make it work. However, especially the English speaking world realized it as a possibility of getting information about Scandinavian research.

Starting from that meeting Gunnar became the editor of Scientifur, and throughout all these 17 years he has carried not only the burden of the editorial work but also the responsibility of finding the financial possibilities to carry on.

Through these years Scientifur has become known all over the world, and one of the great values of it is that in a nutshell it transmits information from east to west, from north to south and vice versa. Thus, it has also initiated many contacts between scientists, which is important for all of us but has been so perhaps even more for our colleagues from the East-European countries. At its maximum, the total number of yearly subscriptions has been over 400, but it has lately declined slightly. In 1992 the total number of subscribers amounted to 350 and Scientifur was distributed to 25 different countries.

Scientifur started publishing mainly abstracts but later on the number of original reports has increased. It has also in increasing numbers informed the readers about new books of interest to fur animal researchers.

The best proof of Scientifur´s value is that, in spite of almost constant economic problems, the newsletter has survived and improved all the time. Fairly early Gunnar Jørgensen also realized that as the number of volumes increased it was necessary to build up an index system to improve the possibilities of searching for information published in Scientifur. The computer system was established in 1985 and an index including the first 10 volumes was published in 1987. Index number II including the following 5 volumes was published in 1992. In addition to the printed ones, a total index for all 15 years and including a search program is now available on disk for use in personal computers.

Many letters from researchers all over the world during the years are also a convincing expression of the value of Scientifur and the work Gunnar Jørgensen has put into it, and I am sure that I speak for many when I express the wish that this valuable work will continue also in the future.

Outi Lohi

Outi Lohi

SCIENTIFUR

SCIENTIFUR and the Scientist

Bruce D. Murphy

My bookshelf has many well-worn volumes of SCIENTIFUR that have accumulated over the last 12 years. I have found that this publication has been extraordinarily useful in the difficult task of keeping abreast with current literature. Even in these days of readily available databases, Current Contents on disk, etc., SCIENTIFUR always proves to be the best and most easily used source of information on fur animals.

One of the important roles of SCIENTIFUR has been to publish the English abstracts from journals from all over the world. Many of these journals are not easily available to me, and the abstract and the address of the author have allowed me to find articles which have been important to my research program.

A strength of SCIENTIFUR is its breadth. The coverage of all of the areas of fur animal science, from genetics to nutrition to pathology and economics, has broadened this reader, and has given me ideas about how to study the problems of fur animal reproduction.

A further important role of SCIENTIFUR has been in the development of a sense of community among those of us who undertake research on fur animals. It was in SCIENTIFUR that I saw the names of my Scandinavian, German and Polish colleagues, and became familiar with their work. It provides us with an audience for original papers, as well as a second forum for presenting our published work, as an abstract to the select, and interested group of scientists. SCIENTIFUR has been useful for advertising Fur Congresses, and these were the important impetus for the founding of IFASA.

The recent innovation of SCIENTIFUR indices of articles abstracted, and their availability on disk, is an important addition to the journal. It will prove very useful for retrospective searches of the literature in virtually every field.

One cannot speak of the value of SCIENTIFUR without recognizing that, as a son reflects his father, a journal reflects its editor. SCIENTIFUR is comprehensive, it is accurate, it appears with highly predictable frequency. SCIENTIFUR is dedicated to providing information, to keeping us up to date, and to serving the scientific community. This high quality and dedication are traits of Gunnar Jørgensen, who founded the journal and has fostered its development through the good and bad economic times of the fur industry. SCIENTIFUR also reflects Gunnar's impish sense of humor, with cartoons, illustrations, congratulations and amusing editorials. It is not clear to me how the journal will survive without him, I hope that it does. We owe a great debt to Gunnar, and I am grateful for this opportunity to thank him, in print, for his friendship, and for the service that he has provided to those of us with an interest in fur animals.

Bruce

Bruce D. Murphy

A Tribute to Gunnar Jørgensen

Tony Rietveld

As a researcher Gunnar has the natural desire to know, as a scientist he feels the need for intellectual intercourse. In his working environment he used the strong base of Scandinavian cooperation. However, his dreams and schemes were about larger objects.

In 1976 the scope became of a worldwide status with the First International Congress in Helsinki.

Scientifur was the brainchild of this Congress. Long before the Iron Curtain went up, Gunnar had made holes in it with his publication.

Information about furbearing animals from all over, was spread all over.

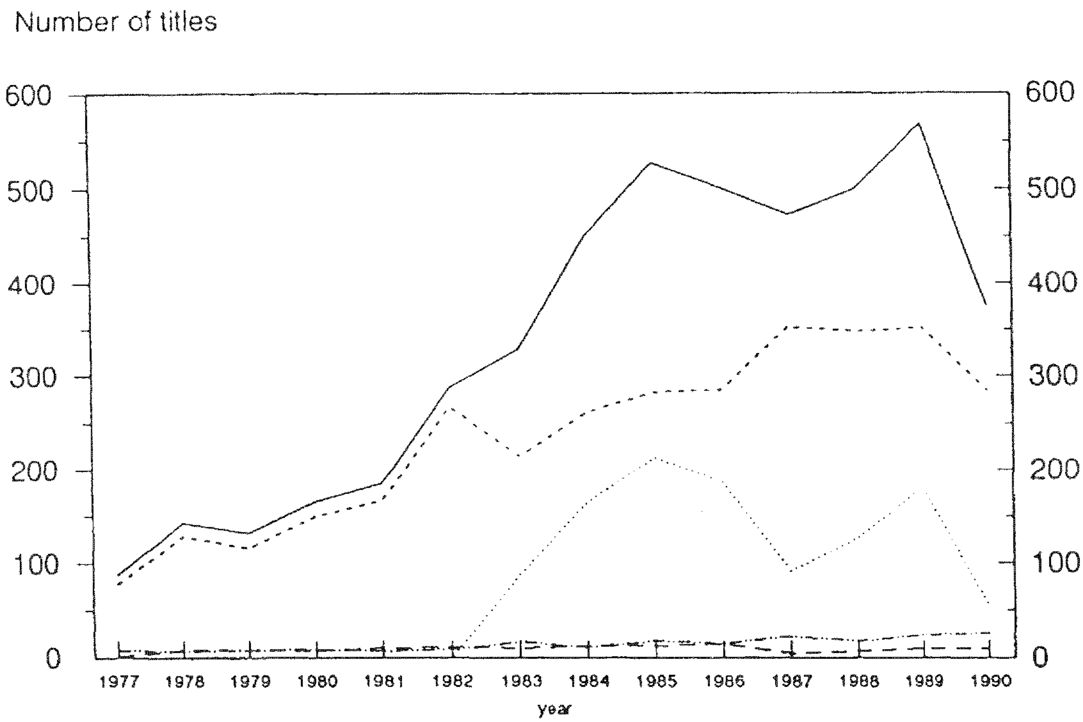
A visit with Gunnar, whether it was during a planeride, in his office in Trollesminde or over dinner, was always stimulating. For me personally the dissemination of knowledge has made the risky business of furfarming much more predictable, leaving as only real variable, the marketplace.

It is only natural to expect that we will hear from Gunnar in the future, in one of the issues of Scientifur.

Tony

Tony Rietveld

Information value of Scientifur.



Eastern Europe and Cooperation with Dr. Gunnar Jørgensen

Stanislaw Jarosz

The research activity of Dr. Gunnar Jørgensen has been known to me since he started his work at the National Institute of Animal Science.

Our personal contacts date back to the International Scientific Congress of Fur Animal Production in Helsinki in 1976. I was then fascinated by the personal charm of Gunnar Jørgensen and his engagement in research work as well as by his readiness to establish scientific cooperation with our research center in Cracow (then still behind the iron curtain). It was during the debates of the Congress in Helsinki that the idea of creating a specialistic research journal "SCIENTIFUR" devoted to fur animal breeding was born, its chief initiator being Gunnar Jørgensen. Since then our strict scientific cooperation has been lasting in the form of exchanging scientific information, frequent visits of Dr. Gunnar Jørgensen to our University of Agriculture in Cracow and our visit to his country as well as a training period of my research worker at the National Institute in Hilleroed and later at Tjele, which is all greatly appreciated by us. In our opinion the Department of Fur Animals of the National Scientific Institute at Tjele is one of the best all over the world, for which credit is also given to Dr. Gunnar Jørgensen. He was the main initiator, too, during the congress in Toronto (1988) of creating IFASA within the framework of which SCIENTIFUR is edited. SCIENTIFUR is highly estimated by people and institutions dealing with fur animal production and fur industry. Both myself and many people from Central and Eastern Europe consider SCIENTIFUR as a periodical fulfilling a very useful task in fast exchange of scientific information on a world scale and it should be continued. All the mentioned achievements are to a considerable degree the result of Dr. Gunnar Jørgensen's activities.

On the occasion of Dr. Gunnar Jørgensen's intention to retire, may I express my personal opinion as well as that of my colleagues of Slovakia and other countries of Eastern Europe that we have fully appreciated our cooperation with Dr. Gunnar Jørgensen and the great friendship which has always been accompanying our contacts. He deserves to be highly praised for all he has done to popularize knowledge of fur animal breeding. We wish him at heart the best of health and further enthusiasm in his professional and personal life. We hope to continue our enjoyable cooperation with Dr. Gunnar Jørgensen after he has retired.

Stanislaw Jarosz

Breeding and Genetics of Mink

Peer Berg

Introduction

Compared to production of other farm animals, fur animal production is a fairly new trade. Just a little over 100 years have passed from the very first efforts to keep mink in captivity, and real farm production has been going on for only about 70-80 years. In spite of this short time and the limited resources, a remarkable development has taken place.

Generally, the existing research capacity has in this period been used to solve the problems most urgent at the time, due to the close contact between producers and researchers.

In the very beginning, the main problems were domestication and reproduction problems. As the success in these fields increased the production, the need to solve problems in relation to feeding and periodically also in relation to diseases arose.

Later, along with improved marketing systems, the fur auctions and qualified grading of pelts, the importance of developing breeding programmes became obvious. For a long time, however, the problems of making reliable grading on the farm, collecting records and calculating breeding values seemed too hard to overcome. In regard to prediction of breeding values and use of selection indices a rapid development has taken place during the last decade.

At the moment, about 40% of Danish mink farms representing about 60% of the breeding stock use breeding programmes on personal computers.

Selection can only slowly change a population. The change is, however, permanent and additive over generations. This means that a given change is realized in all future generations, and further changes can be added in the following generations. Thus selection should be used to pursue long term goals rather than short term problems.

It is presently an important challenge for research to increase the basic knowledge about factors important in breeding programmes and methods for improving the effectiveness of programmes. In the present paper these problems are discussed on the basis of genetic theories and in connection with present and future research at the department.

Selection theory

Response to truncation selection per unit time can be predicted by the classical formula (Rendel & Robertson, 1950)

$$\Delta G = \frac{\Sigma r_{IA} i \sigma_A}{\Sigma L} \qquad [1]$$

where

ΔG is the selection response,
r_{IA} is the correlation between selection criterion and true breeding value,
i is selection intensity, a function of proportion selected,
σ_A^2 is the genetic variance, $\sigma_A^2 = h^2 \sigma_P^2$
L is generation interval,

and the sum is over genetic pathways, e.g. sexes, age groups (cohorts). The numerator is the sum of selection differentials in different cohorts, the difference between the mean of selected animals relative to the mean of the population. The denominator is the sum of generation intervals in different cohorts, the time from birth to the average time when offspring are born. For a one year old breeding male L is 1 and for a two year old breeding male L is 1.5.

To predict response to selection, knowledge about the genetic variation is necessary, and for prediction of correlated reponse to selection, by a modification of [1], genetic covariation has to be known. Selection response is also dependent on the accuracy of breeding values (r_{IA}) and thereby dependent on methods for prediction

Improving breeding programs for mink is a major task for the Department

of breeding values. Selection intensity, generation interval, accuracy of breeding values and the contribution of different cohorts depend on the breeding structure, the breeding plan. In the following these three elements will be reviewed, with the aim of describing the status and perspectives in mink breeding.

Genetic parameters

The review presented is the beginning of a database on genetic parameters in fur animals, and it is our hope that this database will form a basis for an international database on genetic parameters in fur animals. For a full classification of the estimates the trait, colour type, species, the time of measurement, method of estimation, sex, number of animals, mean, phenotypic variance, heritability and common litter effects (c^2), the reference and additional remarks are registered. In Table 1 some of the results are presented within traits. The number of estimates found is given, the range for all and the 50% in the middle, mean, standard deviation, and the median.

Generally a large variation is found between estimates published in different investigations. For example the heritability for litter size varies from 0.1 to 0.2.

A large variation is found for body weight. Berg (1993a) found that generally higher estimates were found in studies with a single population and lower estimates in studies with several populations, which is confirmed by this review. This might be due to an interaction with feeding regime (Berg, 1993a). Of the three measures of size the subjective evaluation of size has the lowest heritability (0.1 to 0.2), weight has a medium heritability (0.2 to 0.4) and body length has the highest heritability (0.4 to 0.6). This indicates that body length might be a good indicator of skin length, but for a general comparison one needs to consider the genetic correlation between these three measures and skin length.

The large variation in the estimates for subjectively evaluated traits can be partly due to effects of the person grading the pelts (Maciejowski & Slawon, 1980) and partly due to the statistical problems of estimation. Heritability estimates are underestimated in analysis of variance using a linear model (Gianola, 1982), which has been confirmed in mink (Berg, 1993b). The estimate depends on the frequency distribution (mean) of the trait (Gianola, 1982).

All the fur and skin characteristics considered in Table 1, have a medium to high heritability, indicating a potential to change these traits by selection. Colour seems to have a higher heritability when measured on live animals than on skins. It should be noticed, that components of overall quality, such as density of wool, and hair length also show a medium to high heritability and thus can be changed like overall quality. However, a high genetic correlation has been found between length of guard hair and wool hair (Berg, 1993b), showing that it is easier to change the absolute length of hair by selection than the relation between underfur and guard hairs.

Heritability estimates for many other traits can be found in the literature, e.g, guard hair coverage (Olausson & Lohi, 1978; Rosberg & Olausson, 1978), leather thickness (Reiten, 1977; Berg & Lohi, 1991), skin weight (Reiten, 1977; Einarsson, 1988a; Berg, 1993b), litter weight (Pastirnac, 1980), guard hair density (Reiten, 1977; Einarsson, 1988a), hair quality (Benthin, 1977; Reiten, 1977; Einarsson, 1988a; Lagerkvist & Lundeheim, 1990), clarity (Jónsson, 1971; Olausson & Lohi, 1978; Rosberg & Olausson, 1978), fur depth (Jónsson, 1971), singe, wet belly, elasticity, bended guard hairs (Reiten, 1977), metallic (Olausson, 1976; Reiten, 1977; Berg, 1993b), overall impression (Benthin, 1977; Rosberg & Olausson, 1978; Kenttämies & Vilva, 1988; Lagerkvist & Lundeheim, 1990), white spots (Fiedler & Siler, 1978; Rosberg & Olausson, 1978), and number of teats and parturition term (Narucka et al., 1982).

Prediction of breeding values

The first breakthrough for the use of more sophisticated methods of breeding value evaluations came with the development of computers and was speeded up with the development of personal computers.

Selection index has been described for a combination of weight, colour and quality using only measures on the animal (Narucka & Gedymin, 1979), for litter size (Christensen et al., 1984; Einarsson & Elofson, 1988) using information on relatives, and for a combination of litter size and overall impression using data on relatives (Rönningen et al., 1980). Lohi (1986) investigated the possibilities of including data from the auction in the calculation of selection indices.

The selection index yields the BLP (Best Linear Predictor) of breeding values given that genetic parameters and fixed effects (means) are known and random mating is performed. Einarsson & Elofson (1988) described a pedigree selection index for litter size including information on the dam, the full and half sisters of the dam and the granddam. Two different strategies are available for predicting breeding values in this setting of data (Henderson, 1963). The first strategy is to solve for one animal at a time (e.g. Einarsson & Elofson, 1988). This involves setting up a number of equations for records on the animal in question and the relatives that are included in the calculation. The number of equations can be reduced by combining equations of groups of relatives (e.g. full-sibs, offspring). The other strategy involves setting of a system of equation equal to the total number of animals and solving for all breeding values at one time. In the first strategy, the same record is processed many times as it is incorporated in the calculation for its own breeding value and in the calculations for all its relatives, but the second strategy can be impossible due to the size of the coefficient matrix and the task of inverting this matrix.

The algorithm described by Christensen et al. (1984) is an updating algorithm, exploiting that new records are conditionally independent of the index based on earlier records (conditionally on the breeding value of the animal, Christensen, 1981), and therefore breeding values can be updated sequentially. The method is in principle based on all available information.

This method can be modified to take account of non-random mating (Christensen, 1981).

Index selection gives a larger selection response than mass selection. Børsting (1989) has reported an 84% and 69% increase in selection response for litter size and fur quality, respectively. Rönningen et al. (1980) presented a method to calculate the discounted economic value of a selection index.

Henderson (1963) developed another method, simultaneous yielding BLUP (Best Linear Unbiased Predictor) of breeding values and BLUE (Best Linear Unbiased Estimators) of fixed effects given that genetic parameters are known at least to proportionality. Compared to a selection index, this method can utilise all available information (all relatives) to predict breeding values when formulated as an individual animal model, where mates are corrected for, which means that random mating does not have to be assumed and it takes into account that fixed effects (means) are not known but have to be estimated from the data. Further, it allows for prediction of other random effects, such as maternal genetic effects, maternal environmental effects, dominance and epistatic effects. Due to the difficulties in solving the BLUP equations directly, many different solving strategies have been proposed utilizing the properties of the infinitesimal model and the structure of the equations (e.g. Schaeffer & Kennedy, 1986; Misztal & Gianola, 1986). Iterative solutions have been proposed for mink (Berg, 1992) applicable on even small computing facilities. A multiple-trait animal model for size, quality and colour has been developed (Westwood et al., 1992).

Many traits in fur animals are not continuous and not normally distributed. Subjectively graded traits are classified in a restricted number of categories. These multinomially distributed traits have non-homogeneous variance and the variance and mean are not independent as assumed in the linear mixed model (Gianola, 1982). Methods have been developed to estimate breeding values in a mixed model for such traits (Gianola & Foulley, 1983; Harville & Mee, 1984; Gilmour et al., 1985), based on the threshold-liability model. These models are, however, not routinely used in mink. Generally, a very high correlation is reported for breeding values predicted under a linear and a threshold model (e.g. Meijering, 1985; Weller et al., 1988; Weller & Gianola, 1989). Most of these comparisons are, however, made in cattle under a sire model with relatively large progeny groups. The correlation might be lower in species with a lower accuracy of the breeding values. This has been confirmed in mink (Berg, unpublished). Secondly, the correlation might not be descriptive of the relative advantage of the methods. Generally, we are only interested in the ranking of the best animals and not the ranking of all animals. A higher realised selection response using a threshold model compared to a linear model for a binary trait has been found (Meijering & Gianola, 1985; Hoeschele, 1988).

Breeding plans and breeding strategies

Only few investigations on breeding schemes and optimization of breeding schemes are reported in fur animals.

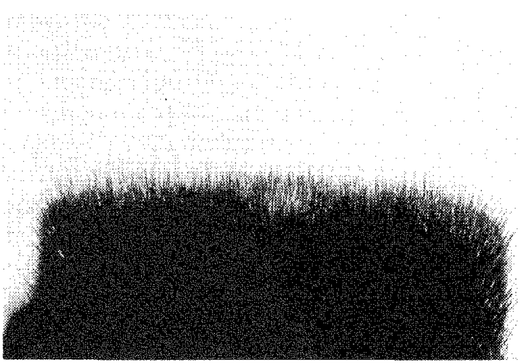
In a collaboration between all Scandinavian countries, the economic weights of skin traits have been estimated (Lohi et al., 1989). These investigations have shown that the relative importance of different traits (quality, colour, size, etc.) is nearly constant over time. This means, however, that the absolute importance of skin traits changes compared to other traits with a more constant absolute economic importance. Elofson (1981) estimated the value of an additional kit per female to be 27 to 56 SEK, depending on average litter size, % of barren females, cost of feeding etc. Lagerkvist (1992) estimated a value of 25 SEK with a price of pelts of 130 SEK and a value of 100 SEK with a price of pelt of 200 SEK. The value of an increase in litter size is a function of both the average production level, production costs and the price of pelts.

As the response to selection is realized in many generations and is cumulative, economic weights should reflect the expectations for the future. Thus, expectations for the future prices of production (including feed) and pelt prices should influence the relative weight of different traits. This means that if low prices are expected, then relatively more weight should be on lowering costs (litter size, feed efficiency). On the contrary, if high prices are expected then more weight should be put on pelt traits (Berg, 1993c). Therefore, it is difficult to define breeding goals for mink. It also shows that uncertainty about the future should be an important variable in a national breeding scheme. A diversity in breeding goals among populations can ensure a genetic diversity among populations. This can be considered as an insurance, because the material to meet future needs and conditions is maintained, and uncertainty reduced (Smith, 1985). Einarsson (1988b) has discussed the principles for choice of breeding goals in fur animal breeding within populations.

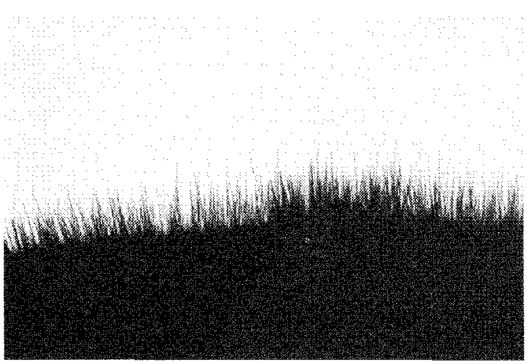
A considerable variation has been found between populations of individual farms, accounting for approximately 9% for litter size (Venge, 1961), approximately 20% to 30% for weight and skin length (Jónsson, 1971; Berg, 1993a) and approximately 10% for skin characteristics (Berg 1993b). A part of this difference is due to genetic differences between populations (Berg 1993 a and b), the rest being a result of factors like feeding, general management and pelting process. As shown by Berg (1993c) combined selection within and between populations as proposed by Smith & Banos, (1991) can increase the response to selection considerably. These investigations have shown a potential for improved breeding schemes by a more efficient utilization of genetic differences between populations.

As shown under selection theory, genetic progress is inversely proportional to generation interval. Rönningen et al. (1980) gave the methods to predict the optimal distribution of age groups, but they only presented results for a fixed distribution over ages. It has been found (Beumer & Berg, unpublished) that generation interval should be very short, especially for males. For selection in two stages genetic progress is maximised by including all available information in the index at each stage (Rönningen et al., 1980).

Crossbreeding has been used to produce scanbrown (e.g. Jørgensen & Lohi, 1984). However, a more systematic exploitation of the advantages of crossing has not been done. Heterosis was reported for price of pelts, skin length and fur defects (Evsikov et al., 1987). Lyngs (1987) found no heterosis for reproduction and skin length but for fur quality (9%-33%) and colour (0%-30%). Lagerkvist (1992) reported a positive heterosis for viability of crossbred kits, but effect of crossing was confounded with the age of the dam. No advantage of continuous crossing was observed for litter size and fur structure (Pingel et al., 1984), but a positive heterosis for litter size of a specific crossbred female. Reciprocal crossings should be tried, as differences between reciprocal crossings were



1. Good quality pelt with short guard hair



2. Uneven guard hair length

There are many details in fur quality

found (Pingel et al., 1984). The advantage of crossing does, however, not only rely on a positive heterosis effect. As shown by Moav (1966 a to c) there can be an advantage due to complementarity between lines, if the two sexes have a different contribution to the total profit. The full potential of crossbreeding in mink is not yet satisfactorily described.

Perspectives

The future work at the National Institute of Animal Science in Foulum (NIAS) will be directed to the following areas.

It has been found that genetic parameters for categorical traits are underestimated by linear models, compared to threshold models (Berg, 1993b). Genetic parameters of categorical traits will be further investigated in the future, with special attention given to fur defects, behavioural response and reproduction traits.

Both in Denmark and Finland, BLUP methods are developed for prediction of breeding values using an animal model. Algorithms have been presented for models with additive genetic effects, but it is possible also to include permanent environmental and maternal effects. This seems to be important for weight and skin size (e.g. Berg, 1993a). It is possible and conceptually simple to include dominance, epistatic and higher order genetic effects in the models, but with increasing numerical problems. Methods have been developed to predict breeding values for categorical traits (e.g. subjectively measured traits) (e.g. Gianola & Foulley, 1983), but these have not been given much attention in fur animal breeding. Development and implementation of methods for categorical traits are planned at NIAS, but numerical problems of obtaining point estimates for all animals under a threshold model remain to be solved. The experience obtained using threshold models to estimate genetic parameters (see above) is expected to give associated developments in this area.

The department is involved in a project 'Optimization of Breeding Plans for Cattle, Pigs, Poultry and Fur Animals' running from 1993 to 1997, in cooperation with scientists working with other species and at other institutions. The aim of this project is to estimate the expected effect of alternative breeding strategies utilizing new technical and biological methods, and to make suggestions for improved breeding schemes for all the animal species. The department is involved in three areas of this project.

The first area is definition of breeding goals. Here the aim is to investigate which economic/genetic principles should be used to define objectives and suggest relevant future breeding goals. Little work has been done in this area in mink breeding, and methods of incorporating uncertainty about the future might be especially relevant to fur animal breeding.

The second area is optimization of multistage selection and selection across groups of animals with the same amount of information. The aim will be to develop algorithms to optimize the effect of multi-stage selection and selection across groups. In mink special attention will be given to the possibility of using multistage selection to decrease the burden of live animal grading of all animals. An example could be a first stage selection based on reproduction (pedigree information) and weight, and a second stage selection further based on live animal grading for fur traits. Further, the effect of different distributions of age categories will be investigated. Little is known about the optimal distribution of breeding animals of different ages. Older animals will have their breeding values predicted with higher accuracy, but they will also increase generation interval, two opposite effects on genetic progress (see [1]).

The third area will investigate the impact of different breeding strategies on genetic variation and inbreeding. The aim is to develop algorithms to predict the effect of different breeding strategies on genetic variation and inbreeding. With more accurate prediction of breeding values (e.g. animal model evaluations), the selected animals will tend to be more related, and the degree of inbreeding will increase more than predicted under random mating and the genetic variance will decrease to a lower equilibrium (e.g. Verrier et al., 1991). These effects are important in order to be able to predict the effects of alternative breeding strategies, especially in small populations.

Table 1. Review of heritability estimates for some selected traits in mink.

Trait	N	Range of estimates 100%	50%	Mean ± SD	Median
Litter size[a]	15	0.07 - 0.30	0.11 - 0.24	0.16 ± 0.07	0,14
Body weight[b]	52	0.00 - 0.98	0.17 - 0.46	0.33 ± 0.21	0.29
Body size[c]	13	0.00 - 0.42	0.10 - 0.18	0.15 ± 0.11	0.14
Body length[d]	9	0.10 - 0.74	0.44 - 0.58	0.52 ± 0.17	0.52
Quality, live animals[e]	21	0.00 - 0.86	0.15 - 0.38	0.29 ± 0.24	0.21
Colour, live[f]	35	0.07 - 0.93	0.27 - 0.63	0.44 ± 0.21	0.45
Density, live[g]	14	0.10 - 0.34	0.12 - 0.25	0.19 ± 0.08	0.18
Skin length[h]	17	0.08 - 0.48	0.15 - 0.40	0.28 ± 0.14	0.36
Quality, skin[i]	13	0.00 - 0.55	0.10 - 0.37	0.23 ± 0.18	0.19
Colour, skin[i]	12	0.00 - 0.59	0.10 - 0.42	0.25 ± 0.18	0.26
Guard hair length[j]	18	0.00 - 0.80	0.27 - 0.64	0.43 ± 0.25	0.40
Wool hair length[j]	17	0.23 - 0.69	0.28 - 0.69	0.44 ± 0.17	0.44

a. Venge (1961); Johansson (1965); Pastirnac (1980); Einarsson (1981,1987); Tikkanen (1986).
b. Jónsson (1971); Benthin (1977); Reiten (1977); Rosberg & Olausson (1978); Jezewska & Maciejowski (1981); Børsting & Clausen (1986); Pingel et al. (1986); Lohi & Hansen (1989); Einarsson (1988a); Hansen et al. (1992); Berg (1993a).
c. Jónsson (1971); Olausson (1976); Olausson & Lohi (1978); Kettämies & Vilva (1988).
d. Reiten (1977); Lohi & Hansen (1989); Einarsson (1988a); Hansen et al. (1992).
e. Jónsson (1971); Benthin (1977), Rosberg & Olausson (1978); Børsting & Clausen (1986); Pingel et al. (1986); Kenttämies & Vilva (1988).
f. Jónsson (1971); Olausson (1976); Benthin (1977); Reiten (1977); Olausson & Lohi (1978); Rosberg & Olausson (1978); Børsting & Clausen (1986); Schumacher & Wenzel (1987); Kenttämies & Vilva (1988); Lagerkvist & Lundeheim (1990).
g. Jónsson (1971); Reiten (1977); Olausson & Lohi (1978); Rosberg & Olausson (1978); Lagerkvist & Lundeheim (1990).
h. Jónsson (1971); Benthin (1977); Reiten (1977); Lohi & Hansen (1989); Einarsson (1988a); Berg (1993a).
i. Jónsson (1971); Benthin (1977); Reiten (1977); Einarsson (1988a); Berg (1993b).
j. Reiten (1977); Olausson (1976); Maciejowski & Slawon (1980); Maciejowski et al. (1980); Einarsson (1988a); Berg & Lohi (1991).

Size, quality, and colour have been and will also in the future be very important factors for the pelt price. The increasing competition on the fur market will, however, emphasize the need for increased selection response and also the need for utilization of special fur properties, creating demand for furs and new marketing possibilities. This new research project will increase considerably the background knowledge needed for improving the effectiveness of existing breeding programs and give recommandations for increased selection response and for successful development of new products.

These research areas are also acknowledged as desired future projects (Lohi et al., 1992) in the strategic plan of the Fur Animal Committee for Breeding, Scandinavian Association of Agricultural Scientists.

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Litter Size and Growth of Mink – Possibilities for Improved Performance

Bente Krogh Hansen

Introduction

The overall breeding goal in fur animal production is to develop a herd with a high reproduction capacity and producing offspring with good size and fur quality. In mink production, both litter size and number of fertile females have gradually been improved over the last 20 years. Since 1971 the percentage of barren females in Denmark has been reduced from 18 to 9 percent, and the litter size per mated female has been increased from 4.27 to 5.68 (DP, 1992). Also the number of large litters, 8 kits or more, has increased.

In practice, however, it is not the number of kits born but the number of kits surviving that is important. Even though also the losses from birth to weaning have been reduced, we still lose an average of 0.5 kit per female before weaning. Therefore, the lactation period is a very important period in mink production. In addition to the surviving of newly born kits, the lactation period also builds up the foundation for growth and final body size.

The development of body size has in earlier research mainly been recorded from weaning to pelting and was used to document the effect of feeding (Jørgensen et al., 1961). Only a few reports regarding the early development can be found.

At Foulum we started in 1989 a project in order to focus especially on the lactation period and the variation between litters during this period. The aim is to find the causes for variation in the early development and to study the relation between early development and final body size, and in regard to female kits the relationship between the early development and their later reproduction and lactation capacity. The project includes investigation of the development of body weight and body length in mink kits from birth to pelting and selection for high respectively low growth from 2 to 4 weeks as an indirect expression of lactation capacity of females. Kit losses, female behaviour, and the level of some hormones in lactating females are also studied and related to kit growth.

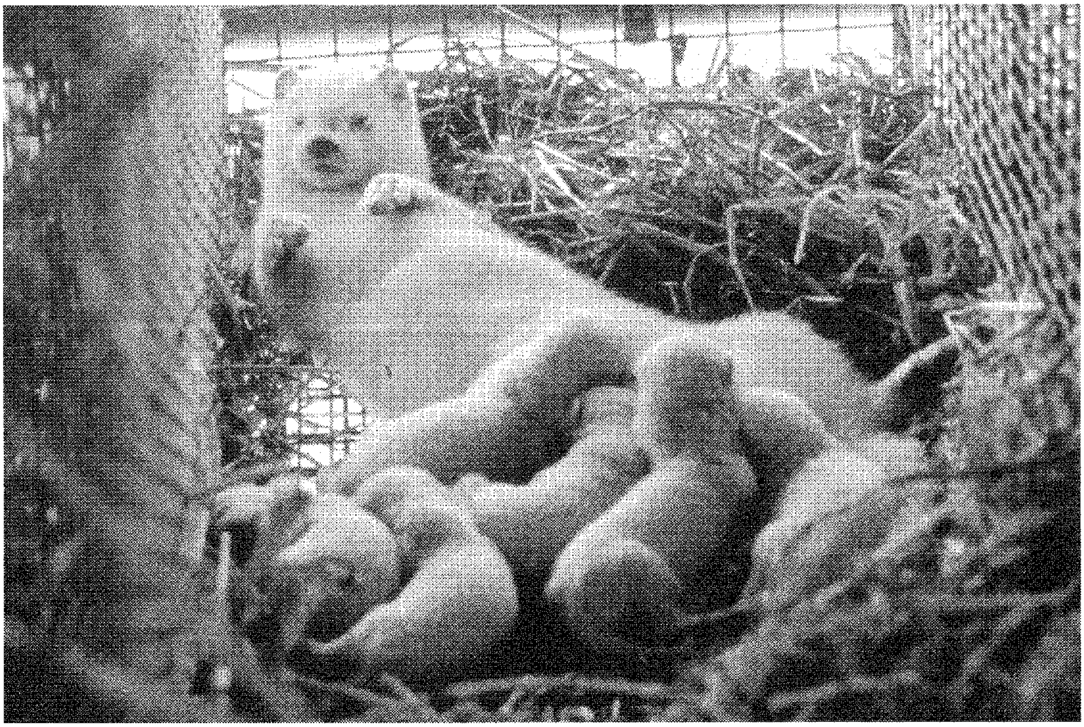
This paper presents a summary of results from other researchers regarding body growth in mink related to the first results from our own project. Further investigations are discussed under future perspectives.

Development of body size

The growth of mink kits can be divided into three periods. The suckling period, from birth to 4 weeks of age, during which period the kits are entirely dependent upon milk from the female, the gradual change from milk to mink feed, from 4 to 6 weeks or until weaning, and the postweaning period, when the kits are normally housed in pairs and fed solid feed.

Importance of the suckling period

Mink is one of the species where the newborns are physiologically fairly immature. They are naked and blind, and their ability to move around is limited. The energy stores in the newborn body are low, because only 1.4% of the body weight is fat (Tauson, 1993). It is therefore extremely important that the female is, immediately after delivery, capable of producing milk. The increasing litter size also requires an increase of lactation capacity in order to guarantee a good start for the kits even in very large litters.



The nursing period is of vital importance for the development of the kits.

Mink kits have an enormous capacity for growth. The average body weight at birth is about 11 and 10 grams for male and female kits, respectively, and is doubled in 3 days and by 6 weeks increased to 30 times the weight at birth (Hansen et al., 1992). There are, however, large variations between litters, and if the reasons could be found, it would increase our possibilities to improve the performance of both the female and her kits.

The daily weight gain is increased all through the nursing period from 4 grams up to 12 grams by weaning (Hansen et al. 1992; Møller, 1992). Møller (1992) described polynomial equations for the weight-age relationship from birth to weaning as follows:

Male kits:

$$bwgt = 10.1 + 0.95 \times age + 0.20 \times age^2$$
$$R^2 = 0.98$$

Female kits:

$$bwgt = 9.0 + 1.80 \times age + 0.14 \times age^2$$
$$R^2 = 0.99$$

where

bwgt = body weight in g.
age = age in days.

In earlier investigations it is shown that high body weight at weaning reflects high weight at pelting (Hoogerbrugge & Baud, 1975a). Likewise, good correlation between weight at weaning and skin size has been reported (Lund, 1980). Nielsen & Olsen (1970) found that if weight gain from 2 to 6 weeks is increased, skin length is increased correspondingly. Tauson (1985) found a relation between feed consumption during early growth and later body and skin length.

For practical reasons, the weight at weaning is most often used. However, the latest investigations show that equally high correlations to skin length ($r = 0.40$) are found at 4 and 6 weeks of age. Fig. 1 and 2 are based on the male kits from material described by Hansen et al., 1992. Fig. 1 shows that body weights at 4 and 6 weeks

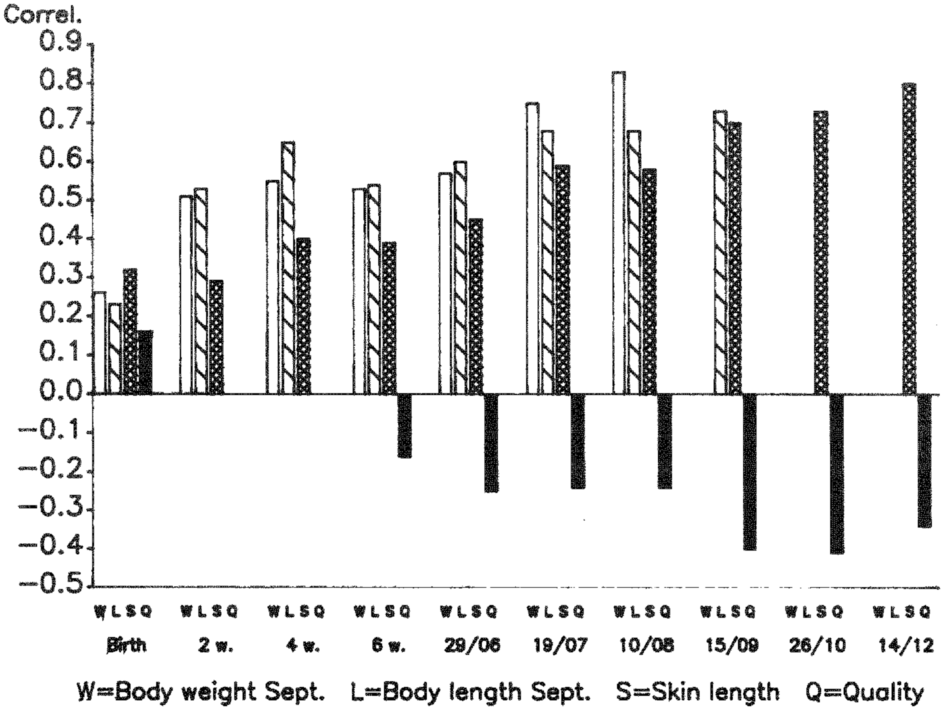


Figure 1. The correlation of body weight at different ages with body weight and length in September and with skin size and quality.

are both good estimates for later body weight and skin length, whereas body weight at 4 weeks is the best estimate for the later body length. For female kits the size at 4 weeks is the best estimate for later body size and skin length (Hansen et al., 1992). Correlation between body length at 4 weeks and skin size is 0.47 and 0.50 for males and females, respectively. Of the figures from the nursing period, the size at 4 weeks thus seems to be the most important.

Factors influencing the growth from birth to 6 weeks of age

Dutch researchers have found a significant difference in male and female body weights already at birth. (Hoogerbrugge & Baud, 1975a; Baud & Hoogerbrugge, 1977). The preliminary results of our project with scanblack mink confirm this in regard to both body weight and body length at birth (Hansen et al., 1992).

The same was found in an investigation with scanbrown colour type in Finland. Already at birth, the males were significantly heavier than the females. The difference persists and becomes more distinct through the entire preweaning period. The Finnish results also showed that the kits, which were bigger at birth were also bigger at the age of 6 weeks (Korhonen et al., 1991). This corresponds well with our results on scanblack mink (Hansen et al., 1992).

Another important factor is litter size. It is possible that the female is not capable of increasing the milk yield in proportion to the number of kits (Hoogerbrugge & Baud, 1975a).

Udris (1970) and Einarsson (1980) have documented a reduction of 0.10–0.24 grams in birth weight per each additional kit in the litter. In our material (Hansen et al., 1992) a similar difference of 0.2 grams in the average birth weight was found between the different litter size groups, and the average weight of the smallest kit gets even smaller when the litter size increases (Schneider & Hunter, 1990). The effect

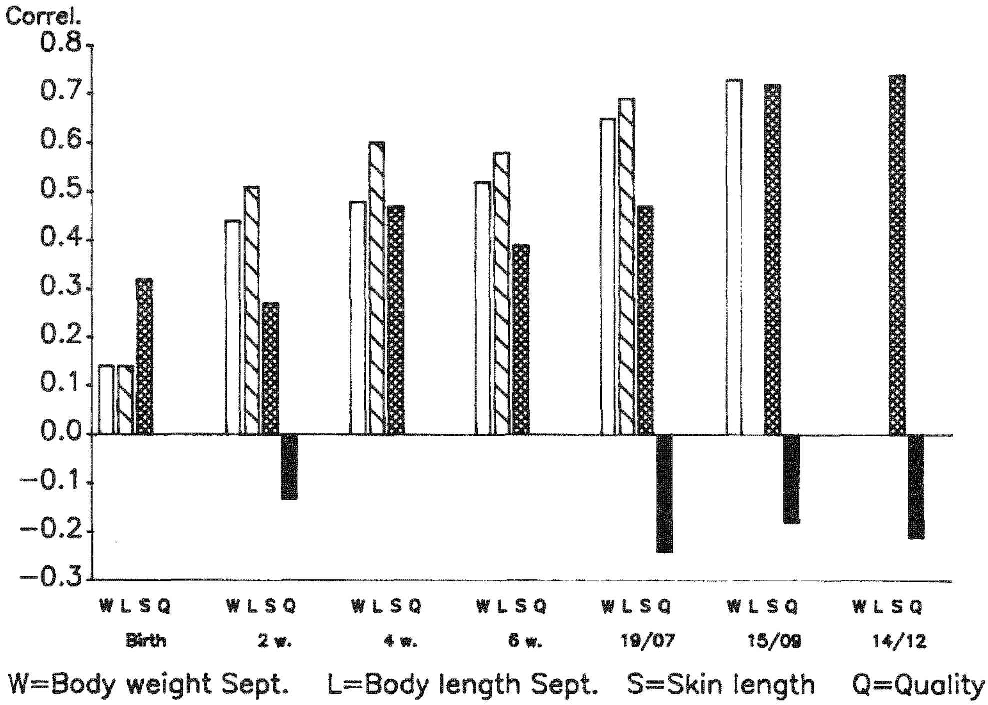


Figure 2. The correlation of body length at different ages with body weight and body length in September and with skin size and quality.

of litter size is obvious through the nursing period but continues to exist even after weaning.

An interesting point in our future studies is the relation between the changes in female condition and the development of the kits. The loss of body weight in females is an indirect indicator of her ability to mobilize energy from her own body reserves. Therkildsen (1989) found that a heavy loss of weight in female was related to better growth in kits. In addition to that there seems to be an interaction between the loss of weight and the age of the female. Hansen (1991) found that second year females lose more weight than the yearlings. Further investigations are necessary to study whether this interaction is due to higher milk yield in older females.

Several studies have also been conducted to illustrate the importance of number of teats and whether it is related to the litter size (Elofson & Swensson, 1982). Pedersen (1978) found that all teats are not always active. He also found a difference between young and old females. There can be more kits in the litter than there are active teats (Hoogerbrugge & Baud, 1975b). Einarsson (1987) suggested that females with few teats should be removed from breeding.

Postweaning period

After weaning, the increase of body weight in both males and females follows an S-formed curve. The early growth is rapid in both sexes, but the sharp increase is slowed down in males in September and in females a little earlier, late in August. The maximum weight, males about 2100 grams and females about 1200 grams, is reached in October (Einarsson, 1987 and 1988; Hansen et al., 1992). As a result of both selection and better feeding, the average maximum weight of both males and females has increased about 200 grams during the last 20 years (Jørgensen et al., 1961; Reiten, 1978). This can also be seen in the increase of skin size at the fur auctions.

The development of body frame is even faster and a plateau is reached already at the beginning of August. The body length of mink is fully developed early in September (Lohi & Hansen, 1989; Møller, 1992) and remains unchanged during the rest of its life (Reiten, 1978).

Altogether, the early growth until August is characterized by a fast increase in proteins and minerals, and the later growth period almost entirely by increase in body fat (Charlet-Lery, 1980; Hansen et al., 1981). A lot depends on the feeding practice, but on average the relation body weight/body length is increased from August (Hansen & Lohi, 1990). French researchers have recorded an increase in body fat content from 15 to 30 percent from August until November (Charlet-Lery, 1980).

Postweaning growth and skin size

Body size is also of great importance when the goal is to produce large pelts. In the present pelting system, standard size drying boards are used, and as the fresh skin is stretched on the board, the length of the pelt is thus a joint result of both body length and the fatness of the animal. Møller et al. (1991) found the following equation for the relationship between skin length and body size parameters:

Skin length (cm) = 26.4 + 0.63×body length (cm) + 8.8×body weight (kg).

Already from September the correlation between body weight and skin size is high ($r \geq 0.70$) (Lohi & Hansen, 1989; Møller et al., 1991; Børsting & Therkildsen, 1992). The highest correlation between body weight and skin size is found at pelting ($r \geq 0.78$) (Hansen et al., 1992; Lohi & Hansen, 1989; Olesen, 1989).

The disadvantage of using body weight in selecting breeders is that in males there is a negative correlation between body weight and fur quality ($r = -0.16$ to -0.40) (Reiten, 1977 a and b; Møller, 1988 and 1991, Olesen, 1989; Hansen & Lohi, 1990). This negative correlation is found already in June but is strengthend towards pelting (Olesen, 1989; Møller et al., 1991; Hansen et al., 1992).

Positive correlation between body length and skin length is earlier documented by Rimeslåtten (1961). In our investigations body length in September and at pelting have almost equally high correlations ($r = 0.72$ and $r = 0.74$) to skin length as body weights at corresponding times (Hansen et al., 1992). On the other hand the negative correlation to fur quality is weaker ($r = -0.18$ to -0.21) (Olesen, 1989; Hansen & Lohi, 1990; Hansen et al. 1992). According to Møller (1991) the relation between body size and fur quality can be based on the following equation:

$$\text{Quality} = 7.2 + 0.35\times\text{length} - 6.41\times\text{weight}$$

where

length = body length in cm.
weight = body weight in kg.

However, body length is not as easy to measure accurately as body weight.

The degree of fatness (g body weight/cm body length) is another important factor reflecting the skin length. Hansen et al. (1992) found at pelting a correlation of 0.83 to skin size but –0.43 to fur quality. This negative correlation is in males found already at the age of 6 weeks. The quality of female pelts is reduced only if the weight/length relation is very high in December (Hansen et al., 1992).

Factors affecting the final body size

It is important to know the normal development of size in order to be able to detect subnormal conditions and to describe reasons for them. Jørgensen et al. (1961) found that early born kits (before May the 2nd) were heavier at pelting and therefore recommended selection of breeders from the early litters. As the effect of litter size was not included in this investigation, there is a possibility that the difference is at least partly due to differences in litter size. Later investigations have shown that the date of birth has an effect until the middle of August, but the late born kits catch up with the early born ones in body length by early September and in body weight by October (Lohi & Hansen, 1989).

Just as the litter size has an effect on the average weight of kits in the preweaning period, the number of kits in the cage has an effect on the later growth, and best growth has been achieved by placing the kits in pairs, male and female, after weaning (Jørgensen et al., 1961), as also used in practice.

In earlier investigations a reduction of the weight of male kits due to litter size was still found at pelting. Each additional kit reduced the average weight by 20 grams (Einarsson, 1980). Lohi & Hansen (1989) and Møller et al. (1991) found a significant effect of litter size until the 15th of August. At pelting, no effect could be seen in female weights. The male weights were slightly reduced, but the difference was not statistically significant. The present results indicate that the medium size litters provide the best growing conditions both in regard to body weight and body length (Lohi & Hansen, 1989). Whether the large variation between litters is of genetic origin and can by selection be used to change this condition, will be one of the main questions in the ongoing project.

Perspectives

It is obvious that a great deal of the improvement in reproduction of mink during the last decade is due to better feeding and health control, but part of it is a result of selection for good reproduction. About 9 percent of females produce litters with 8 kits or more. Therefore it seems obvious that physiologically there are possibilities to increase the average litter size. However, it is important to be able to increase the female's lactation capacity at the same time without an increased loss of females during the nursing period.

In the research project, being conducted at Foulum at present, we focus especially on the lactation period. The aim is to develop methods for selecting females capable of producing and rearing large litters.

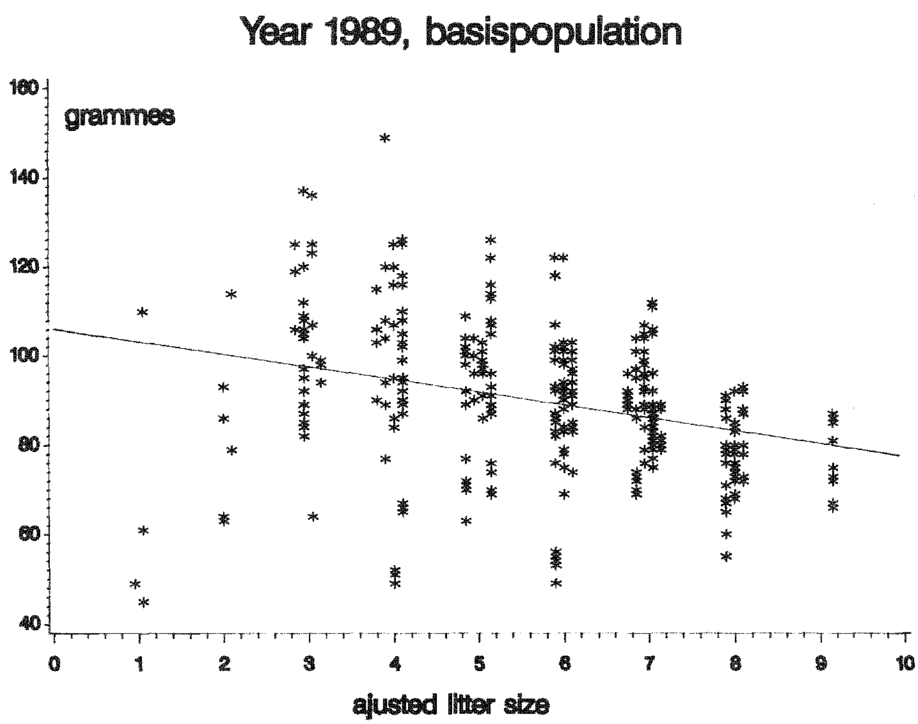


Figure 3. Weight gain of kits from 2 to 4 weeks in the basis population in 1989.

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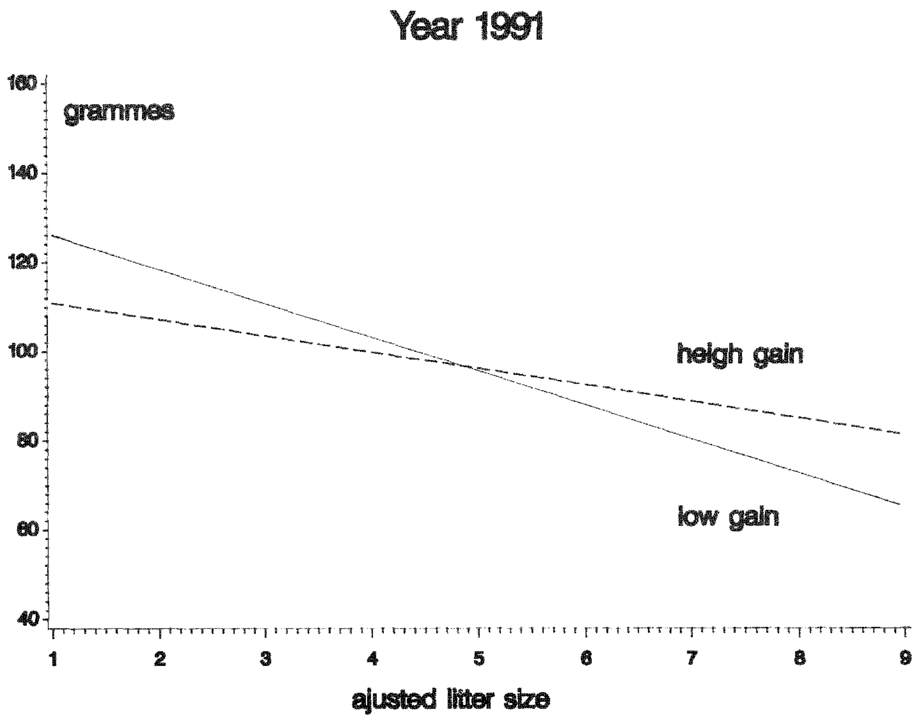

Figure 4. The average weight gain from 2 to 4 weeks in selection lines in 1991 in relation to sex adjusted litter size.

A divergent selection is practised in two lines selecting for high respectively low weight gain from 2 to 4 weeks – the period when the kits are still depending entirely on the milk production of the female. The weight gain during this period is used as an indirect expression of the female's lactation capacity.

In Fig. 3 is shown the average weight gain from 2 to 4 weeks in the basis population in 1989. Fig. 4 shows that the two selection lines differ already after 2 generations.

In addition to the growth of the kits several other parameters are studied in this project.

The weight loss of the female related to her feed consumption will be used to evaluate the lactation capacity and her ability to utilize the energy resources. At the same time plasma is collected to form a picture of the level of the hormones insulin, glucagon, T_3 and T_4 during the lactation period. With these parameters we hope to be able to increase our knowledge about important factors in avoiding health disturbances in the female during the nursing period.

The female's reactions during the weighing procedure and to disturbing sounds as well as the condition of the nest are used to evaluate other mothering abilities.

One of the aspects in the present project is to get more information about the possible reasons for early loss of kits which, during the nursing period, amounts to 17 percent (Therkildsen, 1989; Hansen et al., 1992).

The total material will provide possibilities of evaluating the effectiveness of the present selection method and of estimating the heritability of early growth parameters.

At the end of the project we hope to be able to evaluate the possibilities for improving the lactation capacity and mothering abilities of mink females, including weight gain of the kits, and to evaluate whether it is desirable to include selection for such parameters in the breeding programmes.

Finally the effort is made to suggest an optimal selection method.

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Cytogenetics and Molecular Biology in Fur Animal Research

Outi Lohi

Introduction

Special techniques like cytogenetics and other biotechnological investigations have during recent years made great progress in many farm animal species. Blood groups and electrophoretic identification of proteins are as a routine used in e.g. cattle, pigs and horses for identification of individuals, parent tests, and also for detection of genetic characteristics.

In fur animals, biological markers are less investigated. There are, however, several problems in fur animals, where biological markers for genetic characteristics would be desirable. Animals being heterozygous for recessive genes for diseases or fur defects cannot be detected by the phenotype. Biological markers of genes for coat colour would start a new epoch in research for inheritance of coat colours.

The Department for Research in Fur Animals at NIAS does not have its own specialists or laboratory facilities for biotechnological methods but has, as initiator and cooperator, been involved in several projects in this field. The work itself has been carried out in cooperation with other departments of our own institute as well as other Danish and foreign institutes. Especially in regard to gene mapping, Gunnar Jørgensen's international activities have initiated interesting contacts to the institute from as far as Novosibirsk in Russia.

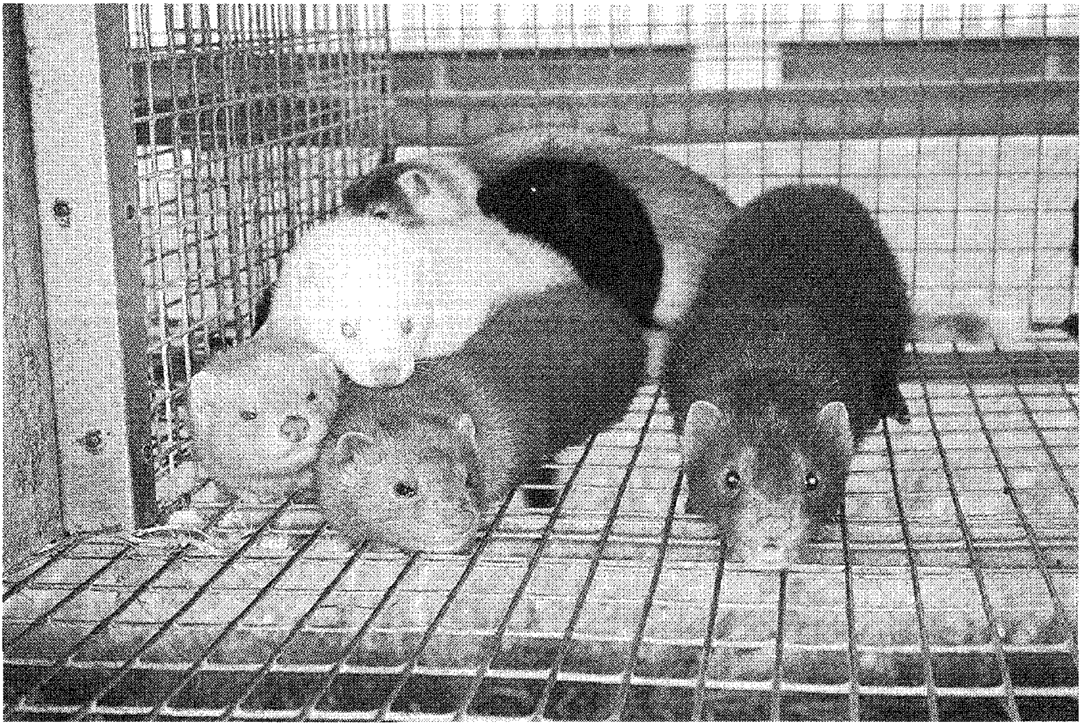
Cytogenetics

Quite a lot of cytogenetic studies on fur animals have been carried out purely from a phylogenetic point of view, to study the relationship between different *Mustelidae* and *Canidae* species. In some connections, however, knowledge about chromosomes has had interest also in fur animal research. In cytogenetic studies the Department for Fur Animals at NIAS has worked together with other Scandinavian research institutes and the Royal Veterinary and Agricultural University of Copenhagen.

In relation to the increasing production of inter species hybrids between the two fox species *Vulpes vulpes* and *Alopex lagopus* in the 1970's, the chromosomal map of both species and their hybrids was studied in Finland (Mäkinen & Lohi, 1976). The number of chromosomes in silver fox, *Vulpes vulpes,* and its colour phases is constant $2n = 34$, not counting the micro-chromosomes. Blue fox, *Alopex lagopus,* showed variation from $2n = 48$ to $2n = 50$ in the chromosome number due to Robertsonian translocation (centric fusion of two acrocentric chromosomes). The inter species hybrids are intermediary to the parents and thus have $2n = 41$ or 42 depending on the chromosome number of the blue fox parent (Mäkinen & Gustavsson, 1982).

Some investigations in cattle have shown that lowered fertility is related to this kind of chromosomal variation (Swenson, 1973). This led to investigations on blue foxes both in Finland, Denmark, and Norway. In the Finnish material, the stable chromosome compositions $2n = 48$ and $2n = 50$ had significantly better litter average than the group where both male and female had $2n = 49$ (Mäkinen & Lohi, 1987). This was also the case in the group $2n = 50$ in the Danish material (Christiansen et al., 1986). The slightly lower litter average in the group with $2n = 48$ in this investigation can depend on the small number of animals in this group.

In investigations of fox karyotypes also special chromosome mutations, existence of microchromosomes and heterochromatin variations



DNA microsatellites will be studied on mink.

were found in blue foxes (Mäkinen et al., 1981). The blue fox seems to be fairly tolerant to heterochromatic imbalance. The possible genetic effects of such imbalance need to be further investigated.

In silver fox, microchromosomes were common and varied in numbers between individuals and between cells of the same animal (Kuokkanen et al., 1985). Most frequent numbers were 2 and 3 and almost linear correlation in the number between parents and offspring was found. No correlation to reproduction or other production traits could be detected and the genetic role of microchromosomes is still unknown.

Blood types

In many domesticated animal species, blood typing is commonly used for control of parents and as gene markers detecting genetic diseases. In fur animals, this field has not been studied very much. However, some studies on blood types in mink were carried out in the U.S.A. in the 1960's by Rapacz and Schackelford (1966). Seven blood factors were detected, controlled by four genes in different loci.

In 1987, NIAS was granted financial support from the Nordic Joint Committee for Agricultural Research for an inter Nordic project on "Research in blood types, tissue types and protein polymorphism in foxes". The project was carried out in cooperation between the Department for Animal Physiology and Biochemistry and the Department for Research in Fur Animals, NIAS Denmark and the Blood Type Laboratory of the Central Association of A.I. Societes in Finland.

The investigation included both fox species, and immunizations were made both within and between species (Christiansen et al., 1989). In general, the antibody response was unexpectedly low (Larsen et al., unpublished). Altogether 11 specificities were detected. Most antisera were produced from arctic foxes, only one anti-

Fur animal gene bank at Foulum includes also different colour types af foxes.

serum by both species and two by silver foxes. Antigens a, c and d were found to be closely related and not independent from antigen j. Another close group was anti f, g and i. The 9 antisera of arctic foxes are thus controlled by five genetic systems. The inheritance seemed to be dominant in all cases. No sex linkage or total linkage between blood group systems was detected. Larger family material is needed to study possible moderate linkages.

Protein and enzyme polymorphism

Polymorphism of protein and enzyme systems in foxes were studied from 1987 to 1992 as part of the inter Nordic project mentioned above. Samples were collected from 8 Danish and 10 Finnish farms from 725 arctic and 698 silver foxes and represent most existing colour types of *Vulpes vulpes* and *Alopex lagopus*.

Three new plasma proteins and two enzymes, which are earlier not reported polymorphic, showed variation in this material (Juneja et al., 1988 and 1989; Simonsen et al., 1990 and 1991).

Altogether sixteen different loci were studied, 11 of these were polymorphic in arctic foxes and 8 in silver foxes (Niini et al., 1992). Differences between farms were detected in some systems, but they can in this material be due to sampling of special families and colour types. The largest variation in arctic foxes was found in α_1B-glycoprotein, transferrin, postalbumin 1 and the enzyme adenosine deaminase. There was generally less variation in silver foxes, most polymorphic were systems postalbumin 1 and pretransferrin 1.

No immediate linkage was detected between the blood types and the protein/enzyme systems (Larsen et al., unpublished). Some relationship to colour type genes has been detected. Further analysis of the results is presently being conducted.

In earlier Russian studies with mink *(Mustela vison)* esterases have expressed variation. Serov (1990) has reported polymorphism in esterases found in heart and red blood cells in three different loci, named ES1, ES2 and ES3. In our material of 412 mink representing 20 families, 7 allelic forms of plasma esterase were found. The interesting result was that the C-allel seemed to be present only in colour types including the royal pastel gene (Simonsen et al., 1992).

In 1992, studies on mink continued and 49 additional proteins were studied. Two new proteins were found polymorphic. C-allel of plasma esterase was again found in royal pastel type (Brusgaard et al., unpublished). However, further studies with material from several different farms and/or countries are needed to confirm connection to the colour gene.

Perspectives

The primary purpose of molecular genetic studies with fur animals has been to find biological markers for single genes or for characterization of pure lines or families.

As variation in blood types and protein and enzyme systems has shown to be limited, other biological markers have to be included in future studies.

DNA-microsatellites exhibit large variation in many species and will therefore be the subject of the new fur animal project commenced in cooperation with the Department for Animal Physiology and Biochemistry at NIAS and The Royal Veterinary and Agricultural University of Copenhagen. In this project we try to identify a series of microsatellites which show variation. These will be analysed in test families including different colour genes from the sire and dam side. Matings in subsequent generations are planned in order to investigate the inheritance of microsatellites and their possible relation to genes for coat colour or for other production traits. Electrophoretic studies to detect protein and enzyme polymorphism are carried out on the same material in order to investigate relations between microsatellites and protein polymorphism. Furthermore, the possibility of using microsatellites for identification of pure lines will be investigated. A method for controlling the purity of breeding lines is necessary if line breeding will be used as a more effective breeding method in the future.

The identification of colour genes with the aid of microsatellites will open new possibilities to solve some very much discussed problems in colour type inheritance and to study relationship of colour genes between species.

Furthermore, if relations between microsatellites and recessive fur defects or diseases can be detected, the analysis will make eradication of the heterozygous carrier individuals from breeding possible.

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Clinical Analysis

Birthe M. Damgaard

Introduction

In the first years of the department's existence, nutrition was the major research area. Gunnar Jørgensen realized, however, very soon that it was not enough to carry out production experiments. It was also necessary to study the basic metabolic relations. He therefore took the initiative to collect blood and organ samples from the animals for laboratory analyses. The purpose was to illustrate the basic metabolism of nutrients in order to be able to feed the animals optimally and select animals which were robust and vigorous under the production conditions given. In the first years, the analyses were performed at laboratories outside the department. The increasing demand led, however, in 1982-83 to the establishment of a hematological and clinical-chemical laboratory with one employee to handle the clinical analyses. To begin with, the laboratory was a 'kitchen table laboratory', but very soon so many analyses were set up that it became necessary to get more space – but still under quite primitive conditions. When the department was moved to Foulum in 1989, new and modern laboratories were established.

Hematological and clinical-chemical analyses

A number of investigations have been carried out to illustrate the normal variation in hematological and clinical-chemical analyses of mink. In one of the earliest investigations, the correlation between the content of hemoglobin in the blood and fur quality was examined (Jørgensen & Christensen, 1966). A later investigation showed that there were sexual differences for a number of hematological variables in mink (Rothenberg & Jørgensen, 1971).

In the department's work to optimize the supplementation of vitamins and minerals to the feed, hematological variables were used to illustrate the effect of supplementation on the animals. Addition of vitamin-C to conventional feed for lactating females and their kits had no effect on hematological variables in the females or on their loss of weight, just as there was no effect on the hematological values and weight of the kits at weaning (Brandt, 1983). A later investigation with mink kits in the growth period showed that supplementation of vitamin-C and iron sulphate had an effect on growth, whereas EDTA-iron did not have any effect (Brandt, 1987).

In further studies concerning supplementation of iron, the feed was added approx. only 50% of the amount of iron normally added to feed at feed kitchens. In all experimental groups, the content of erythrocytes, leucocytes, thrombocytes, and hemoglobin as well as the hematocrit value seemed to be reduced (Brandt & Mejborn, 1986 and 1987).

Feed containing herring offal treated in different ways, which resulted in fatty acid oxidation of various degrees, could in hematological and enzymological examinations not be proved to affect the plasma content of alpha-tocopherol and the cell membrane integrity measured indirectly via enzymes. Growth and fur properties were not affected, either (Brandt & Hillemann, 1987). The effect on the animals of feeding sulphuric acid preserved fish silage in the lactation and growth periods was studied in clinical-chemical and pathological examinations and production trials (Poulsen & Jørgensen, 1986).

Blood sampling of mink requires fixation of the animals or anaesthesia which influences the physiological variables to different extents and in different directions. At the department a project for clinical examination of the effect of various sedatives and anaesthetics was performed (Jepsen et al., 1981). The main conclusion was that most sedatives and anaesthetics affect the respiration and blood circulation of the animals and thus also the hematological and clinical-chemical variables. When evaluating hematological and clinical-chemical values in samples collected with the use of sedatives and anaesthetics, the effect of the substances in question on the measured variables must be taken into account. For total anaesthesia it was found that the use of Althesin® was the optimal, as this substance has the least effect on the hematological and clinical-chemical variables. A combination of Combelen® and Ketalar® and pentobarbital sodium, respectively, for anaesthesia was also satisfactory.

In connection with progeny testing at Favrholm in 1983 and 1984, a comprehensive project was carried out with collection of blood samples and analysis of these for hematological and clinical-chemical variables. This work was continued in genetically well-defined strains in the following years until 1988. The preliminary results showed that when a population is hit by the nutritional muscular degeneration syndrome (NMDS), changes take place in the blood variables also in animals which do not show clinical signs of the disease. Furthermore, a certain genetic background for resistance or the capability of resisting illness and nutritional disorders was found (Lohi et al., 1986).

NMDS is often seen among fast growing mink kits. To find a screening parameter for the syndrome, the frequency of histopathological myocardial degeneration (MCD) was correlated to the total activity of creatine-kinase (CK) in blood plasma. It was found that the frequency of MCD was significantly higher in animals with CK-activities above 200 i.u./l than in animals with CK-activities below 200 i.u./l (Brandt & Henriksen, 1986).

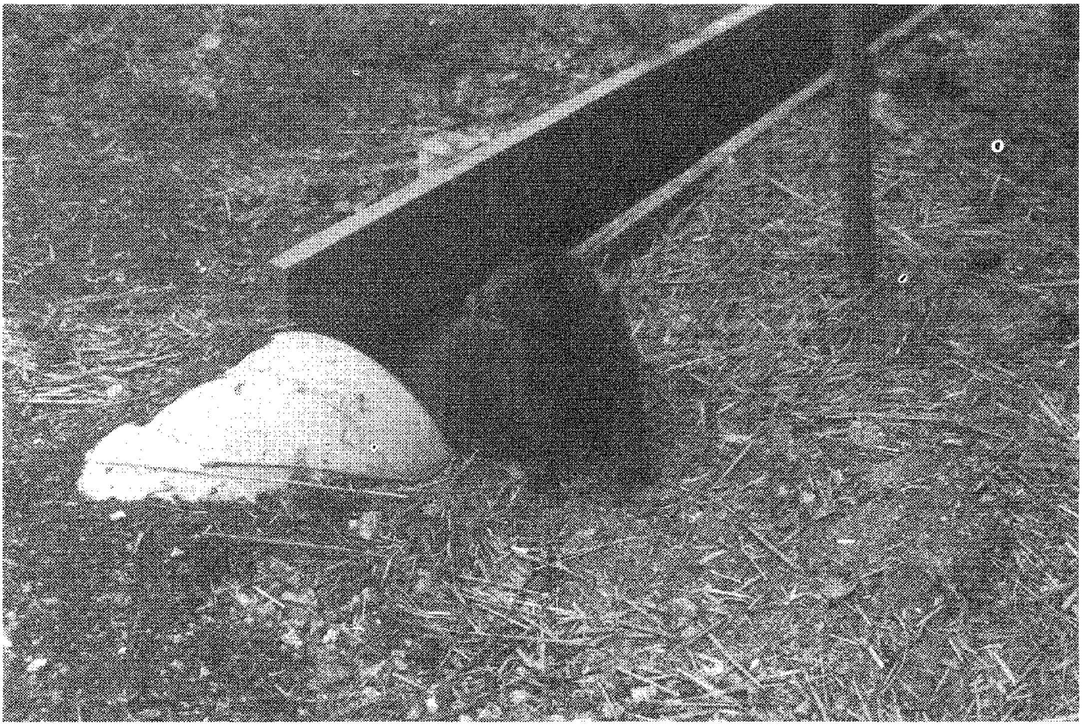
When evaluating hematological and clinical-chemical variables studied in connection with health surveillance and production control, it is important to be able to compare the data collected with reference values from animals of the same age, sex and colour type. This led to the edition of the first reference book in fur animal hematology and clinical chemistry. The book was created in cooperation between the USSR Academy of Sciences, Karelian Branch, Petrozavodsk, the College of Veterinary Medicine, Helsinki, the Finnish Fur Breeders Association, and the National Institute of Animal Science, Department for Research in Fur Animals (Brandt, 1989). The book concerns the collection of blood samples and reference values of hematological variables and clinical-chemical variables comprising proteins, carbohydrates, lipids, enzymes, hormones, vitamins, and minerals. Furthermore, special diseases and organ function tests are described. The stability of the enzymes ASAT, ALAT and CK as well as urea and creatinine in plasma depending on time of storage at –20°C was examined later (Damgaard, 1992). Samples for analysis of CK should not be frozen.

In connection with the project: Adaptation and Domestication of Beech Marten, reference values have been found for hematological and clinical-chemical variables in beech marten (Damgaard & Hansen, 1992). The number of erythrocytes, the hemoglobin content, and the hematocrit value were higher in males than in females. The erythrocytes in beech marten were smaller in size and volume than the erythrocytes in mink.

In relation to the project: Production Factors and Production Control, electrophoretic methods were initiated for illustration of the immune status of mink and foxes. The immune status is illustrated by semi-quantitative determination of the immunoglobulins IgA, IgG, and IgM by means of rocket immunoelectrophoresis. The amount of immunoglobulins in milk, in plasma of females and kits in the nursing period, and in plasma of kits in the growth period from weaning until pelting has been examined.

The content of IgA and IgG in colostrum and milk is important to the immunological protection of the kits in the lactation period. IgM is not present in colostrum and milk, but is produced by the kits' own immune system (Brandt, 1988 a and b).

When examining mink kits from 7 different strains representing 4 colour types in the period from weaning until pelting, it was found that differences in the amount of immunoglobulins are to a higher extent related to strains than to colour types. It was furthermore found that the divergence was highest for IgA, lower for IgG and lowest for IgM (Damgaard, 1990; Damgaard & Lohi, 1990).



The behaviour of mink is also related to clinical-chemical parameters.

An examination of the content of esterase in the blood of the same animals revealed a divergence between strains due to a founder effect and/or selection (Simonsen et al., 1990 and 1992).

In connection with the establishment of a behavioural section at the department, the laboratory initiated stress physiological analyses including eosinophil leucocyte counts, H/L-ratio, and cortisol. In several investigations, the stress physiological variables are supplemented with other hematological and clinical-chemical variables. An examination of the effect of cage size on the hematological variables showed that the concentration of hemoglobin was higher in animals in large cages than in animals in small cages, indicating that mink in large cages were in better physical condition than mink in small cages (Hansen & Brandt, 1989). Later investigations could not demonstrate differences in hematological variables as regards sex and whether the animals had been placed singly or in groups (Hansen & Damgaard, 1991). On the contrary, the activity and the individual variation for the enzymes ASAT, ALAT, and CK were lower in mink, which had most of the time been placed alone, than in mink which had either permanently or most of the time been placed in groups.

In a project selecting the animals according to behavioural criteria, three consecutive generations were examined to illustrate whether selection has had an effect on the hematological and clinical-chemical variables. It has not been possible to prove any divergence in the physiological variables chosen between the three groups selected for different behaviour (Damgaard, 1991).

Challenge test

In connection with the metabolic studies on nutrient metabolism we have during the last year worked with a metabolic challenge test for glucose and a few amino acids as described in the accompanying paper by Børsting & Glem-Hansen.

The physiological examinations in relation to behaviour and stress have shown that there are large variations in the stress physiological variables between individuals, which is important to the adaptability and production qualities of the animals. The activity in the hypothalamus-hypophysis-adrenocortical axis can be described by means of adrenal cortex function tests, dexamethasone suppression and ACTH stimulation tests, which were started at the department in 1988 on mink.

The adrenal capacity of female mink after 7 days of daily immobilization for 60 minutes, was compared to the capacity of females, which had permanently been living in cages without nest boxes, and compared to the capacity of females in conventional production cages. The results showed that females which had been immobilized reacted with a lower increase in plasma cortisol at ACTH stimulation than females which had been exposed to a constant environmental stressor such as nest box deprivation (Hansen & Brandt, 1989). The response in mink kept in conventional production cages was intermediate.

The reduced increase in cortisol excretion following immobilization must be due to a reduced sensitivity to ACTH or to reduced adrenal capacity. Contrary, stress caused by nest box deprivation (long-lasting stress) seems to increase sensitivity to ACTH stimulation.

The dexamethasone suppression/ACTH stimulation test was later performed on beech marten males caught in the wild. The test was carried out on the same males for three consecutive years. The results have shown that the test can be performed with high repeatability on beech marten. Differences in the baseline level for cortisol between years could be related to the way of catching and fixating the animals at blood sampling and to whether they were anaesthetized before the first blood sample was taken (Damgaard & Hansen, 1993).

Perspectives

The work with metabolic challenge studies will be continued to elucidate metabolism of glucose and amino acids. The results already obtained have shown that it is hereby possible to illustrate the metabolism of single components as well as the hormonal regulation of the metabolism. Challenge studies with hormones or metabolites are being introduced to find a measure for the capacity of mink females for milk production which is of importance to the weight development of the kits and later to skin length at pelting.

The large variation shown in the stress physiological variables between individuals makes it difficult to interpret results based on average values for a group of animals. The reason for this variation can either be individual differences in the capacity of the adrenal cortex for secretion of stress hormones, or be an expression of differences in stress sensitivity at the central nervous system and hypophysis level. Our previous results have proven a correlation between the individual activity pattern and the individual physiological stress reactivity.

Examinations on pigs have shown that growth rate, rate of metabolism and reproduction results were higher in individuals with a low cortisol reactivity than in individuals with a high reactivity at ACTH stimulation of the adrenal cortex. It is therefore the intention in future studies to group mink with regard to low/high reactivity in the adrenal cortex to examine if superior production results are an expression of better adaptation to the production environment.

Russian investigations showed that selection for behaviour resulted in pronounced changes in the level of sexual hormones and of corticosteroids, which must be supposed to be of importance to the reproduction properties. By means of hematological and clinical-chemical examinations it has not been possible to demonstrate any differences between three groups of mink selected for temperament. On the basis of the Russian results we want to characterize the three behavioural groups according to cortisol reactivity at ACTH stimulation in order to examine physiological differences between the three behavioural groups.

The number of barren females varies on a national basis between 10 and 20% of mated females. Some American examinations have revealed hormonal reasons why a number of females are not mated.

There is a correlation between the production of stress hormones and sexual hormones, as the regulation of these hormones affect each other mutually. With an ACTH challenge test we will try to characterize barren/pregnant females and females with different activity patterns to try to describe the adaptability and reproduction properties of the animals.

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Feedstuff Evaluation, Nutrition, and Nutritional Physiology

Christian F. Børsting & N. Glem-Hansen

Introduction

This paper reviews the progress in fur animal nutrition conducted at the Dept. for Research in Fur Animals during the 35 years Gunnar Jørgensen has been involved in research within the area. Focus will be on the present status and the perspectives of future research within mink nutrition.

For many years nutrition was the predominant area of research at the department, and, therefore, a lot of research in nutrition has been performed. Feedstuff evaluation has been a very important area yielding much necessary information to the fur industry, and over the years research with most nutrients including protein, fat, carbohydrates, minerals, and vitamins has been performed. Lately, also metabolic studies of the intermediate metabolism of nutrients have been included in the research programme as an important tool to make further progress.

Feedstuff evaluation

As for other species of domestic animals, also for fur animals feedstuff evaluation was one of the first major areas of research.

Over the years, many digestibility trials have been conducted at the department concerning digestibility of both protein, fat, and carbohydrate sources (e.g. Glem-Hansen & Jørgensen, 1972b). A lot of that information was gathered in a Scientifur paper (Glem-Hansen & Jørgensen, 1978), where chemical composition, amino acid composition, mineral content, and digestibility of protein, fat, carbohydrates, and amino acids were given for a large number of feedstuffs of importance to the fur industry.

Later, the list of feedstuffs was broadened further (Glem-Hansen, 1979b) through experiments mainly with dried protein sources of both vegetable and animal origin. Much of the information in these two papers was later included in the Scandinavian feed table for mink (Glem-Hansen et al., 1985) as well as results from the continuous number of digestibility trials performed (e.g. Mejborn, 1983 a to d).

Recently Børsting (1992) published data from current digestibility trials with new and traditional feedstuffs of the three categories: fresh animal feedstuffs, dehydrated animal feedstuffs, and vegetable feedstuffs. The digestibility coefficients of most of the traditional feedstuffs were not changed compared to previous studies and table values. However, for some process-dependent feedstuffs, the new data did imply that existing table values should be changed due to differences over time in manufacturing processes.

Until now, Danish mink feed evaluation has been based on table values for chemical composition and digestibility of individual feedstuffs rather than parameters actually measured in the individual feedstuffs and compound feed mixtures.

However, in the future this ought to be changed for several reasons. Firstly, according to the present EEC-legislation, it is no longer obligatory for feed producers to inform the feedstuff composition of feed mixtures, and table values for individual feedstuffs can, therefore, no longer be used. Secondly, feed control based on a declaration of feedstuff composition and table values for each feedstuff does not discover batches of feed with low digestibility for instance caused by overheating in a drying process. Since Danish mink feed contains substantial amounts of dehydrated protein sources, biological measures like in vitro digestibility would be preferable to table values, especially for protein.

At present an in vitro method to evaluate protein digestibility is utilized in all feedstuffs

Special cages designed for digestibility trials and for mink fitted with a permanent catheter.

tested at the department in digestibility trials with mink. Preliminary results (Børsting, unpublished) have shown high positive correlations between true protein digestibility in mink and the in vitro results. However, to obtain high correlation, the 30 feedstuffs tested until now should be divided into two groups for animal and vegetable feedstuffs, respectively. For animal feedstuffs the two measures were almost equal, whereas for vegetable feedstuffs the mink values were markedly lower than the in vitro digestibilities of protein. However, also for these vegetable feedstuffs the correlation was high.

Early studies showed a much lower digestibility of saturated fats like tallow from cattle compared to vegetable oils. Two studies showed that fat digestibility can be estimated with high precision from the percentage of stearic acid (C18:0). In the first study (Jørgensen & Glem-Hansen, 1972b) the regression

$$y = 99.21 - 0.894x, \qquad R^2 = 0.88,$$

where

y = digestibility of fat, and
x = % stearic acid of total fatty acids

was found by utilizing 12 combinations of soya oil, soya lecithin, lard and shea fat. In the other study (Jørgensen & Glem-Hansen, 1973b) with 26 diets combined from 7 relevant fat sources an almost identical regression was found

$$y = 98.4 - 0.877x, \qquad R^2 = 0.86.$$

Another interesting finding was that despite the high correlation between the content of stearic acid and fat digestibility there tended to be a synergetic effect of unsaturated fatty acids on the digestibility of saturated fatty acids. However, in most diets the proportion of unsaturated fatty acids will be more than sufficient to obtain the maximal synergetic effect.

Regarding digestibility and energetic value of carbohydrates, the most important grains have been studied intensively as well as the effect of different technological treatments concerning heat treatment and grinding processes (Glem-Hansen, 1980c; Glem-Hansen & Sørensen, 1981; Børsting et al., 1993). Besides the trials with individual feedstuffs treated in different processes, also a more general approach has been taken in a study where 81 samples examined in digestibility trials were also tested for their content of α-linked glucose measured with and without autoclaving (Glem-Hansen et al., 1977).

The following regression equation could explain 92% of the variation in digestible carbohydrates:

$$y = 0.98x_1 + 0.46x_2 + 0.20x_3 - 3.37 \qquad R^2 = 0.92$$

where y = g digestible carbohydrate

x_1 = g α-linked glucose determined without autoclaving

x_2 = g α-linked glucose determined with autoclaving minus α-linked glucose without autoclaving

x_3 = g remaining carbohydrate

The regression coefficients show a significant difference in digestibility of the three fractions of carbohydrates, namely 98%, 46%, and 20%, respectively.

This equation, or maybe a similar equation including more modern and specific carbohydrate analyses, could be of interest in future feedstuff evaluation and control especially of feed mixtures where the feedstuff composition is not informed.

Despite the shortcomings of the present feed evaluation and control systems, the need for changes is less acute than could appear at first sight, the reasons being the structure of the mink feed industry and a voluntary feed control scheme. The mink feed industry is run in cooperatives governed by the mink farmers, and the voluntary feed control scheme analyses both the final feed mixtures and the individual feedstuffs, altogether ensuring mink farmers a reasonable insight in the composition and quality of the feed they buy.

Protein and amino acids

The Department for Research in Fur Animals has more or less permanently been investigating different aspects of importance for the safest and cheapest possible way to meet the requirements for protein and amino acids for mink.

During the years different methods have been used for evaluation or determination of these requirements. Prior to 1970 most of the experiments were based on traditional feeding trials evaluating growth rate, pelt characteristics, and reproduction performance (Petersen, 1957; Jørgensen et. al., 1963; Jørgensen & Clausen, 1964; Jørgensen & Glem-Hansen, 1970). From a large scale experiment (Jørgensen & Glem-Hansen, 1972a) it was concluded that the protein requirement was met with the following protein levels as percentage of the metabolizable energy:

Period	*% of ME from dig. protein*
Reproduction (1/1-20/4)	58
Lactation and early growth (20/4-31/8)	41
Late growth and pelt development (1/9-pelting)	41

when the feed composition was based primarily on fish offal.

In the late sixties and early seventies the methods for N-balance experiments with mink were developed (Glem-Hansen & Jørgensen, 1972a; Jørgensen & Glem-Hansen, 1973a). A technique for more advanced feeding experiments during the lactation period was developed and used (Glem-Hansen, 1975 and 1979a). From these experiments it was concluded that the requirement of protein with a normal quality, i.e. NPU = 65%, must constitute 43–53% of the ME during the period from parturition until weaning at six weeks of age.

The protein requirement of mink during the growth period was determined in N-balance experiments and subsequently investigated in production experiments. From the N-balance experiments the requirement was determined to be met with the following protein levels (Glem-Hansen, 1980a):

Age in weeks	*% of ME from dig. protein*
10 - 15	41
16 - 17	42
19 - 21	32
22 - 24	31

However, the following production experiments showed that the quantity of protein, which satisfactorily met the protein requirement for maximal growth, did not meet the requirement for maximal fur development.

The requirements for sulphur containing amino acids at different stages throughout the growth period were determined to be as follows (Glem-Hansen, 1980b and 1982):

Age in weeks	*Minimum level of sulphur containing amino acids (g/16 g N)*
10 - 13	3.3
15 - 19	3.4
20 - 24	4.6 - 5.1
26 - 30	3.7 - 3.8

From the investigations concerning utilization of L-cystine and L- and D-methionine it was shown that D-methionine cannot be utilized by the mink as a source for hair growth, but L-cystine and L-methionine were utilized slightly more effectively than were sulphur containing amino acids from natural feed ingredients.

During the late seventies the requirements for energy to protein and energy retention and for maintenance were investigated in respiration experiments (Chwalibog et. al., 1979; Glem-Hansen & Chwalibog, 1978; Chwalibog & Glem-Hansen, 1980; Glem-Hansen & Chwalibog, 1980). From the experiments it was concluded that the energy in growing mink was retained with an efficiency of 30%, and the total heat loss was about 50% of the gross energy.

The slaughter technique was used for determination of nutrients retained both for mink kits during the suckling period (Glem-Hansen, 1979a) and for mink at different stages throughout the growing period (Glem-Hansen & Hansen, 1981).

The results from 20 years of experiments at the Dept. for Research in Fur Animals together with results from other research institutes were the basis for a review of protein and amino acid requirements for mink (Glem-Hansen, 1992).

At present, the requirements for the individual essential amino acid are not determined. However, from the knowledge of the amino

Table 1. Requirements of essential amino acids (g a.a./100 kcal ME) as estimated by Glem-Hansen, 1992 and Clausen et al., 1993.

	Glem-Hansen, 1992		*Clausen et al.* 1993
g amino acid per 100 kcal ME	Weaning to 15th August	16th August to pelting	Weaning to pelting
Methionine + Cystine	0.20	0.30	0.26
Lysine	0.40	0.40	0.35
Tryptophane	0.03	0.03	0.06
Threonine	0.27	0.27	0.21
Histidine	0.15	0.15	<0.18[a)]
Phenylalanine	0.30	0.30	<0.32[a)]
Tyrosine	0.22	0.22	<0.22[a)]
Leucine	0.50	0.50	<0.54[a)]
Isoleucine	0.30	0.30	<0.28[a)]
Valine	0.35	0.35	<0.38[a)]
Arginine	0.40	0.40	–

a) Exact requirement cannot be determined, because of too low number of animals for pelt grading and because only the amount shown and 50% of that amount was examined for each amino acid.

acid digestibility of the most common feedstuffs and the amino acid composition in feedstuffs of several experiments it has been possible to estimate the levels which most probably meet the requirements during the period of rapid growth and the period of pelt growth, respectively (Glem-Hansen, 1992). For some of the amino acids the minimum requirement could very well be lower, but the requirements estimated by Glem-Hansen (1992) (Table 1) were considered the best estimates at the time.

Lately, determination of requirements of individual amino acids has obtained renewed interest because of the public interest in lower N-excretion from animal production to the environment. Because of the extremely harsh financial situation in mink production also any decrease in costs is welcomed.

In 1990 the department began an intense and fruitful cooperation with Research Farm West on a joint project with the aim of finding the amino acid composition which can support optimal growth, health and fur quality and at the same time diminish N-excretion.

From the first experiment (Olesen et al., 1992) it was evident that the amino acid composition is important also in mink despite the diet containing much more of all amino acids than is retained in hair and body tissues. Almost maximal skin length and fur quality could be obtained with just 20% of metabolizable energy from protein when good quality protein sources with a relatively high content of essential amino acids were fed, whereas fur quality was very poor when the same protein level was obtained mainly from fish offal yielding a lower proportion of essential amino acids.

This proved the need for a more systematic approach in the determination of the requirements of individual amino acids. Therefore, in 1991 and 1992 the effect of individual essential amino acids has been studied by feeding diets where about 50% of all essential amino acids were supplied by synthetic amino acids. This concept gives the opportunity to subsequently remove up to 50% of each amino acid still ensuring fulfilment of the requirements for the remaining amino acids.

Results from 1992 (Clausen et al., 1993) showed that the requirement of some amino acids can probably be met with lower amounts than suggested by Glem-Hansen (1992).

The requirements shown in Table 1 (Clausen et al. 1993) were determined quite accurately for methionine, cystine, lysine, tryptophane, and threonine. Hence, the lower requirements for lysine and threonine are probably more correct than the values from Glem-Hansen (1992), who could only estimate the 'maximum' requirements from experiments not specifically designed to elucidate requirements of individual amino acids. In fact, the new data in Table 1 are the first ever actually measured in mink for other amino acids than methionine and cystine.

The higher tryptophane requirement is probably caused by underestimation by Glem-Hansen (1992), because his estimate was in part based on very old data where tryptophane analyses were either missing or determined with low accuracy.

For the remaining amino acids given in Table 1 the new estimates are still 'maximum' requirements because requirement of these amino acids was determined with only 20 mink per group, and because only the amounts shown and 50% of these amounts were used. For histidin, phenylalanine, tyrosine, leucine and isoleucine, skin length and fur quality were apparently unaffected by a 50% reduction compared to the values given in Table 1 but mortality, liver size and fat content in the liver were influenced. A 50% reduction in valine supply decreased fur quality but increased skin length.

Fat and fatty acids

Most of the year mink diets contain very high amounts of fat. In recent years the fat content in Danish mink feed has increased to more than 50% of the metabolizable energy. Therefore, fatty acid composition and quality regarding oxidative stability are very important factors in mink feed.

When fatty fish species were taken into use as a replacement for slaughterhouse offal, a negative impact on health status and fur quality was found (Jørgensen, 1962). High amounts of marine fat caused an increase in the incidence of yellow fat disease (steatitis), but the use of good quality fish and antioxidants could overcome most of the problem. Later studies revealed increased oxidative degeneration when fatty fish were stored as silage, and showed that the content of free fatty acids (FFA) and peroxides could not predict the impact of the fish silage on mink performance and health (Jensen & Jørgensen, 1975).

Yellow fat disease in mink is recognized as the classical manifestation of the interaction of dietary polyunsaturated fatty acids, their oxidative degeneration products and defence mechanisms in the organism (Brandt et al., 1990). During the eighties, the prevalence of yellow fat disease diminished drastically presumably because of systematic use of antioxidants, fortification of feed with vitamin E and standardization of the feed production (Brandt et al., 1990). In the same period of time the incidence of sudden death in fast growing mink kits increased and a correlation to high incidence of preclinical myocardial degeneration was found (Brandt & Henriksen, 1986).

In a later study (Brandt et al., 1990) no influence was found on the sudden death syndrome of the three dietary factors vitamin E, selenium and of the level of polyunsaturated fat. However, it was found that the negative effects of a high dietary load of polyunsaturated fatty acids – depressed growth rate, enhanced haemolysis, steatitis and depressed erythropoiesis – could be avoided by vitamin E supplementation, whereas there was only a slight effect of selenium.

Production experiments performed in 1990 and 1991 in collaboration with Research Farm West have shown an impact on fat infiltrations in livers and on liver weights. In groups fed a relatively high protein content and a low fat content (40% and 47% of metabolizable energy, respectively) fatty livers were almost absent, whereas in groups with a relatively low protein content and a high fat content (20% and 63%, respectively), the incidence of fatty livers was dependent on amino acid and fatty acid composition (Olesen et al., 1992). In one group with a relatively high proportion of essential amino acids supplied from a number of good quality feedstuffs and with equal amounts of fat from lard, soybean oil and rape seed oil, the incidence of fatty livers was low, whereas in the other group fed the low protein-high fat diet composed of protein from mainly fish offal and fat mainly in the form of lard, the incidence of fatty livers was high. Hence, when the diet is made more inexpensive by decreasing the protein level and subsequently increasing the fat level, also the impact of the fatty acid composition must be studied carefully as well as the impact of other dietary factors of importance to fat metabolism, e.g. choline.

The oxidative stability of marine fat and the correlation to mink performance and health is studied at present at the department. Preliminary results (Børsting & Engberg, 1993) show that even though the health status of mink is negatively influenced by oxidated fish oil, the effect appears to be less serious than for instance in chickens and pigs.

Carbohydrates

As the mink is a carnivorous animal, its natural diet is low in carbohydrates and its ability to digest carbohydrates is low. The two major causes are the rapid rate of passage – 4-5 hours – and lower capacity of amylolytic enzymes (Glem-Hansen et al., 1977). This has led to many attempts to increase carbohydrate digestibility by technological treatments like heat treatment and grinding in order to physically dissolve the starch grains. Exogenous enzymes for enzymatic degradation have also been used.

In an experiment with pure starches from different sources a very clear effect of heat treatment was found. However, there was interaction between the heating process and the starch source as shown in Table 2 (Jørgensen & Glem-Hansen, 1975).

Later it was shown that mink can digest starch from both barley and wheat almost 100%, when these grains are either extruded or treated in a drum drier provided they are finely ground (Østergård & Mejborn, 1989).

Treatment of wheat with amylolytic and cell wall degrading enzymes has also shown promising results in milling fractions of wheat yielding starch digestibilities around 100% and even

Table 2. Digestibility of crude carbohydrates in different sources of starch (Jørgensen & Glem-Hansen, 1975).

Source of starch	Raw	Boiled (95°C)	Autoclaved (110°C)
Potatoes	2.1	77.3	79.9
Maize	63.1	80.7	84.2
Wheat	71.7	86.7	88.0

utilization of part of the cell wall constituents (Børsting & Damgaard, 1991; Børsting et al., 1993).

The effect of grinding on carbohydrate digestibility has been studied most thoroughly in barley (Glem-Hansen & Sørensen, 1981). There was a linear relation between decreasing particle size and increasing carbohydrate digestibility. It was concluded that at least 93% must be able to pass a 0.5 mm sieve to obtain both a high digestibility and a good consistency of the total ration of wet feed. In peas the effect of grinding on digestibility is very clear, and at least 96% must pass a 0.5 mm sieve (Børsting, 1992).

Vitamins and minerals

Regarding vitamin supplementation of mink feed, vitamin E has attracted the most attention, because of its dependence on the content of polyunsaturated fatty acids in the diet of mink. High dietary loads of polyunsaturated fatty acids can decrease vitamin E absorption, whereas vitamin E in the intermediate metabolism is needed as a free radical scavenger inhibiting peroxide formation from the polyunsaturated fatty acids (Brandt, 1987).

The adverse effects of high loads of dietary polyunsaturated fatty acids – depressed growth rate, enhanced haemolysis, and steatitis – can be avoided by adequate vitamin E supplementation (Brandt et al., 1990).

In the latest work on vitamin E it was established that the present Danish recommendation of 60 mg vit. E/Mcal ME is sufficient to overcome adverse effects of high loads of polyunsaturated fatty acids even when these are rather oxidized (Engberg et al., 1993).

Indicators of vitamin B_1, B_2, and B_6 status were searched in a study where urine, blood, liver, kidney, heart, brain, and muscle samples were taken after 28 days of feeding 3 levels of each vitamin (Jørgensen et al., 1975). The elimination through urine gave a good indication of the mink's level of supply for all 3 vitamins. Besides urine, also heart and muscle samples were good indicators of supply – but only for vitamin B_1.

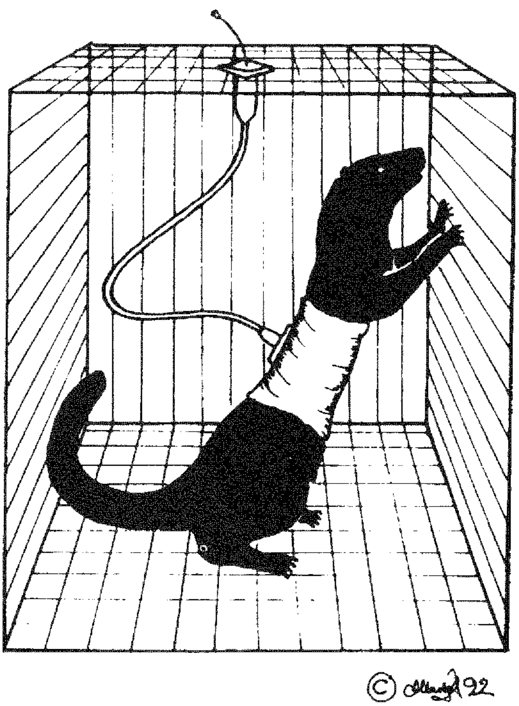
In a field trial, daily feed samples were collected from 5 feed kitchens from August until pelting (Lohi & Jensen, 1991). Both day to day variation within feed kitchen and variations between feed kitchens were quite large for some of the minerals examined. However, it was concluded that the level of all minerals had been adequate in feed from all the feed kitchens over the period as a whole. Mejborn (1989b) also found that 5.1 mg Cu per kg standard feed from a feed kitchen was sufficient to support normal growth and fur quality.

Fur quality and colour can be influenced by dietary content and availability of minerals, especially Ca, Mg, Zn, Cu, and Fe (Lohi & Jensen, 1991). A study of Zn metabolism in growing and adult male mink showed increased Zn balance with increased Zn intake (Mejborn, 1989a). At high Zn intakes, fecal excretion of Zn was the most important for Zn homeostasis, while excretion in urine also contributed significantly to homeostasis at lower intakes. Another study (Brandt & Mejborn, 1987) showed a slight increase in urinary Zn-excretion, when 50 ppm Fe-EDTA was added to the feed.

In another study endogenous Zn excretion was measured in relation to dietary Zn intake. The endogenous Zn excretion was 1.3, 2.0, and 6.4 mg corresponding to 81, 11, and 6% of Zn intake in three groups fed increasing amounts of Zn. Hence, it was concluded that at high intakes homeostatis is maintained via absorption from the digestive tract (Mejborn, 1986, 1987 and 1990). Mejborn (1987) also found increased liver Zn-concentrations, when ingestion of Zn was increased.

Metabolic studies

Recently we have adopted a method for repeated blood sampling in conscious mink to be utilized in kinetic studies of metabolism without the disturbance of neither repeated handling nor anaesthesia which would otherwise be necessary (Børsting & Damgaard, 1992). A silastic catheter is inserted into the jugular vein and the free end is extended subcutaneously to the back of the mink. From here it is inserted into a metal spiral spring with freely turnable swivels mounted in one end to the back of the mink with an adhesive elastic band and in the other end to the cage. With this tethering system it is possible to draw blood samples from outside the cage.



Mink fitted with a permanent catheter for blood sampling.

At present we have two major aims with the metabolic studies. Firstly, we want to study glucose metabolism in lactating females, especially in relation to influence of litter size, feed composition, and etiology of the metabolic disorder – nursing disease. In a recent study Brandt (1988) showed that especially a negative energy balance was a central causal factor, which was followed by low plasma concentrations of insulin and aldosterone, potassium, and haematocrit.

Secondly, we want to study amino acid metabolism and its close relation to glucose metabolism, amino acids being precursors in hepatic glucose synthesis via the gluconeogenesis.

In studies of glucose metabolism with both glucose challenge experiments and glucose turnover studies with ^{3}H- and ^{14}C-labelled glucose we have shown that glucose turnover is of the same order – 2–3% per minute – as in other species despite the fact that the natural mink diet is very low in glucose (Børsting & Damgaard, 1993).

In the lactating female we have found higher clearance rates of a glucose load compared to adult males – and at the same time the load of glucose caused less insulin secretion in lactating females (Børsting & Damgaard, 1993). This shows that the lactating mink, like other lactating animals, have a high demand for glucose in the mammary gland, where glucose uptake apparently is insulin independent as in other species.

Perspectives

As outlined above, there are still many areas within nutrition and nutritional physiology which need further elucidation in order to recommend the most optimal feeding of mink – despite the fact that research in this field has already been performed for many years.

Regarding feedstuff evaluation it appears possible in the future to leave the present system based on table values in favour of an energy evaluation system based on in vitro digestibilities of protein and possibly also of carbohydrates. Alternatively, it may be sufficient to use specific carbohydrate analyses as a basis for carbohydrate digestibility. However, the correlation from in vitro protein digestibility and carbohydrate analysis to protein and carbohydrate digestibility, respectively, must be examined in further detail. Digestibility of fat can be based on fatty acid composition.

Concerning requirements for individual amino acids, even the latest breakthrough leaves rather imprecise estimates for the requirement of many essential amino acids, especially for different stages during the life cycle of the mink.

The new data on amino acid requirement (Clausen et al., 1993) make it possible to lower the content of protein further than the level used at present in common Danish mink feed on the assumption that the levels of essential amino acids shown in Table 1 are met. However, this study has to be followed up by new studies with certain levels of each amino acid – and to minimize N-excretion also different levels of each amino acid should be examined in different parts of the year.

However, knowledge of the requirements of individual amino acids evidently makes it possible to further decrease dietary protein content and consequently reduce feed costs and N-excretion to the environment. Before recommending extremely low dietary levels of amino acids, it is very important to continue the joint efforts in production trials and physiological trials examining metabolic and health parameters, when dietary protein is reduced and especially fat content is increased. If protein level is decreased in the future the effect of fatty acid composition and the oxidative stability of dietary fats must be followed carefully, as well as the requirements of vitamins, especially vitamin E.

Evaluation of feedstuff quality based on a combination of biochemical analyses of fatty acid composition and oxidation products as well as other important quality parameters such as biogenic amines/amino acids and toxins are important research areas in the future. Both for existing and for new analyses of such parameters capillary electrophoresis appears to be an important new tool, which has just been taken into use at the department.

Regarding grains and carbohydrates, the rapid development of exogenous enzyme products used as feed additives will probably point to more research examining the effect of such enzymes compared to other treatments performed to make carbohydrates available to the mink.

In the future the nutritional research will be concentrated on metabolic studies performed jointly with production trials. In the nearest future glucose and amino acid metabolism will have the highest priority. Besides giving results of immediate value to the fur industry, this will, hopefully, further intensify the collaboration with the remainder of the research group of The Danish Fur Breeders Association, especially on Research Farm West.

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Morphology, Morphometry and Histology in Hair and Skin

Palle V. Rasmussen

Introduction

With chief scientist Outi Lohi as prime mover and inspired by her basic ideas about the importance of knowledge of the morphological and histological conditions on the final fur animal product – the pelt – the department in 1986 initiated research on morphological characteristics of hair and skin. In the same year – after a week's introduction by the hair morphologist Leena Blomstedt in Finland – the establishment of laboratory facilities and development of methods were started. Today, the hair morphological laboratory has developed and specialized considerably, both with regard to equipment and the work it is involved in. This has to a great extent been possible because the trade and the department have constantly supported and invested in this research area.

The area is now centrally placed in connection with further development within topics like breeding and nutrition, documentation of effects of experimental treatments, documentation and illustration of the occurrence of fur defects (perhaps also the reasons for such defects), determination of the optimal pelting time, intervention in hair formation, characterization of colour and pigmentation of hair fibres, and last but not least the development of different objective measuring methods.

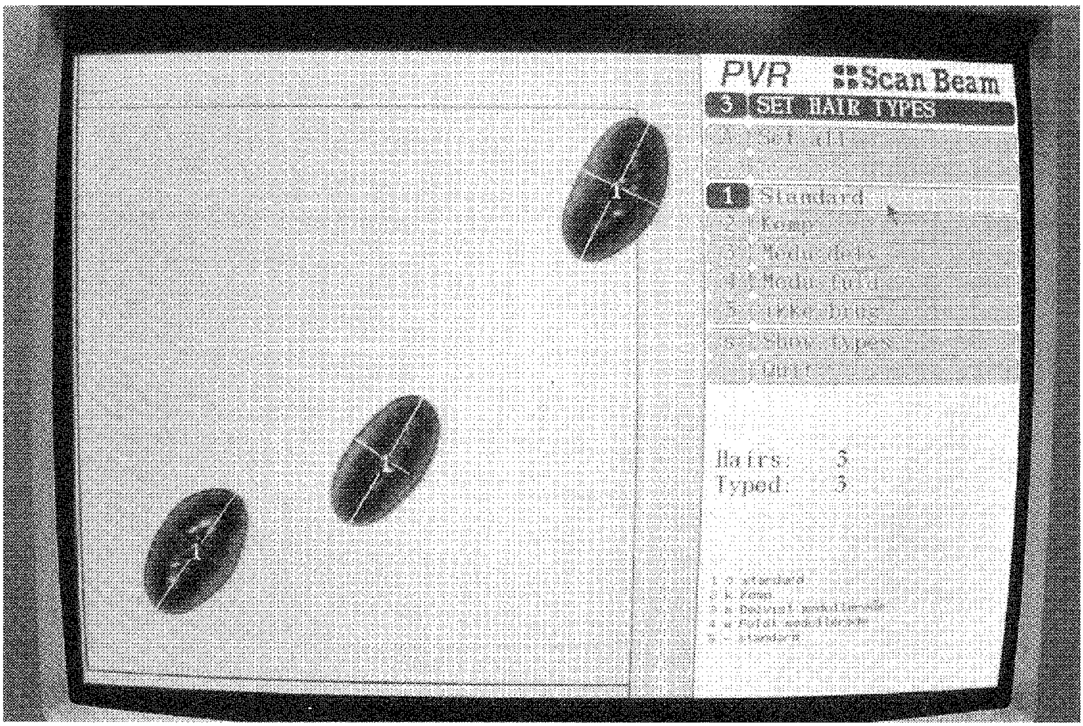

Figure 1. Electronic image analysis. 3 cross sections of guard hairs of mink have been analysed morphometrically.

It is now recognized that research in hair and skin is a necessary condition for specifying the concept of hair and fur quality and thus also for development, improvement and quality control of the final product, no matter whether it is fur from farm raised mink or foxes or special hair fibres of goats and sheep.

Development of objective methods

As the majority of the examinations are of a micromorphological nature, great efforts have been put into developing and adapting methods based on microscopy, which can be divided into several subdisciplines.

Image analysis

Light microscopic methods are the main basis of the objective measuring methods used at the laboratory. The morphometric measurements and histological studies of skin biopsies were initially carried out manually with a light microscope. Later, a semiautomatic electronic image analysis system especially designed for hair morphological examinations (Fig. 1) (Rasmussen, 1989b) was developed together with Scan Beam A/S. In the preliminary phase, prime mover was Asbjørn Brandt, DVM, at the time employed at the department. In 1992 the cooperation with Scan Beam A/S was resumed. This resulted in a reconstruction and upgrading of the original prototype of the system, so that today it must be regarded as a unique tool in hair studies (Scan Beam, 1992; Rasmussen, 1992c). For examination of hair and skin preparations 4 semiautomatic programmes are available, covering: 1) hair fibre analysis, 2) general measurement of area and length, 3) hair density and grouping and 4) areas with defined grey tones. All in all this means that we now have a measuring equipment with a considerable potential even for future demands.

Histological technique

The application of image analysis requires extensive work for the development of selective, histological staining methods. Furthermore, the different analyses often require the use of different embedding media (for instance paraffin or plastic) which again requires current development or adjustment of methods, thus demanding remarkable creativity and patience from the laboratory technicians.

Electron microscopy

On a small scale, examinations of both hair and dried skin have been performed by the use of both scanning and transmission electron microscopy (SEM and TEM). In general, the SEM-examinations at Research Centre Foulum are carried out in cooperation with the Department of Animal Physiology and Biochemistry.

Microspectrophotometry

Based on colour examinations of feathers and later pilot experiments on hair from both mink and foxes carried out at the University of Copenhagen, it was in 1991 possible to apply for and obtain financial support from the Danish Agricultural and Veterinary Research Council for a specific project. This meant that within the discipline microspectrophotometry it has been possible to work with the development of objective micromorphological methods for colour characterization of hair. Furthermore, it has with the upgraded version of the image analysis equipment with advantage been possible to combine and document information of different physical nature but of analogue importance to for instance hair colour.

Macroscopical and physical methods

Not all examinations require the use of microscopy. Macroscopically the length of hairs is measured on standardized skin areas. This procedure is often combined with hair mass determinations. By simple weighing of hair fibres from standardized punched out skin samples, extremely objective and useful information can be obtained.

Important applications and results

The previous chapter has indicated that the research tasks have until now been and will also in the future be rather diversified. Various animal

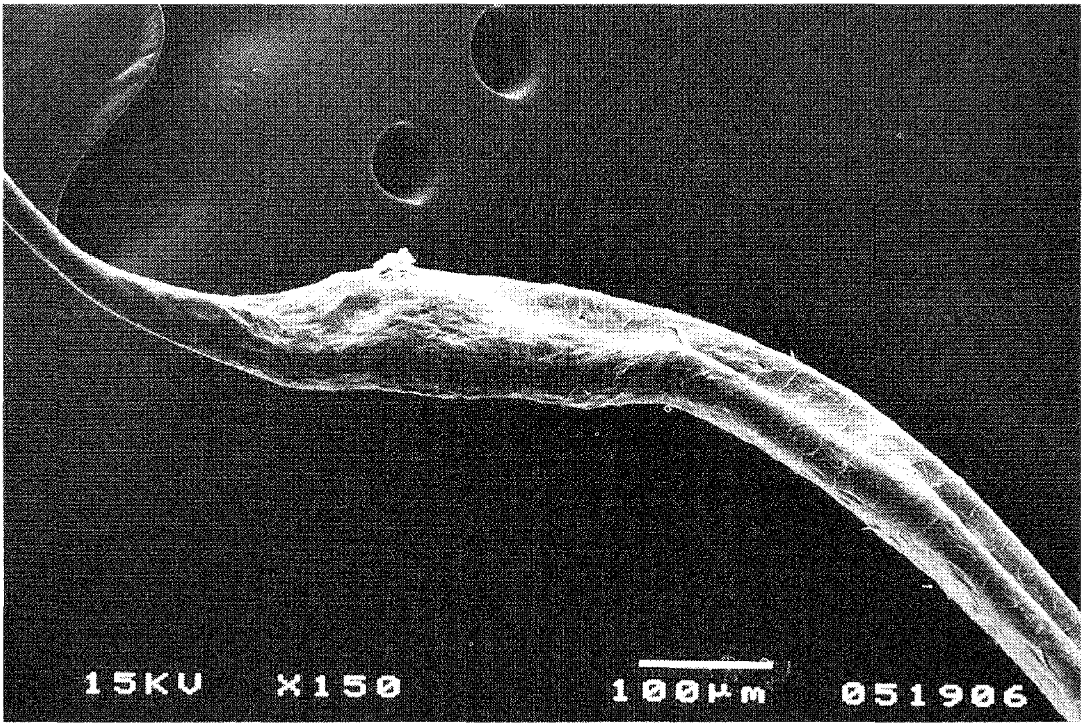

Figure 2. Scanning EM. Micrograph of a 'curly' guard hair of silver fox.

species are in focus. In the following, the most important applications and results for the individual species will be described.

Defects and abnormal hair growth

Most projects have been dealing with farmed mink and foxes. As a sub-project under the project 'Production factors and production control', an investigation of objective methods to evaluate fur quality was started with 200 mink pelts from 20 different farms. The title was 'Morphological differences in hair types from different scanblack strains'. Differences as regards hair mass and the shape and thickness of the guard hairs were documented and compared with a subjective quality evaluation and the mineral content in the hairs (Rasmussen, 1988; Møller et al., 1991).

At the same time another interesting and always relevant research area was started with the common title 'Morphological description of different hair defects'. In general, the purpose is to describe the details of certain hair defects. Exact knowledge about the defect is necessary in studying the reason for it and in order to eliminate the problem. An illustration of the problem with so-called abnormal guard hairs ('flyvehavre') generated lively discussion of this problem at the Annual Meeting of the Danish Fur Breeders Association in Herning 1988. Several other publications on fur defects have followed (Rasmussen, 1988; Rasmussen, 1989c; Rasmussen 1991b).

In connection with an abnormal hair development in silver foxes, resulting in the formation of deformed guard hairs ('curly' silver fox), light and scanning electron microscopic studies were initiated (Fig. 2), showing that the defect is especially seen in the 'silver bar' part (Rasmussen, 1988). Later on, histological examinations of foxes with the defect have revealed remarkable cell morphological changes in the anagen guard hairs (Rasmussen, 1990).

Based on material from the project first mentioned supplemented with experimental pelts from Research Farm West, investigations were started to find an exact description of the skin defect 'reduced hair growth on hips' on mink pelts. This was part of a large project to solve the problem (Rasmussen & Lohi, 1988). It was furthermore documented, that there were relatively few underhair follicles in the hip area of mink with this defect, and that in general the pelts had shorter and thinner guard hairs (Rasmussen, 1989a). In this connection, skin biopsies from 57 mink were also analysed histologically/histochemically for a quantitative determination of the structural protein elastin. No significant effect of the defect on elastin content could be documented. For the introduction of stereological analyses the examination was, however, extremely instructive (Rasmussen, 1992b).

As a regular study of an abnormal change of fur coat, one specimen of the farmed blue fox was subjected to an extremely interesting histological and clinical-chemical examination. It could be concluded that both the hair growth cycle and estrogen profile in plasma of the fox showed an abnormal seasonal variation (Rasmussen & Damgaard, 1992).

At the request of and with support from the Committee for Hair Research of the Danish Fur Breeders Association, the department in 1991 got involved in the problem of the so-called 'oxidized mink pelts'. The Danish Fur Sales had ascertained that dried pelts kept under normal conditions could be subjected to oxidation. This can be seen as yellow or brown areas on the leatherside which for several reasons reduces the value of the pelt. In a recent report it was documented that immediately after pelting the leather side of the skin can be considered almost sterile. Therefore it is important in future research to focus on sources which may contaminate the pelts. It was furthermore demonstrated that different biocides could, to a considerable extent, hamper or prevent the growth of fungi and bacteria (Rasmussen & Jensen, 1993). Further investigations are necessary.

Effect of feeding on hair growth and hair quality

In connection with feeding experiments at Research Farm West, mean hair quantity and hair length as a function of feed composition have been examined. It has been possible to prove several significant correlations between these hair parameters and fur quality (Olesen et al., 1992). The investigation of the effect of feeding is continued in feeding trials with different protein levels and amino acid composition at the same research farm.

Development of new hair types. Product development

In connection with genetic experiments, the winter coat of the American short nap mink (the 'velvet' type) was characterized morphologically in relation to the normal Danish type (scanblack). Preliminary results have shown that the short nap type have shorter guard hairs with changed proportions. Besides, the underhairs are of normal length, but the number of these is increased significantly, as the number of hairs per follicle group is increased (Rasmussen, 1991a). Further results will be published in 1993 on this subject.

Silky hair type is a valuable component of fur quality. At fur auctions, pelts with a silky, i.e. attractive surface, obtain a better price. To clarify which objectively measured hair fibre parameters are important to silkiness, several minor examinations were started in 1991. These combine subjective and objective analyses of the same pelts. The basic question is how the visual and the tactile impression of silkiness differ and whether they are caused by the same physical hair properties. It is important to describe the morphological parameters of the individual guard hairs and to compare these with the result of a subjective pelt evaluation. Table 1 illustrates the result of a pilot experiment (Rasmussen, 1992a). This research area is so relevant that the department has now initiated a selection experiment for development of a silky hair type. The project thus combines morphological and genetic investigations.

Documentation of physical methods

In cooperation with the technical department of the Danish Fur Breeders Association, several

Table 1. Pilot investigation of long guard hairs from scanblack mink in relation to silkiness.

Pelt No.	Objective measurements: No. of scales/80 μm lancet (top) N = 4	No. of scales/145 μm lancet (middle) N = 4	Thickness of lancet (μm^2) (max.) N = 20	Lancet shape (B/A) % N = 20
1	21.2 ± 1.9	28.2 ± 1.8	11331 ± 1174	52 ± 4
2	17.6 ± 2.5	28.3 ± 2.4	8794 ± 887	58 ± 4
3	18.4 ± 2.2	27.0 ± 3.6	11034 ± 854	56 ± 3
4	16.8 ± 1.5	23.7 ± 1.4	10604 ± 792	61 ± 4
5	21.8 ± 0.7	29.0 ± 1.1	10623 ± 988	53 ± 3

Pelt No.	Subjective measurements: Silky	Relatively coarse hairs	Relatively fine hairs	Relatively coarse surface
1	–	++	–	–
2	++	–	++	–
3	–	–	–	–
4	–	–	–	++
5	++	–	–	–

investigations of common pelt material were carried out until the end of 1992. These investigations have documented correlations between actual hair quantity and pelt density measured by means of physical methods at the technical department. An important work regarding further documentation and especially interpretation of the various types of results still remains to be done.

Hair growth in fur rabbits

In 1990 a cooperation with the Department of Poultry and Rabbits was started. The objective was to illustrate the fur quality and fur priming of the Castor Rex rabbit in relation to age and feeding intensity. It could among other things be concluded that a pelting age higher than 6 months can produce a reasonable product (Petersen & Rasmussen, 1991). Later on, further investigations of pelt development have been performed, but as regards product development considerable work still remains to be done.

Quality tests of hair fibres in mohair goats and sheep

In 1991, electronic image analysis for examination of both muscle and hair fibres was demonstrated at the Agricultural Exhibition in Herning. With research scientist Bente K. Hansen as intermediary, this resulted in a contact with the Danish Mohair Association and gave rise to the development of an objective Danish quality test of hair fibres from mohair goats. This has developed further to a close contact with new collaborators within the National Institute of Animal Science and the Agricultural Advisory Centre, Skejby. As a result of this, the laboratory has developed an objective test by which the average fibre diameter, cross sectional shape and the percent of medullated fibres and unusually large fibres are determined. The test is sold to private producers (Rasmussen, 1992 d and e). Furthermore, analyses of hair samples from animals participating in specific projects are performed. Our intention is to develop and standardize the mohair/wool test to the extent where it can be recognized as an international test method.

Perspectives

Even though it can be said that the morphological hair research at the department has come a long way, it can also be established that there are still important, unsolved tasks waiting ahead. This applies to mink and foxes as well as fur rabbits, mohair goats and sheep. As regards mohair goats and sheep, a large and almost unexplored research area for the National Institute of Animal Science is waiting.

It is, however, even more important, and extremely positive, that the department – but especially Gunnar Jørgensen – has succeeded in convincing the Danish Research Secretariat of the Ministry of Agriculture that hair and skin from mink and foxes are to be regarded as raw materials which can be further developed. It has further been established that morphological and biochemical research and accumulation of knowledge are crucial for causal interpretations and for development and quality control of the product – the fur animal pelt. In practice, this has resulted in a 5 year project of morphological and biochemical studies, for which the Danish Research Secretariat has granted the department the amount of 3.5 mill. DKK in connection with the project 'Biomass for Non Food Purposes'. The project period is 1993-1997.

The morphological programme will in the time to come be concentrated on further development of methods to produce objective parameters for the breeding and nutritional research work. Further accumulation of specialized knowledge of methodology and normal values of hair and skin parameters is imperative in itself.

More specificly, the development of methods for colour characterization of hair will be in focus, including both the measurement of the colours of hair fibres and the determination of the degree of pigmentation in growing (anagen) hairs. Here the preparation of models for illustration of various colour relations will have a central position. At the same time it will obviously be a great challenge objectively to describe quality concepts like for instance silkiness. The important, overall objective is to give the primary producer powerful and effective tools to control and influence the quality of his pelt production.

Finally, it should be mentioned that with a combination of the morphological and biochemical disciplines, there will hopefully be good possibilities to further develop the experimental, cell biological research in hair and skin.

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Biochemical Investigations in Relation to Fur Development and Fur Quality in Mink

Søren Michaelsen

Introduction

Changes in the shape and appearance of hairs, as well as defects in hair and skin found by subjective evaluation of the product have been observed for a long time. The differences in appearance and the various defects are likely to be caused by differences in the biochemical composition of the hair and skin. There is a potential for affecting the development of hair and pelts towards products of higher quality, if knowledge of the composition of normal pelts and pelts with disorders is obtained. Due to the close physiological relationship between skin and hair, biochemical investigations on the composition of both skin and hair are likely to give answers to what the differences are, and which factors they are caused by. The potential value of the biochemical investigations is independent of whether the changes or defects are caused by genetic, nutritional or environmental effects.

The work is very extensive, as it includes determination of normal values and individual variations in the contents of various compounds found in hair and skin with an expected connection to the quality of the pelts. On the low molecular level, the compounds of interest include amino acids comprising also the special amino acids hydroxy prolines, hydroxy lysines, desmosine, and isodesmosine, as well as glycosaminoglycan disaccharides. Proteins, proteoglycans, and enzymes are included on the high molecular level. It is essential to have a number of analytical methods for determination of these compounds. These methods should be as simple and cheap as possible and yet give precise and specific information on the compounds of interest. Therefore, the development of new methods and adaptation of existing methods to the analysis of hair and skin components have a high priority.

The biochemical investigations on mink skin were initiated in 1982 on the initiative of Gunnar Jørgensen and Outi Lohi. In 1988 a collaboration with Hilmer Sørensen, Chemistry Department, The Royal Veterinary and Agricultural University was established for continuation of the biochemical studies and education of a specialist in this field. Recently, a large part of the investigations and analyses was transferred to the department.

The investigations can be grouped in studies based on low specific methods and high specific methods. The low specific methods included measurement of proteins and glycosaminoglycans as amounts precipitated under various conditions, residues left after enzyme treatments, and spectrophotometric methods for quantitation of uronic acid, proline, hydroxy proline, lysine, and hydroxy lysine after hydrolysis and/or reactions with chromophores. The high specific methods used until now include solid phase extraction, HPLC, FPLC, capillary electrophoresis, gel electrophoresis, and specific enzyme assays. The methods have been used for quantitations of individual amino acids and glycosaminoglycan disaccharides, analyses of individual proteins, and determination of enzyme activity.

Low specific methods

Generally, the investigations performed have included determination of hydroxy proline and hydroxy lysine as a measure of the content of collagen, proline as a measure of the content of protein, hexuronic acid as a measure of the content of glycosaminoglycans, and neutral sugars as a measure of the content of glycoproteins. All results are reported here as content in dried defatted skin.

Topographical differences

Topographical differences in the content of proline, hydroxy proline, and hydroxy lysine have been reported in scanblack skin by Blumenkrantz (1985a). The content of proline, hydroxy proline, and hydroxy lysine was higher in the dorsal samples than in the ventral samples of the corresponding location. From tail to head the composition changed in a non-systematic way. Higher contents of water and fat were found in samples from the ventral side compared to samples from the dorsal side.

The effects of feeding

Studies on a possible connection between connective tissue components and fur quality were performed in two experiments with biopsies from two groups of scanblack mink (Blumenkrantz, 1985b; Blumenkrantz & Lund, 1989). In the first experiment, one group was fed ad libitum, and the other group was fed restrictively. In the second experiment, two different energy levels were used. Both experiments were performed from July to December. The parameters investigated were hydroxy proline, hydroxy lysine, proline, hexuronic acid, and neutral sugars. No statistically significant correlation between the biochemical parameters and the subjectively determined fur quality was found. It may be questioned, whether the methods of analysis are specific enough to reveal such differences. However, the biochemical parameters investigated were affected by the feeding. The contents of hydroxy lysine and neutral sugars were slightly higher in the restrictively fed and low caloric groups, and the contents of hydroxy proline and proline were higher in the ad libitum fed and high caloric groups. The hexuronic acid content was unaffected.

The effect of feed containing silage of filleting scrap or rapeseed meal on the connective tissue components in skin of pastel mink was also investigated (Blumenkrantz & Hillemann, 1989a). Decreased body weight and skin length were found for the groups given the two experimental diets compared to the control group. Increased content of water, hexuronic acid, and hydroxy proline, and decreased content of fat and proline was found in the skin of mink fed rapeseed meal. The skin from mink fed fish silage showed increased content of hexuronic acid and hydroxy proline, and decreased content of proline and neutral sugars. In conclusion, the energy intake and the feedstuffs used may influence the composition of the skin and the subjectively determined quality of the skin. However, at present no correlations between the biochemical composition and the quality of mink skin have been found.

The effect of chelators

Feeding mink kits (4 to 8 weeks of age) a diet added the antiviral-antibacterial agent Vantocil 1B® (polyhexamethylene biguanidine hydrochloride), and whose dams received the same feed during the gestation and lactation periods, produced depigmentation of the underfur (Blumenkrantz & Hillemann, 1989 a and b). Vantocil was shown to be able to chelate copper, iron, and zinc ions, and the activity of the enzyme tyrosinase, which is involved in melanogenesis, is known to require copper and iron ions. Furthermore, Vantocil can solubilize melanin. The effect of Vantocil may be a result of decreased enzyme activity and/or interference of the tyrosine-melanin metabolic pathway due to solubilization of melanin. The latter necessitates the presence of Vantocil or Vantocil products able to solubilize melanin in the hair bulbs. Feeding a diet containing Vantocil to animals from ten weeks of age to mid-November resulted in improved quality but decreased colour intensity determined subjectively. Furthermore, the content of water, hexuronic acid, and hydroxy proline increased, and the content of proline and hydroxy lysine in skin decreased (Blumenkrantz & Hillemann, 1989a). The authors concluded that careful biochemical studies of products to be added to mink feed should be performed to avoid unwanted problems.

The effect of daily subcutaneous injections in scanblack mink of Cu^{2+} and Fe^{2+} ion chelators was studied by Blumenkrantz & Blomstedt (1987). Some of the chelators gave skin and hair depigmentation, increased elasticity of mink skin, and disturbed growth of hair. However, no alteration of mink fur quality was found for any of the chelators. The depigmentation was probably caused by decreased or no activity of the enzyme tyrosinase due to the lack of copper. The increased elasticity might be caused by the lack of Cu^{2+} and/or Fe^{2+} as cofactors for the enzyme lysyl oxidase and thereby decreased collagen and elastin crosslink formation. The results support the proposed action of Vantocil in depigmentation of hair.

The methods used did not give any information on whether changes in the composition of e.g. collagen or proteoglycans/glycosaminoglycans could be found or of the exact structure of the compounds. Furthermore, the methods applied could not determine which of the two possible isomers of hydroxy proline or which of the four possible isomers of hydroxy lysine are present in skin. Because this information was considered of interest on the background of the results obtained, and because of the value of having more specific determinations, high specific methods were developed and applied.

High specific methods

Amino acids

The first results on amino acid content in skin and hair of mink pelts were reported in 1971 by Jørgensen & Eggum (1971). The amino acid composition of skin and hair was very different. Especially the content of glycine was much lower, and the contents of tyrosine and cystine were much higher in hair compared to skin. The amino acid composition of hair from tail, dorsal, and ventral samples did not differ much.

In 1988 the investigations on individual amino acids in the skin were taken up again. Methods of analyses were evaluated, and a combination of the σ-phthaldialdehyde (OPA) derivatization of primary amino groups and dansyl (DNS) derivatization of primary and secondary amino groups and amino acids was chosen for the analysis by HPLC (Fig. 1) (Michaelsen & Sørensen, 1989a; Michaelsen et al., 1991). Hydroxy lysines, desmosine, and isodesmosine can be detected by both methods of derivatization, whereas proline and hydroxy prolines can only be detected by the DNS method.

It has been shown that the four possible isomers of both 4-hydroxy lysine and 5-hydroxy lysine can be separated by the HPLC method for analysis of OPA-amino acid derivatives prepared by use of a chirale reagent (Michaelsen et al., 1991). However, this HPLC method cannot be used for the determination of the stereoisomers of hydroxy proline, and therefore another method is needed. In this connection the development of a capillary electrophoresis method is of interest. The amino acids in skin all have the 2S configuration. The stereoisomer of hydroxy proline found in skin is believed to be (2S, 4R)-4-hydroxy proline, whereas the configurations of the 4- and 5-hydroxy lysines are unknown as well as the relative content of these hydroxy lysines (Alsly et al., 1990).

The amino acid content can be expressed as the amount per gram of skin, the amount per area unit of skin, or as the relative amount (mol percentage), and the relevance of these measures has been discussed (Alsly et al., 1990).

Both fresh and dried skins were analysed, and differences were found in the absolute content of amino acids, whereas the relative contents of amino acids showed limited variations (Michaelsen et al., 1991). This is most likely a result of the different amount of fat in the biopsies, and the extent of stretching of the skin during drying. In fresh skin the amount per area of skin and the relative amount are the most relevant measures, whereas in dried skin all three measures can be used.

The topographic variations in pelt protein amino acids were large (Michaelsen et al., 1991). Dorsal and ventral samples as well as samples from the hips and samples obtained from head to tail contained different amounts of amino acids. This means, that the results from pelt protein amino acid determinations have little value, unless it is clearly stated from where the sample has been taken.

Amino acid determinations from skin of standard mink with and without the disorder 'reduced hair growth on hips' showed characteristic differences (Michaelsen et al., 1991). The relative contents of glycine, lysine, hydroxy lysine, and threonine were lower in skin with the defect, and the relative contents of alanine, aspartic acid/asparagine, glutamic acid/gluta-

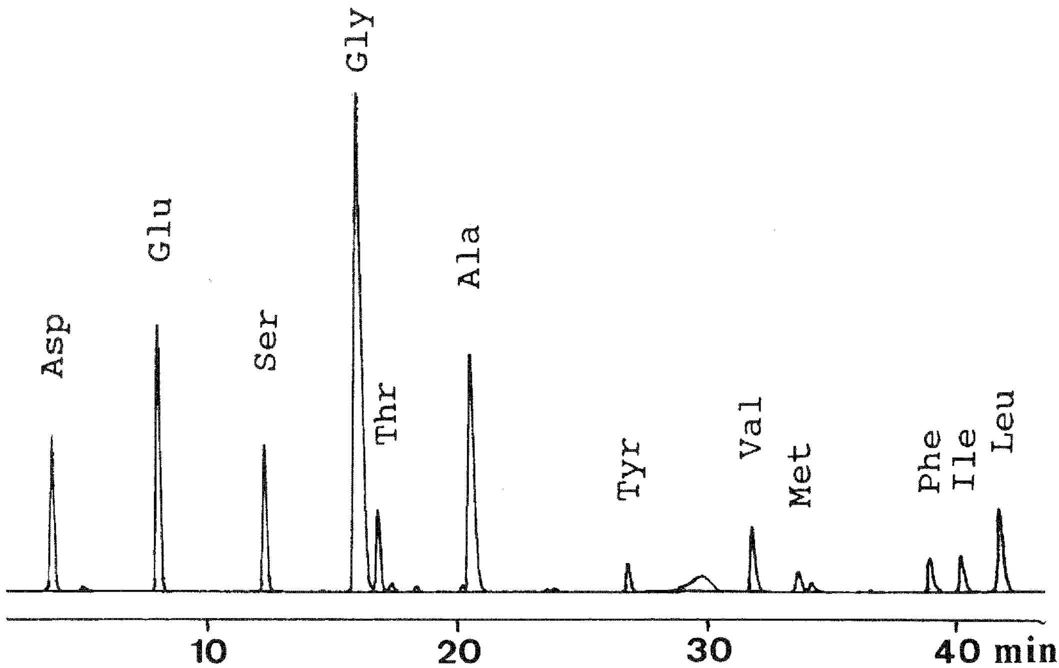


Figure 1. HPLC chromatogram of OPA derivatized neutral and acidic amino acids obtained from hydrolysed mink skin.

mine, and arginine were higher in normal skin. This indicates that there are appreciable differences in the types and composition of structural proteins occurring in the two types of skin, and that the defect may thus reflect metabolic differences during skin/hair growth.

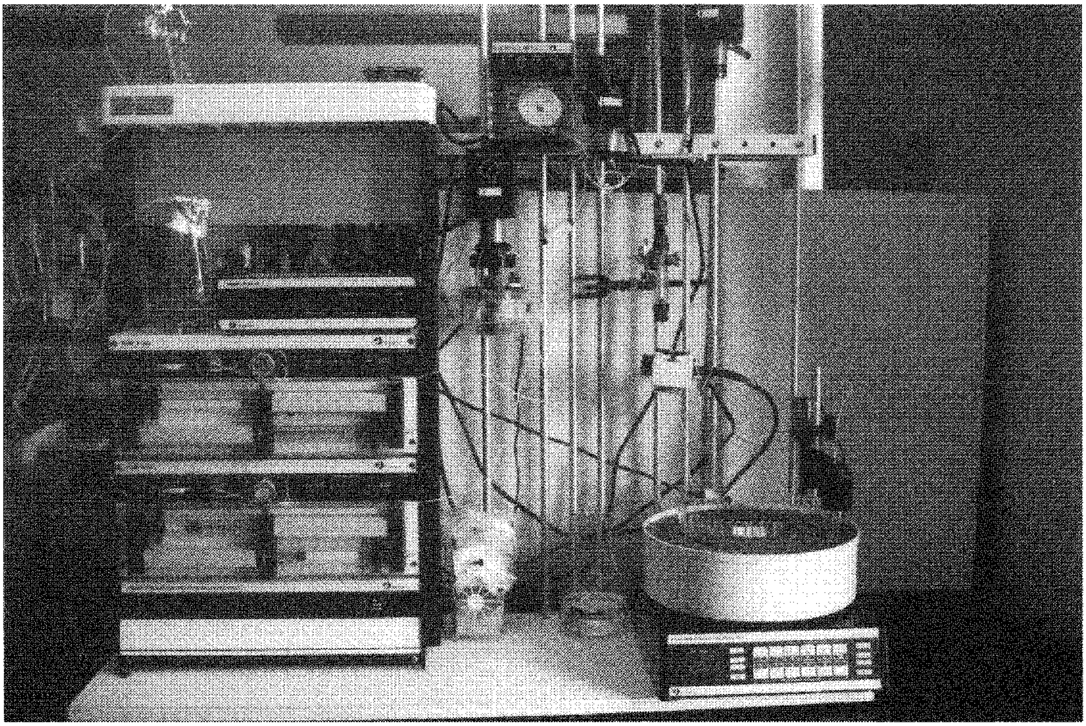
Glycosaminoglycans

Glycosaminoglycans are skin constituents assumed to be important for the properties of mink pelts (Blumenkrantz, 1985a; Michaelsen et al., 1993). Information on structure and quantity of glycosaminoglycans in mink skin is therefore wanted. A new method of capillary electrophoresis based on micellar electrokinetic capillary chromatography has been developed for the analysis of glycosaminoglycan disaccharides (Michaelsen et al., 1992 and 1993). The method gives efficient separation and quantitation of the analysed standards and disaccharides obtained from chondroitin sulfates. Skin samples are protease and chondroitinase ABC treated to obtain the disaccharides. At present the protease treatments of the skin have to be further improved to obtain sufficient material from samples to perform accurate analyses.

Proteins and tyrosinase

The amount and properties of intact proteins in skin have been investigated. Testing several extraction methods revealed very large differences in the amount of protein extracted from skin. An extraction buffer consisting of Tris, sodium chloride, and sodium cholate resulted in the highest amount of proteins extracted (Michaelsen & Sørensen, 1989 b and c). However, for some of the following analyses this buffer created problems.

Intact proteins extracted by methanol-water yielded a total of 20% of the proteins with molecular weights above 10 kD and 80% with molecular weights under 10 kD (Michaelsen & Sørensen, 1989a). The amount of glycoproteins in the proteins extracted was only around 2%.



FPLC equipment for protein and enzyme analyses.

Preliminary isoelectric focusing analyses revealed different patterns and intensity of proteins in the protein fractions according to the isoelectric points of the proteins present (Michaelsen & Sørensen, 1989a). Furthermore, SDS-PAGE analyses of protein subunits showed many subunits with molecular weights between 15 and 90 kD. The number and intensity of subunits found depended on the protein fraction analysed.

Studies of the metabolism of proteins in the mink skin have been initiated. The precursers used have been ^{14}C-labelled amino acids as well as glucose. The aim of these studies is identification and isolation of special radioactively marked skin constituents such as collagens, elastins, glycoproteins, glycosaminoglycans, melanins, and special amino acids. The preliminary results show that only 2 to 10% of the subcutaneously injected radioactivity was excreted in the urine during day 1 to 9 after injection. The chance of finding radioactively labelled skin constituents is therefore high.

The amount of tyrosinase activity extracted from skin by various methods was also investigated. The same system as for proteins was chosen due to high total activity extracted and high specific enzyme activity (Michaelsen & Sørensen, 1989b). It appeared as if the activity in the skin generally decreased with increasing age of the animals. This may be caused by a tighter binding of the enzyme to the membranes of the cells of the older animals or to differences in the developmental stage of the fur. However, the enzyme activities found in all skin samples were low, and a more sensitive assay would be preferable.

Minerals

The mineral content in mink hair in relation to the mineral content in feed has been investigated (Lohi et al., 1991). A clear correlation between the amount of Ca, Mg, Na, and Se in feed and in hair was found. The pelt quality could not be related to minerals in feed or hair, whereas a significant correlation between hair colour and the content of Ca, Mg, and P in hair was found. Of the single minerals only Ca showed correlation to morphometric characters of hair.

Consequences of suboptimal storage

Mink pelts stored under different conditions and with and without stickiness of fur were analysed for amino acids, biogenic amines, and fatty acids. The analyses of protein bound amino acids and non-protein bound neutral and acidic amino acids did not reveal any differences between the pelts. However, the fatty acid composition of hair and especially skin as well as analyses of biogenic amines from the pelts did show that appreciable microbial activity had occurred in the sticky furs. Protection against microbial activity is therefore required to avoid these changes.

Perspectives

Generally, production of data showing the normal content and the normal variation for different compounds and groups of compounds in skin and hair of fur animals is essential. So is the determination of correlations between these skin constituents and the quality of the pelts or the occurrence of pelt disorders. Furthermore, the investigations will include further method development with respect to some of the specific methods. Especially the possibilities and advantages of making cheaper and more effective analyses by the use of capillary electrophoresis will be investigated.

Amino acid investigations will have to include amino acids in hair as well as determination of the special amino acids in skin. Calculation of the contents of special proteins in skin (e.g. elastin) based on the determination of specific amino acids will be performed. The influence of feeding various levels of protein and different qualities of protein to mink is one of the important nutritional projects. In connection with this project biochemistry is used to investigate the effect of feeding on the amino acid content of the skin and possible correlations between skin composition and fur quality.

The glycosaminoglycan studies will have to include improved protease treatments of the skin, in order to obtain accurate results from the analyses. Furthermore, isolation and characterization of proteoglycans (glycosaminoglycans and attached protein) may also be included in the studies.

Continued investigations of intact proteins including characterization of collagens and elastin will also have a high priority. The studies of metabolism of proteins will be a valuable tool in this connection. Furthermore, additional enzyme studies including tyrosinase activity and properties as well as other important enzymes such as prolyl hydroxylase, lysyl hydroxylase, and lysyl oxidase are also necessary. Finally, the characterization of melanins should be continued by the use of some of the new and promising methods. Acute problems in the production and storage of pelts will be included in the biochemical investigations to the extent required.

In conclusion, biochemical investigations in hair and skin are necessary to obtain knowledge on what the reasons are for – and how to avoid – differences in quality of pelts and the occurrence of pelt disorders. It is a comprehensive work, but various promising results have already been obtained, and with the new techniques available much more is possible within this area.

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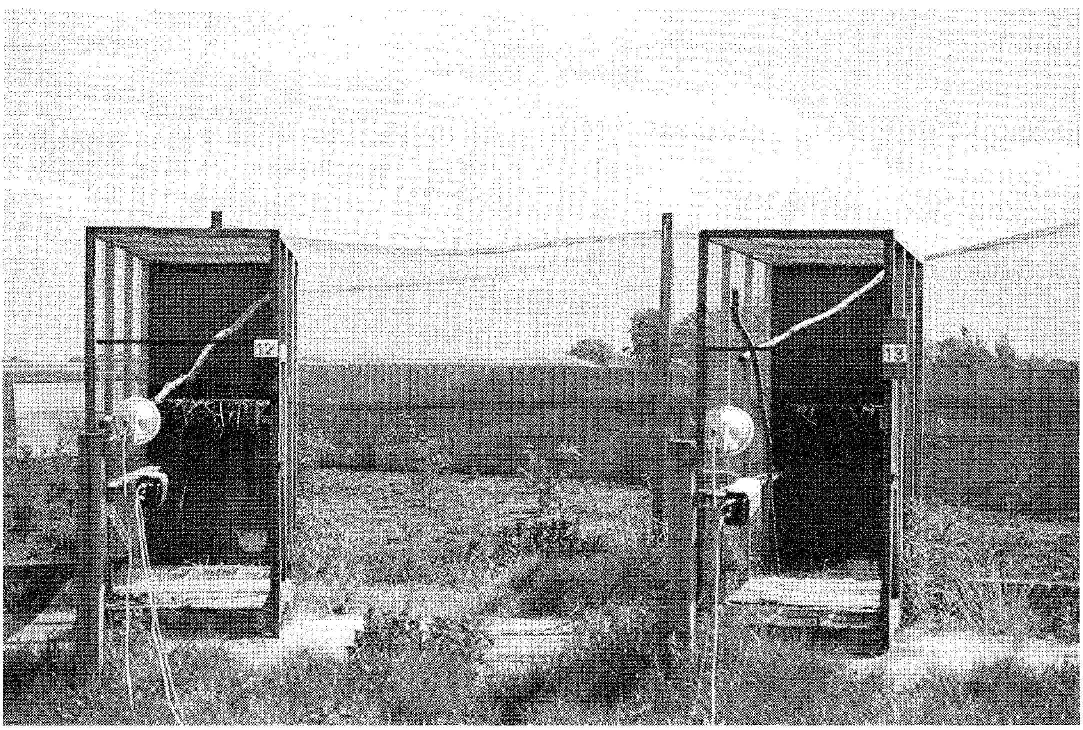
Stress and Behavioural Biology

Steffen W. Hansen

Introduction

At the initiative of Gunnar Jørgensen, the department in 1985 started the first ethological examinations of farm mink in a close and fruitful cooperation with the University of Copenhagen. The cooperation lead to an intensified effort in the behavioural area, as several ethological theses (Dodd, 1985; Jonasen, 1987; Damsgaard, 1987; Kristensen, 1988) were prepared at the department. Concurrently, the University of Copenhagen carried out basic investigations at the research farm Trollesminde on the behavioural and stress physiological reaction patterns of farm mink (Heller & Jeppesen, 1985 and 1986; Jeppesen & Heller, 1985 and 1986).

In 1985 the section for behaviour, production and environment was started at the Department for Research in Fur Animals. At an early stage, the ethological work with farm foxes was, for practical reasons, placed at Research Farm North in Jutland, while the investigations on farm mink continued at Trollesminde. When the department was moved to Foulum, the physical conditions for the ethological work were improved considerably. A special farm section with improved possibilities of behavioural observation was established in the new sheds. Besides, an outdoor pen was established for more extensive housing of fur animals.



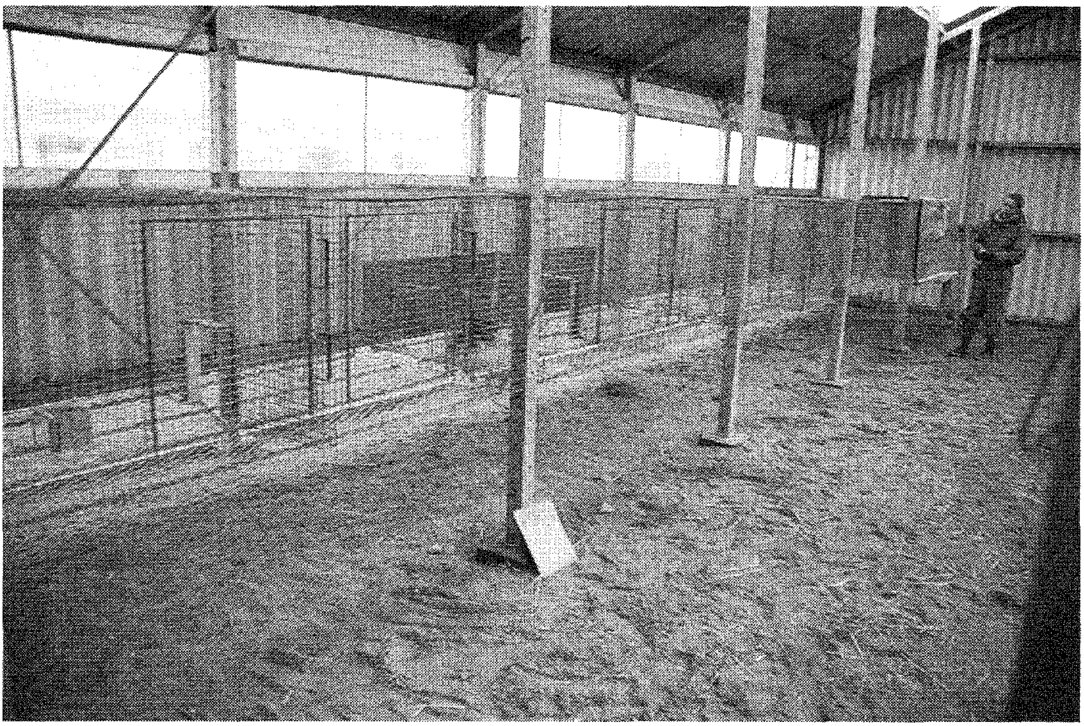
Cages for observation of behaviour under extensive conditions.

The influence of the production environment on the behaviour of farm mink

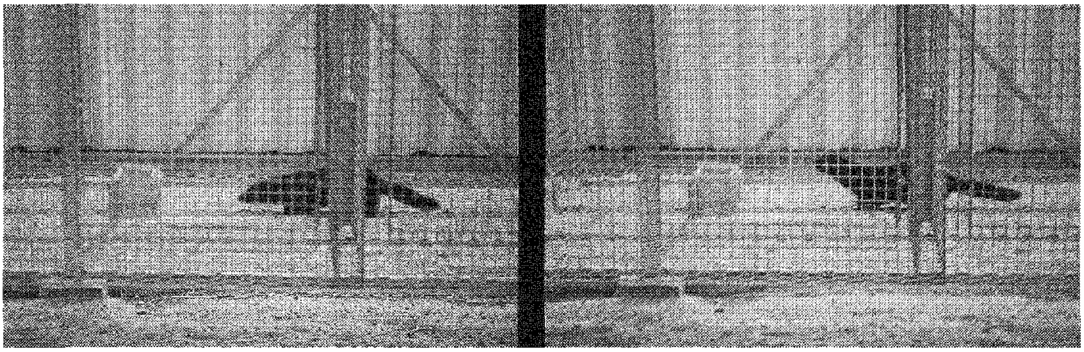
The work at the new section has in the ethological area been many-sided. An accumulation of basic knowledge included the development/adaptation of methods and tools for description of the normal behaviour of farm mink under production conditions.

The importance of the physical production environment to the behaviour and productivity of mink has been examined, partly in relation to cage sizes (from 0.1 m^2 to 1.0 m^2), and partly in relation to nest boxes (Hansen, 1988a; Hansen & Brandt, 1989; Hansen, 1990a; Hansen & Damgaard, 1991a; Hansen et al., 1992). The results showed that cage size did not affect the stress level of farm mink. On the other hand, the existence of a nest box was an important factor in securing the welfare of mink. The nest box also had a beneficial effect on fur quality. The possibility of enriching the production cages by means of shelves and water trays was also examined (Hansen, 1989a; Hansen, 1990b; Hansen, 1990c). In general, mink choose a resting place above floor level when resting out in the cage (Hansen et al., 1992). In the nursing period, the shelves may function as a 'refuge' for the female, but only as long as the kits cannot get up there. Water trays have an occupational effect for the mink, just like for instance 'toys', and especially immediately after they have been installed in the cage. There is, however, no indication that the water trays are meeting any behavioural needs of the mink. That the kits could supplement their need for liquid by licking water from the female's wet pelt has not been observed.

The importance of the social environment has been examined by varying the time of weaning of the kits (Hansen, 1987a), by letting the kits grow up alone, in pairs – male and female kit together – or in groups (Hansen & Damgaard, 1991b; Hansen, 1990d). The latter investigations furthermore provided the possibility



Open field is used to test reactions in new environment.



Mink approaching the object in open field.

of illustrating the flexibility of farm mink, partly to establish territories, and partly to adapt to social dominance hierarchies (Houbak, 1990 a and b; Hansen, 1990e). Despite the solitary life of farm mink in nature, the results show that placing in pairs or even in groups involves certain advantages for the mink in the insufficiently stimulated production environment. The frequency of pelt bites is, however, increased when the mink are kept in groups. The feed intake and circadian rhythm of the farm mink at ad libitum feeding have been examined (Hansen et al., 1992; Hansen et al., 1993) and discussed in relation to corresponding investigations at restrictive feeding. At ad libitum feeding, the mink adjusts its activity rhythm to the light-dark rhythm. At restrictive feeding the activity rhythm is synchronized to feeding time. The occurrence of stereotyped behaviour is under production conditions a common form of activity, which is more connected to the activity rhythm than to the actual feeding time. Stereotyped behaviour is disadvantageous in relation to feed economy, as this behaviour increased feed consumption considerably.

Besides illustrating the 'normal' behaviour of the farm mink, the investigations mentioned have also included data relevant to production such as growth, reproduction and fur properties (incl. pelt bites).

As part of a larger examination concerning production factors and production control, the variation between farms in the behavioural and physiological properties of mink has been studied (Møller et al., 1991; Hansen & Møller, 1988). On the farms involved, also the effect of handling routines on the behaviour of mink kits was examined (Møller & Hansen, 1988). These examinations led to more detailed investigations at the department's research farm in Foulum regarding the effect of 'pleasant' and 'unpleasant' handling of mink kits in their sensitive period (Houbak, 1991).

Topics in relation to the debate on welfare

The debate regarding the welfare of our domestic animals has resulted in increased efforts in two areas important to the continued, serious debate regarding the welfare of farm mink.

The first area was domestication of farm mink. This was based on the assertion that because of their short time as domestic animals, farm mink were still to be regarded as wild animals. The second area was determined by the necessity of developing and demonstrating applicable, objective stress indicators for evaluation of the welfare of farm mink.

Domestication of farm mink

During the last six years, a selection experiment has been running, where mink are selected for breeding according to their reaction to human contact. A behavioural test has been developed as a special tool in the selection (Hansen, 1987b; Hansen, 1989b; Hansen & Møller, 1989; Hansen, 1991a). This test is also used in corresponding experiments with farm foxes in Denmark and Norway. Regular use of the test has proved a seasonal variation in the temperament of farm mink (Hansen, 1992a). An aggressive behaviour response is in kits first seen in the month of December and may be caused by a beginning sexual maturity or be a consequence of the restrictive feeding introduced in this period. It has therefore not been possible by means of this test to select individuals with an increased tendency towards aggressive behaviour before the pelting period. On the other hand, we have succeeded to select two lines – a 'timid' line and a 'tame' line – which react to human contact by running away from or by approaching human beings, respectively. Timid behaviour in our domestic animals is regarded as undesirable and may often be an indication of reduced welfare. The preliminary results from the selection experiment show that the reproduction result is reduced in the timid individuals compared to the production result of the 'tame' individuals (in preparation). For both lines the test shows that the aggression of the males increases in the mating period and that the aggression of the females increases in the nursing period.

That the temperament of farm mink is primarily a result of genetic and/or prenatal influences was shown by switching 1-2 days old kits between selection lines. At a later behavioural test, the foster kits reacted like their biological mothers and do therefore not develop the characteristic behaviour of the foster mother (Hansen, 1991b).

It is a reasonable assumption that in the 80 generations of domesticated farm mink, an unconscious selection towards a higher adaptation to production conditions has taken place. Reduced reproduction because of poor adaptation to captivity was in the first generations an important factor. Besides, it has been normal farm practice to 'pelt' individuals with deviant behaviour, such as extremely timid or screaming individuals. This selection has had the result that most of the farm mink today react to human contact with curiosity rather than with fear.

The constant production conditions, to which the farm mink has been subjected, has contributed to this domestication. That farm mink – like other domestic animals – still possess the same behavioural properties as their conspecifics in nature does not mean that they are not domesticated, as the threshold values for release of several behavioural elements have changed considerably.

The early stages of the domestication process

To be able to illustrate the early stages of the domestication process, the section started in 1988 investigations of beech marten *(Martes foina)* caught in the wild and kept under extensive captive conditions.

The following areas of the biology of the beech marten are being/have been examined: circadian and annual rhythm in activity pattern (Hansen, 1992b; Hansen, 1992d), weight development, feed consumption (Hansen, 1993b), defecation pattern, reproduction cycle, development of kits, mother-kit relations, behavioural and stress physiological patterns of adaptation (Hansen & Damgaard, 1993). It will be attempted to compare the results obtained partly with beech marten born in nature and beech marten born in captivity, and partly with corresponding investigations concerning farm mink. Hematological, enzymological, and clinical-chemical variables have been examined for beech marten and compared with other mustelidae (Damgaard & Hansen, 1992).

Stress indicators

Another important area, partly because of the debate on welfare, was the development/demonstration of applicable objective stress indicators (Hansen & Jørgensen, 1989; Hansen, 1989c; Hansen, 1989d; Hansen, 1993a). An estimation of the validity of new as well as old stress indicators has been a central area for the section in evaluation of the influence of production environment and production routines on the welfare of farm mink (Hansen, 1992c). Existing knowledge of the independence of stereotyped behaviour of the originally releasing stimuli means that stereotyped behaviour is rather the expression of an active type of adaptation than an indication of reduced welfare (Hansen & Damgaard, 1993). That some individuals perform stereotyped behaviour can therefore not be taken as an indication of reduced welfare in relation to individuals not performing stereotyped behaviour. The applicability of eosinophil leucocytes as a parameter in the evaluation of welfare has proved to be insufficient, as the level can be both increased and reduced in cases of longerlasting stress depending on the strength and character of the stimulus (Hansen & Damgaard, 1991a). An evaluation of welfare should be based on several mutually supplementing behavioural and physiological stress indicators with due consideration for the individual pattern of adaptation (Hansen, 1992c).

Future perspectives

The experiments with selection for behavioural properties in the farm mink mentioned before will be continued. In 1992 an unselected control line was added to the design. The result will be used for estimating the heritability of the properties regarding behaviour and production. The physiological changes after a long time of selection will be described and related to the stress response of the animals.

The variation between the stress response of individuals has proved to be dependent on the individual type of adaptation. A correct interpretation of the physiological and behavioural stress indicators therefore requires knowledge of the way in which the individual adapts to the production environment. It is the intention to characterize – behaviourally, physiologically and productionwise – individuals in relation to the two types of adaptation – an active and a passive type – which have previously been described (Hansen & Damgaard, 1993). The type of adaptation used by the individual depends on a number of factors such as genetic conditions, age, sex, and the experience of the individual. It is therefore not surprising that even among siblings a large variation is found in productivity and in stress response at the same amount of stress. The objective of the coming investigations is to be able to point out one of the two types as being the best as regards production capacity and welfare under the actual production conditions. Furthermore, by means of behavioural and/or physiological characteristics of the preferred type to be able to select the best production animals for next year's breeders. Preliminary results indicate the possibility of reducing the often far too high percentage of barren females at the Danish mink farms by means of these investigations (Hansen, 1990d).

The section has several times taken the initiative to examine the practical problem 'pelt biting'. Each year pelt biting causes a considerable reduction in the earnings of the trade. The possibility of illustrating the problem by means of skin statistics, seems to be insufficient, as these statistics also include damages occurred at pelting. It must be vital for a solution of the problem to verify who is causing the 'pelt biting', the animal itself or the other animal in the cage, or both. Furthermore, a description of the time when the problem occurs/develops in relation to other forms of behaviour is essential. These tasks are purely ethological. Most likely several factors influence the occurrence of pelt biting. Genetics, production routines, and feeding have been mentioned in this connection. At the Department for Research in Fur Animals there is a close cooperation between the sections working within these areas. If the trade finds the problem sufficiently important and worthy of economic support, it is possible to study the problem from several angles and thus maximize the possibility of an early reduction of the problem. The selection lines at Research Farm South, where selection is performed for individuals which have been kept together with individuals with pelt bites, could give valuable material as regards experimental animals.

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Research in Housing Conditions and Welfare of Farmed Foxes

Vivi Pedersen

Introduction

During the eighties, it was considered a task of high priority to study the housing conditions, behaviour and welfare of farmed foxes. The initiative to commence an inter Nordic project 'Improval of cage- and nest box systems for farmed foxes' was taken by Gunnar Jørgensen at the National Institute of Animal Science and Hans Edvard Teglers from the International Fur Trade Federation (I.F.T.F.). In 1987 the cooperation between researchers from Norway (Bjarne Braastad, Morten Bakken), Finland (Mikko Harri) and Denmark (Leif Lau Jeppesen, Vivi Pedersen) started. The main conclusions of the Norwegian and Finnish research are described below. In Denmark the main object was to examine the use of different whole-year shelters and to reveal what impact whole-year shelters had on foxes' behaviour and welfare.

Pilot studies were carried out at the National Institute of Animal Science, Trollesminde, Hilleroed, under the supervision of Gunnar Jørgensen. Experiences gained in these pilot studies were used to make studies in a larger scale at the Research Farm North, Loekken. The Danish Fur Breeders Association and I.F.T.F provided a fox farm and a fox keeper.

Research in Norway

The research was concentrated on infanticide and related problems in fox farming: 70-80% of cub mortality with infanticidal vixens was caused of bites due to the mother. In 50% of infanticidal occurrences the vixens started by biting off the cubs' tail. The cub-killers rested less inside their breeding box compared to non-killers. No indication was found that disturbances from humans could provoke cub-killing and cubs were not killed more frequently during the day time (Braastad & Bakken, 1993). Tunnel entrances in the breeding box reduced the occurrence of infanticide and increased the mean litter size at weaning (Braastad, 1990 and 1991). Tunnel entrances also made vixens more relaxed during working hours (Braastad, 1993). M. Bakken studied the utilization of individual variation in behaviour among farm-foxes and found that it was possible to predict a female cub's later reproductive performance by behavioural testing at the age of 30 days (Bakken, 1989 and 1993a). A correlation between competition capacity, reproduction and timidity was found showing that vixens with high competition capacity were also less timid and reproduced better compared to vixens with a low competition capacity. The relationship between neighbouring vixens showed to have great impact on the reproductive performance (Bakken, 1993b and c).

Research in Finland

In Finland the main object was studies of the physical parameters of different designs of whole-year shelters: Newborn mortality was highest at the time of birth and the following 2-3 days. The heat produced by the vixen only increased the air and wall temperatures in the nest box by 1-2°C (Harri et al., 1988). By huddling the cooling rate halved. The overall conclusions were that newborn foxes were not able to maintain homeothermy under farm conditions. They needed an external heat such as their mother. The nest boxes should be designed in such a way that the vixens choose to stay inside the box for longer periods. This is more crucial for the survival of her newborn cubs than providing isolation etc. (Harri et al., 1991a).

Blue foxes spent an average of 6.8% of their daily time on platforms. The platforms were more often used at temperatures above 0°C than at lower temperatures. High wind combined with high temperature promoted the use of platforms (Harri et al., 1991b). The conclusions were that platforms were not used as protection against bad weather. Inter-individual variation in preference for platforms was high. Silver foxes used platforms as a place for observation and used the platforms less frequently during weekends than during working days. The familiarity with the platform in the cage environment was not a significant factor affecting its use (Harri et al., 1992).

Research in Denmark

Housing conditions

From November 1987 to December 1989, 50 silver and 50 blue fox vixens were kept with 3 different designs of whole-year nest boxes and a shelf. All vixens had free access to all 4 types of shelters. 50 vixens of each species were kept in barren wire-mesh cages as control animals.

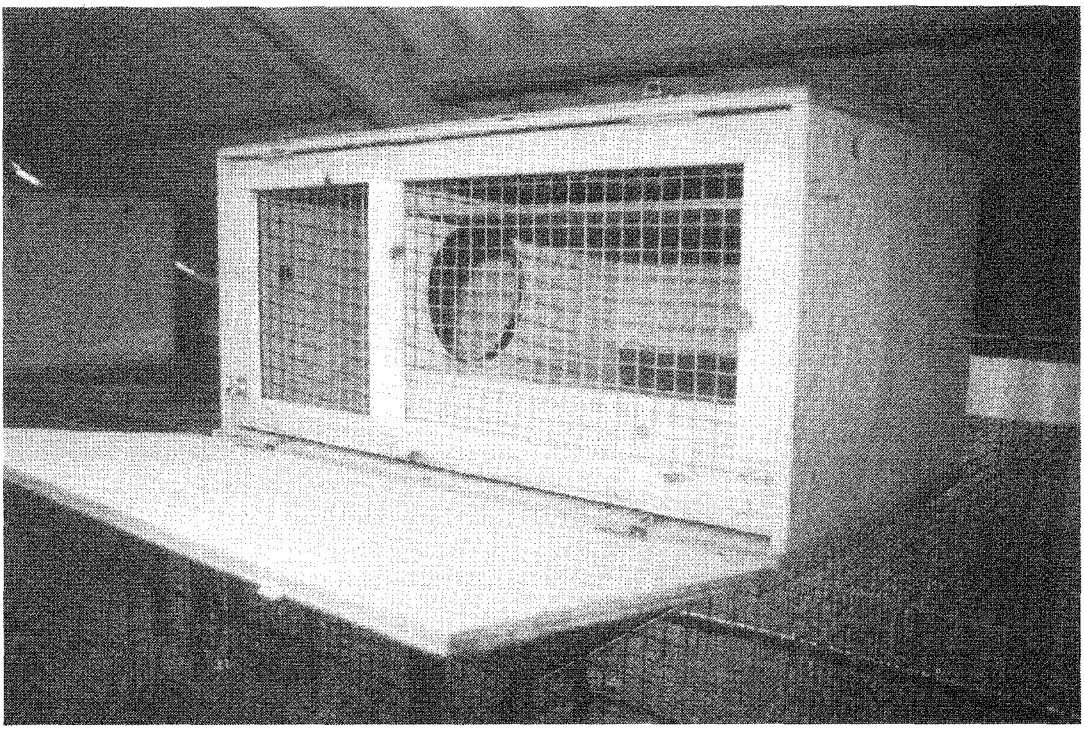
In the first 2 years of the study the foxes' use of the shelters was registered regularly with scan samplings of 10 minutes intervals. Faeces from all shelters were removed and weighed every second week. The results revealed a preference for the shelf and the top box in both species. Blue foxes used the top box sligthly more than the silver foxes and in all, blue foxes used shelters twice as frequently as silver foxes. During disturbances most blue foxes fled into the top box or the side box whereas silver foxes fled to the opposite side of the cage and some fled into the top box. It was concluded that farmed foxes use whole-year shelters when provided with them, and that different shelters were used to a different extent. Differences in use between individuals, between species, and during the year, may have reflected different sensitivities to disturbances or different strategies between species (Jeppesen & Pedersen, 1990; Pedersen & Jeppesen, 1993). Defecation mainly took place on the cage floor near the feeding site, or in the open box. The other shelters were not soiled to the same degree, and in the end of the study defecation in the top and side box was minimal, ranging between 0-5 grams per 14 days (Pedersen & Jeppesen, 1992).

In October and November 1989 the foxes were exposed to various tests in order to evaluate behavioural and physiological responses to an acute stressor. First their responses to different test situations in the cage were registered. Then the foxes were exposed to an open field runway and blood samples were taken just prior to and after the open field exposure. The results revealed that compared to controls the foxes kept with whole-year shelters (experimental animals) were more defensive and less fearful in the in-cage behavioural tests and the experimental animals more vigorously resisted being caught. In the open field runway the experimental animals showed more field crossings than control animals, they were faster to proceed the runway in general, and had lower latencies to reach the end of the runway compared to control animals. The experimental animals also showed lower initial levels of cortisol and lower levels of eosinophil leucocytes throughout. The conclusion of these results was that whole-year shelters had a beneficial effect on the foxes' everyday well-being (Jeppesen & Pedersen, 1991; Jeppesen & Pedersen, 1992).

Early experience

The possibility of foxes to escape from disturbing stimuli may cause failure to habituate to the farm situation, and for that reason the top box was developed in such a way that foxes could be exposed to the farm situation, also when they were inside the box. The new top box was also made more convenient for the farmer. As the fox farm at the National Institute of Animal Science moved from Hilleroed to Foulum these newly developed top boxes, together with shelves, became standard equipment for all adult foxes in the new farm (Photo 1).

At Foulum a study was commenced in 1990 to examine if early experience with the farm environment affected the foxes' later behavioural responses towards different stimuli. Litters, which were unharmed at 2 weeks of age, were divided into 2 groups. In the experimental group the solid front door of the top box was removed (see photo). Behind the solid door was a



Top box used for early experience.

wire mesh door which prevented the vixen and her cubs from escaping. Behavioural testing was performed at the age of 12-16 weeks and again at 23-28 weeks of age by a person unknown to the foxes. The results revealed that foxes reared in open top boxes showed lower levels of fear towards the person compared to control animals reared in closed top boxes (Fig. 1) (Pedersen, 1991). Similar results have been obtained by Clark & Galef (1977) who worked with Mongolian gerbils reared either in laboratory cages or cages with tunnels and shelters.

At present, all adult vixens at Foulum are divided into two groups: one in which the vixens are kept with an open top box throughout the year and a second group, in which the vixens are kept with a closed top box. Twice a year all vixens and their cubs are tested for behavioural responses towards humans and other stimuli. This way we will experience if the possibility to hide, when disturbed, has a greater impact on the fear behaviour of foxes compared with the possibility to escape into a 'safe' but open top box.

Early handling

Research concerning effects of handling on behaviour, reproduction, physiology, and stress sensitivity in foxes has been performed during the last 5 years in addition to research concerning housing conditions. The main object was to find some reliable methods to achieve less fearful and less stress-sensitive foxes in the farm. Different handling procedures, age intervals, duration and intensity have been studied and the results and conclusions have been (or will be) published in international journals. In this contribution only a short summary will be given:

Gentle handling of foxes at the age of 2 to 8 weeks showed to reduce their later fear responses and increase exploratory behaviour towards both known and unknown humans. Handled foxes also showed a higher degree of exploration in an open field runway and less fear responses during capture in the cage. The physiological parameters (cortisol, eosinophil leucocytes) indicated long term stress in control

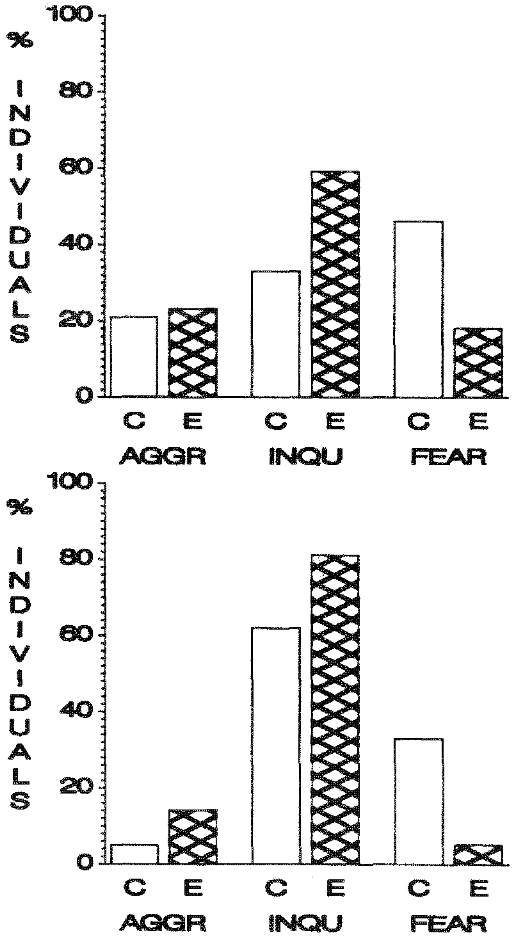


Fig. 1. Results from the behavioural test carried out at the foxes' age of 12-16 weeks. Percentage of control (C) and experimental (E, with open boxes) animals (silver foxes above; blue foxes, below) showing aggressive, curious or fear responses when confronted with an unknown human being. Differences are significant at the P < 0.05 level for silver foxes and at the P < 0.001 level for blue foxes, $\chi 2$ test, two-tailed.

animals compared to handled animals. Different processes and terms such as imprinting, early experience and habituation were discussed (Pedersen & Jeppesen, 1990).

To reveal more about the sensitive period of primary socialization in foxes and to maximize the handling effects, it was examined at what age gentle handling had the maximum effect on the later behaviour of the foxes (Pedersen, 1992). 344 silver fox cubs were randomly distributed in 8 different groups and received gentle handling at different age intervals and with different duration. During their juvenile phase the foxes were exposed to different behavioural tests and once, at the age of 26 weeks, blood samples were collected. The most pronounced reduction of fear responses was found in cubs handled during weaning (6-9 weeks, 6-12 weeks of age) or post-weaning (9-12 weeks). The conclusion of the study was that handling for three consecutive weeks during and post-weaning was sufficient to reduce the later fear responses towards humans. Handling pre-weaning only reduced fearfulness when carried out for six consecutive weeks. Frequent and repeated contact with humans could be a way of achieving less fearful foxes and thus foxes which are more adapted to the farm environment. Similar results have been achieved with rabbits (Kersten et al., 1989) and pigs (Hemsworth et al., 1986).

In the above studies, the handling procedure was gentle, stroking and fondling the animals, but it was experienced that some foxes reacted with fear or defensive aggression during the handling sessions. Great emphasis was put on making the handling either a very positive or negative experience for the fox in the next study. The gentle, unforced handling included very slow movements and presentation of supposedly delicious feed items during the handling session. The forced handling procedure included abrupt, quick movements with capture, transportation, and confinement. The results of this study (Pedersen, 1993) showed that both handling procedures were efficient in reducing later fear responses of foxes towards humans, but the forced handling procedure also reduced the foxes fear responses towards unknown stimuli.

Ongoing studies concern the long term effects of the two different handling procedures on fear responses and stress sensitivity of the foxes. In addition, the effects of the different handling procedures on the reproduction of foxes, maternal behaviour, physiology and various production parameters are examined.

Different production conditions will be compared in the new research project.

Future perspectives

Results from the above mentioned studies have been used by the Council of Europe's Standing Committee for the Protection of Animals kept for Farming Purposes in their recommodations concerning fur animals. Still there is a great public interest in the research concerning housing conditions and welfare of farmed foxes, and the Ministry of Agriculture has now funded a project concerning alternative housing conditions for farmed foxes. In this project (1993-96) the traditional housing systems will be compared with a system using the results from the preceeding years of fox research, and with an alternative system providing the foxes with more space and equipment and a solid floor. The comparisons of the 3 systems will be on several levels: reproduction, behaviour, health, welfare, fur qualities, costs of production etc. This project is conducted by Leif Lau Jeppesen and Vivi Pedersen at Research Farm North. At Foulum continuous research of the foxes' habituation to the farm environment through the new top boxes will be conducted by Vivi Pedersen in cooperation with Outi Lohi.

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Production Systems and Management

Steen Møller

Introduction

Decisions regarding housing systems and management pratices have always been based on the experience and intuition of the individual mink farmer. In the early stages, this also applied to feed composition and health control. As the advisory service was established, help was first received in health control and feed plans. In the 1960's the responsibility of composition and quality of feed was handed over to central feed kitchens (FK) making and delivering ready mixed feed for many farms. In the late 1960's systematic feed control was established.

Reproduction and feeding have been subjected to several investigations, but the entire production system and management are studied very little. Thus, many questions about everyday management on the mink farm are still unsolved, and the practical implementation of scientific results is usually left to the advisers and farmers.

In the late 1970's, Gunnar Jørgensen saw that a more efficient production should be built not only on experimental results. How the alredy established sum of knowledge was implemented to improve the production on private farms was just as important. To investigate this, focus should be put on the production on private farms, and the reasons for the large variation between their results.

The use of different production figures as a guideline in planning and production control was little used and depending on the individual adviser. In the late 1970's and early 1980's, the Danish Fur Breeders Association started collecting information of losses caused by deaths, nutrient composition of feed and feed consumption, and pelt production to central data bases.

In the early 1980's, the use of this kind of information was discussed between The Danish Fur Breeders Association (DFBA) and the National Institute of Animal Science, and a survey of the total production line was planned in order to build up a model for improving the production systems.

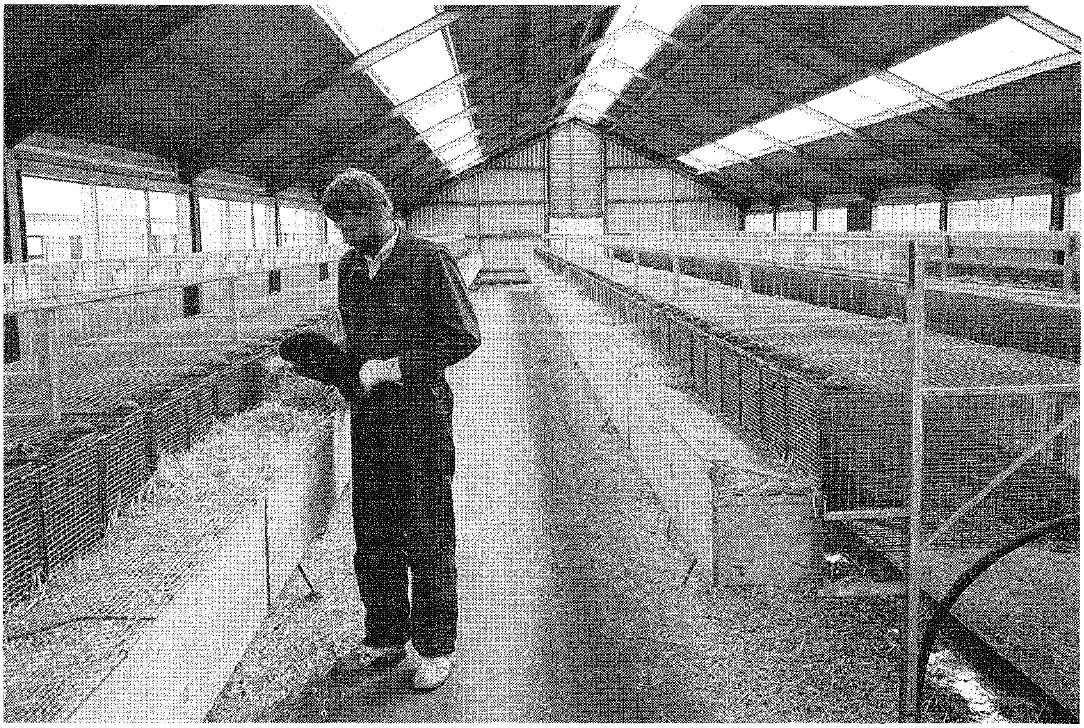
The project "Production Factors and Production Control" was started in 1985 and was carried out in cooperation with DFBA, local advisers, feed kitchens, and farmers. To cover most of the variation, 4 FK and 23 farms were selected from different parts of the country representing farms of different size and quality. Most research disciplines were involved, and research in ethology and in production systems and management was initiated in this project. In 1989, the results were incorporated in the formulation of a detailed production guide for mink farmers. This was done in fruitful cooperation with the Fur Breeders Association of Mid-Jutland, and the guidelines were tested on 6 private mink farms in the "Demonstration Farm Project".

These projects resulted in a lot of information about the present production systems and initiated several research experiments. In the following results from some of the main areas within the projects are presented and discussed.

Mink production systems

The production system consists of labour, buildings, machinery, and animals. In the project "Production Factors and Production Control", most focus was put on housing conditions. The size of the project farms varied from 125 to 2800 females, while manpower varied from 0.5 to 4. Each man tended 533 ± 169 female mink. The first man tended approx. 400 females, while each extra man tended approx. 650 females. This increase in efficiency with number of females was due to large-scale operation, rather than to degree of mechanisation.

The design of sheds and cages was limited to a few standards. The reason is that most sheds and cages in Denmark are pre-fabricated in sec-



Multi-row sheds are common in Denmark.

tions of 2 meters containing a battery of 6 breeding pens or 8 furring pens. The sheds were usually with 2 or 6 rows of cages, but up till 11 rows were seen.

On the 23 private farms, differences were found in whelping results between two-row and multi-row sheds as shown i Table 1 (Møller 1992, a and b).

Though a higher kit loss in multi-row sheds has been reported by Jørgensen & Hansen (1970), the result was unexpected. The explanation may be differences in climate or light conditions in the sheds. To examine this, a method to measure the light conditions in the sheds with a normal photometer was developed. Light conditions were always good in two-row sheds, as light falls in through the sides. In spite of large variation in light conditions, no correlation to the reproduction result was found in multi-row sheds (Møller, 1991a). An experiment is planned to thoroughly examine the effect of shed type.

The design of nest boxes showed large variations in shape, size, and extra equipment such as drop-in bottoms and wind shields. The effect of nest box and drop-in bottom was examined experimentally during the nursing period of mink (Møller, 1990). No effect on litter size at birth or

Table 1. Reproduction of the scanblack type in two-row and multi-row sheds.

Rows	Farms	% barren females	Kits per litter at birth	Kits per litter at 3 weeks	% lost kits
2	12	8.9 ± 2.9	5.5 ± 0.3	5.3 ± 0.3	3.6 ± 3.1
>2	10	12.8 ± 6.5	5.2 ± 0.3	5.0 ± 0.4	5.2 ± 2.6
		p = 0.10	p<0.05	p<0.05	p = 0.21

number of barren females was found. The better physically defined the nest was, the better the kits performed with regard to survival and weight gain in the first weeks after birth.

The explanation may be that the thermo regulation of the kits does not work properly before the age of 25 days. Younger kits go into a sort of cold rigor if they are not kept warm. A good nest box is important because the easier the kits get away from the nest, the more often this cold rigor will occur. The rigor reduces milk intake and gain and increases mortality, if the kits are not brought in by the female.

In spite of the variation between the private farms, the floor area of nest boxes used during the nursing period did not vary, and no differences in production results were found (Møller, 1992a).

Drinking water

The watering system on the private farms consisted of a plastic hose running along the backside of the cages. The mink got water by releasing a drinking valve. The valves were of different makes, but only details differed (Møller, 1992a). The farmers did much to ensure fresh water for the mink throughout the year. During lactation, extra arrangements were made to help the kits to find the water, and to keep the water fresh. In the winter, water was circulated or heated to keep it from freezing. In spite of these efforts, not much was known about the effect of different watering systems or the drinking behaviour of the mink. Though no effect of the different watering systems on the private farmes was found, a series of experiments has been carried out, to reveal the drinking behaviour and the effect of watering systems (Møller, 1988b, 1991b and 1993b; Møller & Lohi, 1989).

Supplementary watering systems

In the latter part of the lactation period, a supplement to the ordinary supply of drinking water was often used. A drip watering system has been tested throughout two lactation periods (Møller & Lohi, 1989).

The supplementary watering system only had effect when the ambient temperature during the day reached above approx. 25°C. In the warm and dry lactation period of the second year, the weight loss of the females was reduced, and the weight gain until 7 weeks was higher for kits in the group with drip watering system. Kits with drip water supply took in water earlier than the control group, by licking water from the tongue of the valve. However, they did not learn to release the valve earlier (Møller & Lohi, 1989).

In spite of the variation between the watering systems on the farms, they all provided water for the kits during the nursing period, and no differences in production results were found (Møller, 1992a).

Mink kits find the water supply and start to drink as their activity in the pen increases at the age of 5 to 6 weeks. As they start to eat at the age of 4 weeks, there is a difference of approx. 2 weeks from the time the kits begin to eat and to drink (Møller & Lohi, 1989). This interval was surprising, as the general opinion was that the kits start to eat and drink at the age of 3 to 4 weeks. This interval in time has been confirmed (Møller, 1991a). In this 2 week period, the kits often lick saliva from the female's mouth. Saliva licking occurred most frequently in the group without drip water supply and at high ambient temperatures.

An experiment with a spray watering system has confirmed that supplementary water affects the weight gain of the kits (Møller, 1993b). It seems that any source of available water may help the kits to drink, until they are old enough to release the drinking valve.

Drinking water temperature

The temperature of the drinking water offered to mink varies from 0°C in the winter to more than 45°C in the summer. The temperature preference of mink has been investigated comparing the intake of 40°C warm water to water from the tap (6-17°C) (Møller, 1988b and 1991b).

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. With only one temperature at a time, no difference in the intake pattern was seen.

An adult male mink drinks about 25–30 times a day, from a nipple type watering system. In each drinking session between 2.5 and 5 ml of water is ingested depending on water temperature. In all cases a higher quantity of cold water was spilled, both totally and per drinking session (Møller, 1988b).

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 (Møller, 1991b).

It was concluded that mink do not refrain from drinking 40°C hot water, but the drinking behaviour differs between warm and cold water. Waste of water is in all cases significantly higher for cold water than for warm water.

The variations in design of nest boxes and watering systems were correlated to the region probably due to the location of the equipment producers as well as a regional or advisory effect. These differences indicate that a variation in production results between farms using feed from the same FK, may be due to differences in production systems (Møller, 1992a).

The variation in production systems indicates that the basic needs for housing of mink can be met in many different ways.

All ordinary mink cages complied with the recommended guidelines regarding size. Recommendations as regards nest boxes, screening, feeding, and water supply were met on all farms.

Management

Management on the project farms during pregnancy, birth, lactation, and weaning differed only in detail, and no significant effects were found. Large variations between farms were found in mating routines and selection of breeders, but only few effects on production results were demonstrated.

Placing of animals

Some variation was found in the placing of animals on the private farms. In the reproduction period males and females were either caged in separate groups, or in series of 1 male and 5 females. Whelping results were better where the breeding animals were mixed than where males and females were placed in separate groups, as shown in Table 2 (Møller, 1992a and b).

A logical explanation would be olfactory stimulation from the males, but male urine sprayed on the females' pens prior to mating had no effect (Møller, 1991a), neither had odour from the anal sac of males (Therkildsen, 1991). As the placing of animals was related to FK, other factors may influence the results. An experimental clarification of the effect of placing of animals for breeding is planned.

None of the farmers separated the females by an empty cage or a cage filled with straw during the breeding and whelping period, though previous experiments have shown good results. Therefore, the effect of separating the females in the mating and whelping period was investigated. The separated females were less active than the control group throughout the lactation period. Neither whelping results nor weight gain of the kits were influenced by the separation or the difference in female activity (Møller, 1991a).

Table 2. Reproduction results of scanblack for breeding animals caged in groups by sex or mixed for mating.

Placement	Farms	% barren females	Kits per litter at birth	Kits per litter at 3 weeks	% lost kits
Groups	11	12.5 ± 4.9	5.2 ± 0.3	5.0 ± 0.3	5.8 ± 2.7
Mixed	11	10.1 ± 4.4	5.6 ± 0.3	5.4 ± 0.3	3.8 ± 2.4
		$p = 0.25$	$p<0.05$	$p<0.05$	$p = 0.11$

After weaning the majority of kits were placed in pairs of one male and one female on the private farms. An old female with a male kit was also common (Møller, 1992a). Placing mink kits individually from September till pelting resulted in a lower weight at pelting (Møller, 1991c). This could be expected to improve the pelt quality, but the results rather showed a declining tendency. Most fur bites arose from November till pelting – for animals living alone as well as in pairs. The number and degree of fur bites were reduced in the separated group, but no sexual difference in the frequency of bites was seen. The difference in number of fur bites is likely to be caused by the partner. The benefit of the reduction in pelt bites could not compensate for the cost of reduced skin length in kits housed alone.

The differences in mating routines and selection of bredeers were correlated to the feed kitchens, indicating a regional or advisory effect. Thus, a variation in production results between farms using feed from the same FK, may be due to differences in management.

The level of pelt bites varied between farms, but also between years. In a year with low temperature in November, few pelt bites occurred, which indicates that climate is the common environmental effect within years.

The main impression from the project was that many different management practices may work perfectly. The important thing is that the farmer is skilled at what he is doing, and that he is confident with his management practices (Møller, 1992a and b).

In the "Demonstration Farm Project" the production on 6 private farms closely followed a common guideline. The farmers were often visited by the advisers, who also helped in the selection of breeding animals. The preliminary results showed a reduction in the feed cost by 15 %, while improvement of other production results were insignificant. The participants felt that their skill as mink farmers was improved, but noticeable results in the production have not yet been seen. The final results are not published yet.

The combination of data from many different sources in the calculations of farm performance was not without difficulties. Most data were gathered for some specific purpose, which they may serve well, but care should be taken when data are used for another purpose. For instance, pelt grading at the fur auction is done in order to get uniform sales lots. However, different kinds of damaged skins fall into the same category of 'bite marks', which is therefore not an exact measure for how many animals were subject to bites during production. A thorough knowledge about the auction grading system is therefore necessary, when auction data are used to evaluate the skin production on different farms. The project showed, that with proper knowledge about the data from external sources, they can give valuable information for production control.

Weight gain and feeding practice

One of the characteristics of mink production is that the product can be evaluated only at the end of production. In the early stages of production, only indirect measures, such as weight, can predict something about the final skin properties, i.e. the skin size. By following the growth curves of the animals, the farmer can control the feeding and management in general. Therefore, the mink were weighed regularly on the 23 private farms, and the weighing program was evaluated as a tool for control of the production.

All farmers fed ad libitum during the lactation period in order to get the kits as large as possible before weaning. The growth curve was followed by weighing ten litters of 4-9 kits every 10 days during the lactation period on each farm. The birth weight was set at 10 g for males and 9 g for females, and the following functions were fitted for mink kit growth:

Male kits:

$$\text{weight} = \underset{\pm 0.7}{10.1} + \underset{\pm 0.41}{0.95}\times\text{age} + \underset{\pm 0.01}{0.20}\times\text{age}^2 \qquad R^2 = 0.98$$

Female kits:

$$\text{weight} = \underset{\pm 0.5}{9.0} + \underset{\pm 0.28}{1.80}\times\text{age} + \underset{\pm 0.01}{0.14}\times\text{age}^2 \qquad R^2 = 0.99$$

where

weight = body weight in g
age = age in days

The growth functions give a good description of the growth from birth till weaning, and can be used as standard curves for scanblack mink kits until weaning.

Though litter size was restricted, the weight of kits at the age of 31 days depended on litter size as well as on farm. It is therefore important to select homogenous groups for weighing and to weigh at a certain age. A difference between years indicated a common environmental effect.

The early development of the kits gives a good impression of their final weight and of the skin size, and therefore a high weight at weaning is desirable in practice. However, a considerable part of the growth potential is linked to litter size. The effect of kit growth during the lactation period seems to be related to protein deposition (Glem-Hansen, 1980). Other reasons for variation in weight at weaning can often be compensated for after weaning. It is therefore more important to meet the protein requirement of the animals than to feed for maximum weight at weaning (Møller, 1992a).

All farmers continued to feed ad libitum during the summer. In order to avoid welfare disease, feeding was restricted for the period from August to October. The growth curve was followed by weighing 25 pairs of kits from each farm every 2 weeks from weaning till pelting for 3 years. The following growth functions were fitted from weaning till pelting:

Male kits:

$$\text{weight} = \underset{\pm 60}{-701} + \underset{\pm 1}{30}\times\text{age} - \underset{\pm 0.004}{0.077}\times\text{age}^2 \qquad R^2 = 0.95$$

Female kits:

$$\text{weight} = \underset{\pm 33}{103} + \underset{\pm 0.6}{11}\times\text{age} - \underset{\pm 0.002}{0.025}\times\text{age}^2 \qquad R^2 = 0.95$$

where

weight = body weight in g
age = age in days

The functions give a good description of the weight development, and can be used as standard curve for scanblack mink kits after weaning.

As regards weight development, skin length, skin quality, and feed intake differences were found between farms and, with the exception of feed intake, also between years. A difference between years indicates a common environmental effect, e.g. the weather. Some farms had almost the same growth curve each year, while others had large fluctuations between years.

On all farms and at all times of weighing, the standard deviation amounted to approx. 10% of the weight. There was a close relation between the weight of males and females. The relation was constant over years, indicating that only one sex needs to be weighed to draw the general growth curve of the animals.

The amount of energy fed varied with up to 43 kcal/animal/day corresponding to 4-5 kg of feed per skin produced.

In the total Danish production a positive correlation was found between average skin length and average pelt quality of individual farms. A grouping of the project farms according to skin length and quality showed no difference in weight nor in daily gain. Feed intake was correlated to skin length and quality. The weight in August and in October was correlated to skin length, but only the weight in October had negative influence on pelt quality. The reason may be a difference in body length resulting in less fat animals and thus a better quality at the same weight (Møller, 1992a).

The differences between the mink strains on the farms must be a result of different selection in regard to size and quality. An ideal growth curve must take into consideration the body length and the quality of the animals on the individual farm as well as an effect of year.

The correlation between the size of the mink and the size and quality of the pelt was also investigated experimentally (Møller, 1988a and 1991a). The length of the skin is correlated to the body length and the fatness of the mink, but the fatness is negativly correlated to pelt quality. As weight is a combination of both length and fatness it is highly correlated to skin size, but also to skin quality. As the quality grading

Close contact to the researcher.

of live animals is difficult, many farmers may benefit from selection for body length rather then for weight, in order to get longer skins without a drop in pelt quality (Møller, 1992a and c).

The breeding animals lost weight quickly just after the pelting season, irrespective of the farmer's strategy regarding weight development. There was a negative correlation between the mean temperature from January to March and mortality on a national basis. No fixed weight limits can be given, but a weight reduction of more than 30%, or to less than an average of 900 g, seemed unfavourable. As first year females are especially sensitive, whereas the weight of the males is of minor importance, it would be most efficient to weigh first year females.

Behavioural response to weighing

Some farmers found that the animals in the weighing group became more difficult to handle. The influence of the weighings on the behaviour of the animals was measured by the stick test. A stick was thrust into the cage and the reaction of the mink was noted. In general, the females reacted more timidly than the males. On most farms the weighed animals reacted more timidly than the control group, but on a few farms they were less timid than the controls. Thus it seems that the reaction to weighing expresses the minks' experience with handling (Møller & Hansen, 1988; Møller, 1991a).

A weighing programme

Based on the many weighings, a programme for control weighing of mink in the nursing period, growth period, and in the winter period is suggested. The purpose of the weighings, the need for selection and size of groups to be weighed, and times for weighing as well as the application of the weighing results are discussed (Møller, 1993a). Irrespective of farm size, a group of 25 mink from litters of 5-7 gives a good idea about the mean weight of the population.

Perspectives

The comparison between the production on private farms and experimental knowledge about production factors showed that well documented effects could rarely be demonstrated in practice, while other factors, not previously regarded as important, seemed to have effect. In practice, many factors influence the results, in contrast to experiments where only one factor is investigated. This indicates that the interactions between the many aspects of mink production in practice, i.e. housing, feeding, health and management in general, are just as important as the single factors. The interesting part is to find these interactions and improve the chance for the private farmer to get these factors right. The Demonstration Farm Project shows, that it is not enough to do the right things, they also have to be done right! In this context the farmers' qualifications and intuition seem to be crucial.

In the future, these interactions will be investigated through a systems analysis of the production periods. When the farm is regarded as a cybernetic system, the farm is organized as a production system and a management system as discussed by Sørensen and Kristensen (1993). The systems analysis shows how the farmer maintains the farm in harmony with the goals, by use of controllable factors. Corrections in management are often necessary to correct for the effect of disturbances by the uncontrollable factors, such as climate, price relations, and restrictions by society. Management is considered as a chronological series of: measurements of systems behaviour, comparisons with a goal, and adjustments in controllable factors. An important factor is the farmer's possibility to control the production. In order to do so, the lack of production control measurements is a problem, and one reason why the farmers' qualifications and intuition seem to be so important.

The long period between management decisions and the consequence for the production result means, that the aim of management focuses on the result of the actual period, though consequences reach into future periods. Therefore, development and implementation of control measures has high priority.

Further investigation of the unsolved questions that have emerged (e.g. type of shed, placing of breeders) would be natural steps forward. The best solution is to find the underlying mechanism, rather than investigate all possible factors. For instance, when the factors that govern ingestion and waste of drinking water are known, the effect of any type of watering system can be estimated. If the light conditions to secure a normal production were known, many problems and worries in practice could be avoided.

The common environmental effects, connected to the year, on production parameters (e.g. weight development, pelt properties) create a need for information about the production level of the year. The basis for this could be farm panels within each FK. Since the projects started, many farm panels for exchanging and discussing experiences from the mink production have been established.

The value of data from external sources can be improved if the production control aspect is included in the purpose of data collection.

The optimal feeding practice in relation to growth curves, pelt quality, feed efficiency, and lactation capacity also needs further investigation.

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