Genetic models for the inheritance of the silver colour mutation of foxes

FLEMMING SKJØTH[1]*, OUTI LOHI[2] AND ALUN THOMAS[3]

[1] *National Institute of Plant and Soil Science, Department of Biometry and Informatics, Research Centre Foulum, P.O. Box 23, DK-8830 Tjele, Denmark*
[2] *National Institute of Animal Science, Department for Small Farm Animals, Research Centre Foulum, P.O. Box 39, DK-8830 Tjele, Denmark*
[3] *School of Mathematical Sciences, University of Bath, Bath BA2 7AY, UK*

(*Received 22 November 1993 and in revised form 25 May 1994*)

Summary

We consider genetic models for the inheritance of the particular colour patterns of silver foxes. The models are evaluated by computation of statistical likelihoods based on observations of related foxes in extended pedigrees. Problems caused by incomplete paternity information are addressed by inferences based on phenotypic observations. The unreliability of subjective evaluations of fur colour also provides difficulty, in particular *crossfoxes* emerge as being difficult to differentiate. No evidence of linkage between Agouti locus and Extension locus is found in this dataset.

1. Introduction

Naturally occurring mutations producing variations in the fur colours of animals have long been of interest to geneticists. In the case of the Red Fox, *Vulpes vulpes*, mutations producing silver-coloured animals are of economic importance to fur farmers since unusual colours command higher prices. It is clear that alleles at at least two loci can affect this trait but there are different theories about the effect of the genes and the mode of inheritance. Warwick & Hanson (1937) proposed in 1932 a theory with two independent loci each with a recessive black colour allele. Iljina (1934) introduced to this theory a third recessive locus for cross marking. The mode of inheritance of genes for fur colour has been widely investigated for some animals, in particular mice (Silvers, 1979). Searle (1968) discussed the homology between colour loci of mice and other mammals.

The different types of foxes as well as two genetic models are described in Section 2. In Section 3 we describe the data collected on foxes in four Scandinavian fur farms. Animals within each group are related by large extended families, or pedigrees. Section 4 describes briefly the statistical elements that we use to model the genetic trait. This model leads to complex likelihood calculations that require considerable computational resources. An efficient

method for making these calculations is briefly mentioned. Also in Section 4 we describe an *ad hoc* solution to the problem of uncertain paternity in some litters. The results of the analysis are presented in Section 5 and our conclusions are given in Section 6.

2. Types of Silver foxes

(i) *Classification system*

The earliest known colour mutation of the wild Red fox is Silver fox, which is black with varying amount of silvery hairs (guard hairs with a shorter or longer silver bar in the middle of the hair). It soon became evident that silver foxes were not all genetically identical. Crossings between Red fox and Silver fox produced several types of cross foxes with variation in amount and distribution of red and black colour. Thus the fur colours of foxes range from red to black in a large number of nuances, making it hard to establish a classification system. Below a brief description is given of the different types, into which the foxes are mainly categorized. For a more precise description the reader is referred to Nes, Einarsson & Lohi (1988).

Red fox: The Red fox is generally red coloured with a more or less dark brown line on the spine, which becomes broader at the shoulders, forming a cross. The breast and lower part of the neck can be white or light grey. The underfur is grey with a reddish brown

* Corresponding author.

zone at the top. The belly is often white or the main red colour of the fur. The paws and the outer part of the ears are covered with dark brown and black hairs. The tip of the tail is, with very few exceptions, white.

Gold fox: This type differs from the Red fox in having black or dark grey ventral spots. The legs and the ears are black.

Gold Cross fox: This fox is generally darker than the Gold fox. On the dorsal part red colour still dominates but the line along the spine and across the shoulders is black thus forming a clear dorsal cross. The white spots on the belly are changed to black. The nose, throat, legs and tail are black.

Silver Cross fox: This type is again darker than the Gold Cross fox. The red colour is seen mainly just above and below the shoulder cross. The shade is more brownish than yellow. The darkest individuals have a few red hairs on the shoulders and inside the ears.

Silver fox: This term covers several types of Silver foxes all being generally of black colour with a white tip of the tail. There are two types on which most breeding has been based. Alaskan Silver has grey/brown hairs on the ears and sometimes on the sides. This fox is somewhat larger and has more coarse hairs than the Standard Silver fox, which has only black and white hairs.

Most of these black foxes have agouti hairs with a silver band under the black tip, thus giving them their name. However, all the types above can also have agouti hairs. Through deliberate selection the breeders have obtained a wide range of silver foxes from almost black to foxes with silver hairs all over the back with very wide bands of white, hence making the fur very fair.

(ii) *Model 1: Two di-allelic loci*

Adalsteinsson, Hersteinsson & Gunnarsson (1987) suggested the following theory including two di-allelic loci A (agouti) and E (extension of black eumelanin *vs.* red phaeomelanin pigment) based on homology with mice and sheep.

(*a*) *The A locus.* The agouti locus controls whether the hairs will have a regular pattern of eumelanin and phaeomelanin pigment. The dominant agouti allele A^r inhibits the production of eumelanin and switches it over to production of phaeomelanin in certain areas of the body or in certain fibres. This allele gives the typical red colour of the Red fox, only leaving traces of black pigment on the hair tips.

The recessive non-agouti allele *a* has no effect on the pigment production. The black colour of the Standard Silver fox is the result of homozygosity of this allele. The heterozygous $A^r a$ results in a Gold fox type.

(*b*) *The E locus.* This locus determines extension of eumelanin pigment versus phaeomelanin. The allele E^d is dominant and epistatic to all alleles at the A

Table 1. *Phenotypes and genotypes for the fox according to Adalsteinsson et al. (1987). The genotypes missing are Silver fox types*

Red fox	$A^r A^r$	EE
Gold fox	$A^r a$	EE
Gold Cross fox	$A^r A^r$	$E^d E$
Silver Cross fox	$A^r a$	$E^d E$
Standard Silver	aa	EE
Alaskan Silver	$A^r A^r$	$E^d E^d$

locus, eliminating the inhibiting effect on eumelanin production, thus resulting in extension of black eumelanin pigment all over the body.

The recessive allele *E* has no effect on the pigment production. Heterozygosity $E^d E$ does not result in full extension. The homozygous $E^d E^d$ is usually darker than the heterozygous.

The colour types of this theory are listed in Table 1.

(iii) *Model 2: a di-allelic E locus and a tri-allelic A locus*

The following theory is an extension of the theory of Adalsteinsson *et al.* (1987) described above. It is hoped that this model can provide a better separation of the phenotypes with respect to the genotypes.

In the established agouti series for mice an intermediate agouti allele A^i is found (Silvers, 1979). This allele is dominated by the agouti allele A^r but dominant to the non-agouti allele *a*. The second locus is assumed to be identical to the extension locus described above.

It is assumed that the inhibiting effect of the A^i allele on eumelanin production is not as strong as that of the A^r allele. Thus presence of A^i results in a phenotype less red than caused by the A^r allele but not as black as by the *a* allele.

The homozygous $A^i A^i$ is assumed to be darker than a $A^r a$ but more red than a $A^i a$. The heterozygous $A^i a$ is assumed to be a dark Red fox or a Gold fox. Red fox and Gold fox types are assumed to be homozygous EE. The $A^r A^r \ E^d E$ genotype is possibly an intermediate type between Gold fox and Gold Cross fox. The very dark Silver Cross fox is assumed to be mainly a double heterozygous $A^i a \ E^d E$. Silver fox types are either homozygous aa or $E^d E^d$.

3. Data

The data are a set of pedigrees of farm-bred foxes. The material was collected from four private Scandinavian farms: Hannu, Ilkka, Olavi and Sten, containing information on 247 animals born between 1976 and 1982. For each fox in the pedigrees information is available on the fur type and colour.

The quantity used for analysis is the subjectively evaluated fur colour in regard to the amount of black

Table 2. *Colour classification scale and frequency in dataset*

Colour	Description	N
2–3	Red fox and foxes with very little grey or black on the belly and feet	45
4	Gold fox resembling Red fox but with dark belly	31
5	Gold fox with dark under fur (have almost a cross marking as pups). + Gold Cross with unclear cross	18
6	Gold Cross with clear cross	20
7	Very dark Gold Cross or Silver Cross	7
8	Very dark Silver Cross foxes	3
9	Silver fox	123
		247

pigment compared to red in terms of the above mentioned fox types. The colour classification is made according to the scale given in Table 2, revealing that the types are not represented uniformly.

The animals are classified by several persons and at different ages. Hence it is questionable how comparable these classifications are. A particular problem faced in the dataset is that the pedigrees in themselves are uncertain. All farms but Hannu have cases where litter paternities are unsure, each of the eight cases having two males as possible sire. As an illustration

the genealogy of Olavi farm is given in Fig. 1 (filled symbols indicate that the individual is involved in a paternity case).

(i) *Initial data analysis*

The classification of the animals according to the amount of black and red colour is not always consistent with a classification based on the overall coat colour.

The Gold fox type ought to associate with Colour 4 or 5, nevertheless 27 Gold foxes have either Colour 2 or 3. These Gold foxes may be very red or be missing the black or grey hairs on their belly and feet. It may also reflect a difficulty in distinguishing between Red and Gold foxes. Likewise the Silver Cross type should associate to Colour classes 7 and 8, but 7 Silver Cross foxes are classified as Colour 6. This illustrates that it can be hard to distinguish between the Cross foxes.

The parent–offspring colour frequencies are given in Table 3. Assuming the classifications are correct the genetic model must be consistent with these frequencies.

(ii) *The problem of uncertain paternity*

In many cases a dam is mated with two sires to ensure fertilization, and sometimes the matings are too close in time to tell which male is the father. The most likely
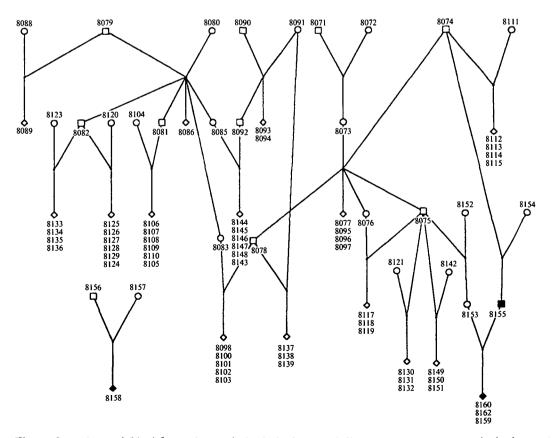

Fig. 1. Genealogy of Olavi farm, the symbols circle, box, and diamond represent respectively dams, sires, and unmated offspring. Filled symbols indicate that the individual is involved in a paternity case.

Table 3. *Frequencies of parent–offspring Colour combinations*

Parent colour	Offspring colour								Total
	2	3	4	5	6	7	8	9	
2 × 2	2								2
2 × 6			2		1				3
2 × 9	3	1	9	9	2			2	26
3 × 9	4	9	4	3	3			11	34
4 × 9	2	8			4	2		21	37
5 × 9		2	4		2			8	16
6 × 7			1			1		1	3
6 × 9	2	2	3		5		2	8	22
7 × 9	1				2	1		3	7
9 × 9								9	9
									160

Table 4. *Cases where litter paternity is unsure. Indices refer to the reported Colour classification of the animal*

Farm	Mother	Father	Father2	Offspring
Ilkka	8011_4	8025_5	8051_9	$8052\text{–}8056_{9,7,4,4,3}$
	8018_9	8014_6	8019_9	$8020\text{–}8023_{6,9,7,4}$
	8024_9	8014_6	8019_9	$8025\text{–}8029_{5,9,9,3,4}$
	8064_5	8031_9	8065_9	$8066\text{–}8069_{7,3,9,9}$
	8013_4	8025_9	8030_9	$8036\text{–}8038_{9,4,4}$
Olavi	8153_9	8155_9	8158_9	$8159\text{–}8162_{6,9,8,9}$
Sten	8236_2	8235_9	8174_9	8181_3
	$8178_?$	8176_9	8177_9	$8203\text{–}8206_{9,9,2,2}$

candidate is stated as father to all pups in the litter and in cases of doubt the second sire is registered as an alternative. However, it is possible that pups from both sires appear in the same litter. Doubt about paternity occurs in eight cases, see Table 4. The case from the Olavi farm illustrates the difficulty in choosing the true father. Both sires mated with animal number 8153 are Silver foxes, and so are the offspring numbers 8160 and 8162. From this information the paternity cannot be established, but a qualified guess might be given if all relevant information available in the pedigree can be used.

4. Methods

(i) *Probability modelling*

In the following we describe the model and the methods used for analysis. Consider a trait which is assumed to be hereditary, thus it is affected by the genotype of the individual, which affects its observed phenotype. Let $\mathscr{G}$ represent the genealogy containing the set of individuals $\mathscr{I}$ and their relationship. Let $\mathscr{F} \subseteq \mathscr{I}$ be the set of founders, i.e. those whose parents are not in the pedigree. The set of individuals with known phenotype is denoted $\mathscr{D} \subseteq \mathscr{I}$. Let ϕ_i be the

phenotype of $i \in \mathscr{D}$, and let the set of observed phenotypes be

$$\phi(\mathscr{D}) = \{\phi_i, \quad \text{for all } i \in \mathscr{D}\}.$$

The genetic model $\mathscr{M}$ describes the trait, i.e. mode of inheritance and effect of the genes. The model consists of the following three components:

(π) Genotype population frequencies: a randomly chosen individual from the population has genotype γ with probability $\pi(\gamma)$.

(ρ) Penetrance probabilities: the relation between genotype and phenotype is described as the probability $\rho(\phi|\gamma)$ that an individual with genotype γ has phenotype ϕ.

(τ) Transmission or segregation probabilities: let $\tau(\gamma_o | \gamma_m, \gamma_f)$ be the probability that an offspring has genotype γ_o given that the genotypes of the parents are γ_m and γ_f.

These are conditional probabilities which are based on the following assumptions about conditional independencies:

(π) Random mating and equilibrium of the population genotype frequencies.

(ρ) The phenotype does not depend directly on shared environmental factors nor on the genotypes or phenotypes of the parents (or others).

(τ) Transmission of chromosomes from the parents to one offspring is conditionally independent of that to another offspring.

The probability of observing the actual phenotypes given the pedigrees and proposed genetic model forms the so-called likelihood function

$$L\{\phi(\mathscr{D}), \mathscr{G}, \mathscr{M}\} = P(\phi(\mathscr{D}) | \mathscr{G}, \mathscr{M}),$$

on which the following analysis is based.

By varying the model $\mathscr{M}$ inference can be made on the components, π, ρ or τ. If we let the genealogy $\mathscr{G}$ vary, $L(\mathscr{G})$ becomes the likelihood of the specific pedigree structure and can thus be used in paternity testing. Eventually unobserved phenotypes can be estimated, i.e. as in genetic counselling on prospective offspring using $\phi(\mathscr{D})$ as the variable.

The conditional independencies listed above enable efficient calculation of the likelihood function. It can be expressed in terms of π, ρ, and τ and as a sum over all genotypes for all individuals. The following factorization of the function can be obtained:

$$L\{\phi(\mathscr{D}), \mathscr{G}, \mathscr{M}\}$$

$$= \sum_{\gamma} \left[\prod_{i \in \mathscr{F}} \pi(\gamma_i) \prod_{j \notin \mathscr{F}} \tau(\gamma_j | \gamma_{pa(j)}) \prod_{k \in \mathscr{D}} \rho(\phi_k | \gamma_k) \right],$$

where γ_i is the set of all possible genotypes of individual i and γ is the set of possible genotypes for all individuals. The term $\gamma_{pa(j)}$ represents the genotypes of the parents of individual j.

If the trait involves more than one locus, linkage must be considered. Let the probability of observing recombination between the two loci be θ, where $0 \leqslant \theta$

Table 5. *Segregation probabilities, assuming parents have genotypes* $(AB)(aB)$ *and* $(AB)(ab)$, *and the probability of observing recombination is* θ. *The parentheses separate the maternal and paternal parts of the genotype*

Offspring genotype	Segregation probability
$(AB)(AB)$	$(1-\theta)/4$
$(AB)(ab)$	$(1-\theta)/4$
$(aB)(ab)$	$(1-\theta)/4$
$(AB)(aB)$	$\theta/4+(1-\theta)/4$
$(AB)(Ab)$	$\theta/4$
$(aB)(aB)$	$\theta/4$
$(aB)(Ab)$	$\theta/4$

$\leqslant 1/2$, the limits indicating full linkage and no linkage, respectively. The recombination fraction θ influences the segregation probabilities τ of the genetic model. An example is given in Table 5.

Linkage is conventionally investigated by calculating the log-odds of θ against the hypothesis of no linkage, i.e. $\theta = 1/2$, as

$$\text{LOD}(\theta) = \log_{10}\frac{L(\theta)}{L(1/2)},$$

where

$$L(\theta) = \sum_{\gamma}\left[\prod_{i\in\mathscr{F}}\pi(\gamma_i)\prod_{j\notin\mathscr{F}}\tau(\gamma_j\,|\,\gamma_{pa(j)};\theta)\prod_{k\in\mathscr{D}}\rho(\phi_k\,|\,\gamma_k)\right].$$

A high LOD score at $\theta \simeq 0$ is taken as indication of linkage.

(ii) *Paternity testing*

The usual approach to paternity testing is to compare the likelihoods of the pedigrees where each possible father appears as the actual father. In the case of Ilkka farm this gives essentially 2^{21} possible pedigrees to compare. Instead we will use the fact that the genotypes can be estimated. Based on the given phenotypic information the genotype distributions of the males in question can be estimated. These are then compared with the expected genotype distribution of the unknown father. A reasonable approach is to avoid representing them as fathers of the child and represent the unknown father as a founder with known phenotype only if the males tested have the same phenotype. The most likely father can be picked out either by exclusion or by comparing their most probable genotypes.

(iii) *Calculation and maximization*

Calculation of the likelihood as formulated above will require computations of order $g^{|\mathscr{F}|}$, where g is the number of genotypes and $|\mathscr{F}|$ is the number of

individuals. Thus likelihood calculation seems to be impossible in practice, but by exploiting the structure of conditional independence represented in the pedigree, the likelihood function can be factorized using the peeling algorithm (Cannings, Thompson & Skolnick, 1978). This is implemented in the Pedpack program package (Thomas, 1987), which is used for calculation of likelihoods and genotype distributions and for drawing the marriage-node graphs. The simplex method of Nelder & Mead (1965) has been used for the search of maximum likelihood estimates (Press, Flannery, Teulosky & Vetterling, 1988).

Some kind of parameter reduction is sensible if one is considering highly parameterized models like ours. Furthermore it is reasonable to put constraints on the penetrance to make the computations correspond to the described models. Hence, estimates maximizing the likelihood are only found for penetrance parameters initialized non-zero. In this way the number of parameters to estimate is reduced.

Optimal genotype frequencies are determined under the assumption of genetic equilibrium in the population. Maximum likelihood estimates of population allele frequencies are found optimizing the likelihood function from random starting values. By plotting the LOD-score for different recombination fractions the possibility of linkage is studied.

The maximization process is performed iteratively and sequentially, restricting the number of varying parameters in each step to at most three or four.

5. Results

The CPU-times for one likelihood calculation on the two models discussed below were approximately 6 min and 16 h respectively on a SUN-4 machine with 32Mb RAM. In this light the second model was optimized on the basis of the observations from farms Hannu, Sten, and Olavi only; this brought the CPU-time needed down to 7 min. Still the total CPU-time required depends on how many likelihood calculations are needed in the optimization process.

Based on the theory of Warwick & Hanson (1937) a fairly simple model was implemented with two independent di-allelic loci penetrating into 5 phenotypes, see Section 2(i). This model was fitted and subsequently used for testing paternities. All results given below are based on the paternity tested material.

The estimates reported are those associated with the best likelihood value found in a number of maximizations repeated with various modifications, like new starting points or changed sequence of parameters optimized simultaneously. The observed variation on the penetrance probabilities was more than that of the recombination fractions and population allele frequencies.

Table 6. *Model 1: resulting penetrance and population frequencies based on the paternity tested data. The indices h, l and s, referring to respectively high, low and sure, illustrate the relative size of the parameters in the initial penetrance table*

Genotype	Phenotype								Population frequencies
	2	3	4	5	6	7	8	9	
$A^r A^r$ EE	0.66_h	0.16_h	0.18_l	—	—	—	—	—	0.02
$A^r a$ EE	0.19_l	0.30_l	0.37_h	0.14_h	—	—	—	—	0.19
$A^r A^r$ $E^d E$	—	—	—	0.45_l	0.54_h	0.00_l	—	—	0.01
$A^r a$ $E^d E$	—	—	—	—	0.67_l	0.27_h	0.06_h	—	0.09
$A^r A^r$ $E^d E^d$	—	—	—	—	—	—	0.003_l	0.997_h	0.001
aa EE	—	—	—	—	—	—	0.02_l	0.98_h	0.43
$A^r a$ $E^d E^d$	—	—	—	—	—	—	—	1_s	0.01
aa $E^d E$	—	—	—	—	—	—	—	1_s	0.22
aa $E^d E^d$	—	—	—	—	—	—	—	1_s	0.03

(i) *Model 1: two di-allelic loci*

The relative sizes of the initial penetrance probabilities are shown as indices in Table 6. The decision on which Colour phenotype ought to have high or low probability is based on the scale of the Colour phenotype given in Table 2. E.g. the genotype $A^r a$ EE is a Gold fox, which is either Colour 4 or 5, thus these penetrance probabilities ought to be high. In Section 3 we identified some inconsistencies concerning the Gold fox type, that many Gold foxes were classified with Colour 2 or 3, thus these are also assumed to be probable Colour phenotypes of the $A^r a$ EE genotype. The actual entries in the penetrance matrix are not crucial, almost all probability mass is placed on the proposed highly probable phenotypes. The initial population frequencies are estimated from the observed phenotypes. Initially the loci are assumed to be not linked, i.e. $\theta = 1/2$. The initial model has log likelihood:

$$\log L = -416.7.$$

The genetic model obtained through the model search has penetrance and frequencies as given in Table 6.

The population allele frequencies are

$$P(A^r, a) = (0.18, 0.82)$$

$$P(E, E^d) = (0.81, 0.19).$$

The linkage analysis results in the LOD score curve shown in Fig. 2. The final genetic model has log likelihood

$$\log L = -357.5.$$

(ii) *Model 2: a di-allelic E locus and a tri-allelic A locus*

The model is initialized in the same manner as the previous model; the relative sizes of the penetrance
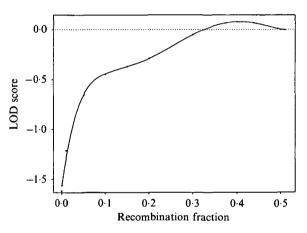

Fig. 2. Model 1: LOD score curve for linkage of loci.

probabilities are indicated as indices in Table 7. The structure of the Ilkka farm genealogy results in very time-consuming calculations on this model. Hence it was excluded in the maximization process. The proposed model has, on the restricted data, the following log likelihood

$$\log L = -369.9,$$

and on the complete data it is

$$\log L = -480.8.$$

The model search results in the penetrance and genotype frequencies shown in Table 7. The population allele frequencies are

$$P(A^r, A^t, a) = (0.16, 0.21, 0.63)$$

$$P(E, E^d) = (0.62, 0.38).$$

The linkage analysis results in the LOD score curve shown in Fig. 3. This genetic model has, on the restricted data, log likelihood

$$\log L = -258.1,$$

and on the complete data it is

$$\log L = -398.1.$$

Table 7. *Model 2: resulting penetrance and population frequencies. The indices h, l, s referring to respectively high, low and sure illustrate the relative size of the parameters in the initial penetrance table*
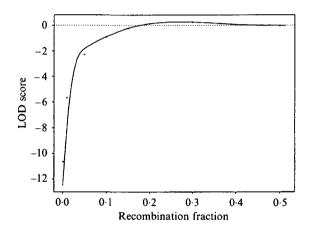
Genotype	Phenotype								Population frequencies
	2	3	4	5	6	7	8	9	
$A^r A^r$ EE	$1\cdot00_h$	$0\cdot00_l$	—	—	—	—	—	—	0·009
$A^r A^i$ EE	$1\cdot00_l$	$0\cdot00_h$	—	—	—	—	—	—	0·02
$A^r a$ EE	—	$0\cdot67_l$	$0\cdot24_h$	$0\cdot09_l$	—	—	—	—	0·07
$A^i A^i$ EE	—	—	$0\cdot30_l$	$0\cdot70_h$	—	—	—	—	0·02
$A^i a$ EE	—	—	—	1_s	—	—	—	—	0·09
$A^r A^r$ $E^d E$	—	—	—	$0\cdot96_h$	$0\cdot03_l$	—	—	—	0·01
$A^r A^i$ $E^d E$	—	—	—	—	1_s	—	—	—	0·03
$A^r a$ $E^d E$	—	—	—	—	$0\cdot75_l$	$0\cdot25_h$	—	—	0·09
$A^i A^i$ $E^d E$	—	—	—	—	—	$0\cdot00_h$	$1\cdot00_l$	—	0·02
$A^i a$ $E^d E$	—	—	—	—	—	—	$0\cdot00_h$	$1\cdot00_l$	0·13
$A^r A^r$ $E^d E^d$	—	—	—	—	—	—	—	1_s	0·004
$A^r A^i$ $E^d E^d$	—	—	—	—	—	—	—	1_s	0·01
$A^r a$ $E^d E^d$	—	—	—	—	—	—	—	1_s	0·03
$A^i A^i$ $E^d E^d$	—	—	—	—	—	—	—	1_s	0·01
$A^i a$ $E^d E^d$	—	—	—	—	—	—	—	1_s	0·05
aa EE	—	—	—	—	—	—	—	1_s	0·13
aa $E^d E$	—	—	—	—	—	—	—	1_s	0·19
aa $E^d E^d$	—	—	—	—	—	—	—	1_s	0·07

Fig. 3. Model 2: LOD score curve for linkage of loci.

With respect to the second model with an intermediate agouti allele the obtained penetrance in Table 7 is more restrictive than initiated. The very dark Silver Cross fox, Colour 8, has genotype $A^i A^i$ $E^d E$ and not as proposed $A^i a$ $E^d E$, which instead is a Colour 9. The model shows no difference between penetrance of $A^r A^r$ EE or $A^r A^i$ EE. It appears that the genotypes aa EE, and aa $E^d E$ are the most frequent Silver fox types. The analysis of linkage does not give any evidence on possible linkage of the loci (see Fig. 3). Hence, the loci are most probably placed on two different chromosomes.

With respect to both models it is seen that phenotypes which are very rare according to the data will obtain very low penetrance probabilities.

6. Conclusion

From the results for Model 1 it appears very clearly from the population frequencies that the Standard Silver fox genotype and its hybrids are the most frequent genotypes, whereas the Alaskan Silver fox genotype is the most rare. The obtained penetrance matrix diverges markedly from the initial penetrance. The Gold Cross $A^r A^r$ $E^d E$ does not penetrate into Colour 7 as expected. The Silver Cross genotype $A^r a$ $E^d E$ penetrates almost exclusively into Colour 6 or 7; only 6% are penetrating into a 'very dark Silver Cross'. The Silver fox genotypes $A^r A^r$ $E^d E^d$ and aa EE mainly penetrate as Colour 9, Silver foxes. The LOD score curve for linkage (see Fig. 2) gives no evidence of linkage between the two loci. This first model does not provide a clear classification of the genotypes with regard to phenotype as classified by the Colour scale.

(i) Summary of results

For each model the improvement in likelihood shows that the final models give a better fit to the data than the initial models. Model 2 should be capable of giving a more detailed description of the data than Model 1; this should be expressed in a higher likelihood if they are evaluated on the same dataset. The likelihoods show that Model 1 describes the data at least as well as Model 2. Hence there seems to be no gain in having the more complicated model, at least as it is represented in Table 7. The second model is however optimized without using the data from the Ilkka farm. Unless the farms are homogeneous with respect to phenotype classifications and genotypic composition, it is inevitable that the data from the Ilkka farm will not be in good accordance with a model fitted to the data from the other farms. This is

2

actually seen as a net decrease in gained likelihood when comparing the final likelihood with the initial, with and without the data from the Ilkka farm. It is evident that much information has been lost due to the exclusion of the Ilkka farm data. The lack of sufficient information showed up in that some of the genotypes could penetrate into a specific phenotype within a wide range of probabilities without affecting the likelihood. Regarding the population frequencies and recombination fractions the observed variation was small, both models considered.

Although the second model is at first glance more complex it has several penetrance parameters at or close to 1. The first model however needs more penetrance probabilities and so has more parameters.

The conclusion on the second model is that it is not unlikely that a third allele does exist on the agouti locus, but no evidence is found that it should be expressed as assumed above.

A proper evaluation of especially the more detailed hypothesis of Model 2 requires very precise data, where all phenotype and family information can be taken as certain. It is evident that a very detailed model is more sensitive towards misclassifications, since this inevitably brings possible inconsistencies into the data which the model cannot embed. These inconsistencies will then be modelled as wide ranges for penetrance of the genotypes. If more certain phenotype classifications were available it is believed that the data could be used to propose and evaluate better hypotheses.

References

Adalsteinsson, S., Hersteinsson, P. & Gunnarsson, E. (1987). Fox colors in relation to colors in mice and sheep. *Journal of Heredity* **78**, 235–237.

Cannings, C., Thompson, E. & Skolnick, M. (1978). Probability functions on complex pedigrees. *Advanced Applied Probability* **10**, 26–61.

Iljina, E. D. (1934). The inheritance of basic coat colour in the fox. *Zoologitscheskij Journal* **13**, 701–713.

Nelder, J. A. & Mead, R. (1965). A simplex method for function minimization. *Computational Journal* **7**, 308.

Nes, N. N., Einarsson, E. J. & Lohi, O. (1988). *Beautiful Fur Animals – and their Colour Genetics*. Denmark: Scientifur.

Press, W. H., Flannery, B. P., Teulosky, S. A. & Vetterling, W. T. (1988). *Numerical Recipes in C*. Cambridge: Cambridge University Press.

Searle, A. G. (1968). *Comparative Genetics of Coat Colours in Mammals*. London: Logos Press.

Silvers, W. K. (1979). *The Coat Colors of Mice*. New York: Springer-Verlag.

Thomas, A. W. (1987). Pedpack: user's manual. *Technical Report 99*. Seattle: Department of Statistics, University of Washington.

Warwick, B. L. & Hanson, K. B. (1937). The breeding of fur animals. In *Yearbook of Agriculture* (ed. F. G. Ashbrook), pp. 1379–1395. U.S. Department of Agriculture.