

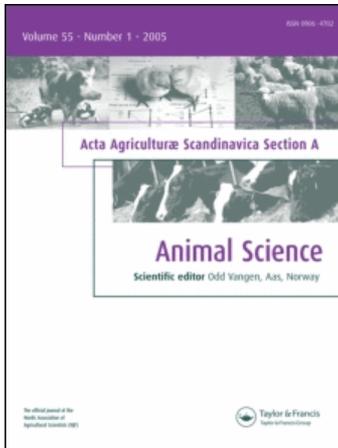
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Acta Agriculturae Scandinavica, Section A - Animal Science

Publication details, including instructions for authors and subscription information:

<http://www.informaworld.com/smpp/title~content=t713690045>

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Online publication date: 23 April 2010

To cite this Article Olsen, H. F. , Klemetsdal, G. , Ruane, J. and Helfjord, T.(2010) 'Pedigree structure and genetic variation in the two endangered Norwegian horse breeds: Døle and Nordland/Lyngen', Acta Agriculturae Scandinavica, Section A - Animal Science, 60: 1, 13 – 22

To link to this Article: DOI: 10.1080/09064701003639884

URL: <http://dx.doi.org/10.1080/09064701003639884>

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ORIGINAL ARTICLE

Pedigree structure and genetic variation in the two endangered Norwegian horse breeds: Døle and Nordland/Lyngen

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Abstract

To examine the genetic variation in two endangered Norwegian horse breeds, the pedigree structures were investigated, and key parameters such as inbreeding coefficients, effective population size, effective number of founders, effective number of ancestors and effective number of founder genomes were calculated. The data consisted of 31,142 individuals of the Døle horse and 1973 individuals of the Nordland/Lyngen horse, for which the complete generation equivalent was 10.5 and 7.2, respectively. In both breeds, the pedigree data were more than 98.5% complete in the fourth generation, allowing the rate of inbreeding and the effective population size to be precisely estimated, actually measuring the fractional loss of heterozygosity, comparable across generations (not so for the other measures). The level of inbreeding was about 12% in both breeds, with a rather wavy pattern during the past 50 years in the Døle. Considering the last generations only, the effective population size was found to be 152 in the Døle and 42 in the Nordland/Lyngen. For both populations selection in the future should be based on optimal contribution.

Keywords: Breed conservation, effective population size, pedigree analysis, probability of gene origin.

Introduction

Norway is a country that is relatively rich in domestic animal genetic resources (Ruane, 2000). However, for a variety of reasons, such as changes in farming practices, many of its breeds are in danger of extinction in the near future. Conservation of these resources is a challenge, requiring a multi-faceted approach, covering a range of aspects such as increasing public awareness about their importance or ensuring that the breeds are managed in a sustainable way so that problems of inbreeding and loss of genetic variation are minimised. Here, we focus on this latter aspect and report on the pedigree structure and genetic variation in two endangered Norwegian horse breeds.

The populations of the Norwegian Nordland/Lyngen and the Døle (www.ansi.okstate.edu/breeds/horses) are small, and breeders also exercise artificial selection. Selection reduces the effective population size (N_e), because the number of

breeding animals is reduced, and in an already small population it accelerates genetic drift. In such small populations genetic drift, in which allele frequencies at a locus change at random from one generation to the next, increases the probability of fixation of alleles and reduction of additive genetic variation.

Changes in allele frequencies also result in changes of genotype frequencies, and a locus will contribute to a change of the population mean due to inbreeding if a dominance effect is present (Falconer & Mackay, 1996) (in addition, mean change from allele frequency changes on non-neutral loci). The frequencies of homozygotes increase and recessives accumulate in the population, leading to the phenomenon known as inbreeding depression. The smaller the population is, the more it genetically drifts, and the larger the probability is that the alleles become identical by descent, defining the coefficient of inbreeding (F). In a case with a neutral locus, no mutation and no migration, the inbreeding

coefficient thus reflects how far the process of genetic drift has progressed, relative to the base population, whereas the inbreeding rate describes the speed of the process. This study examined the pedigree structure of two endangered breeds and calculated various parameters describing genetic variation, such as the effective population size, from the rate of inbreeding and the coefficient of kinship, and those based on probabilities of gene origin, with the aim of drawing inference as to future genetic management of the breeds.

Materials and methods

Data were received from the Norwegian Equine Centre in autumn 1999. Recent birth years are not included in this work due to loss of data in the process of changing data registration system at the Norsk Rikstoto in 2006 (zeroing parents not recorded in the studbook as an individual by itself, see below, meaning that the most remote generation of ancestors could not be included in the analyses). The original material used for this study contained all the animals ever registered in these two breeds: 30,712 registered animals of the Døle and 1969 registered animals of the Nordland/Lyngen. The first registered Døle was a stallion born in 1846. In the Nordland/Lyngen, the first registered animal was a mare born in 1906. In both breeds, the last birth year of registered animals was 1998. All the ID-numbers were renumbered, and the files were edited to remove obvious errors such as ID-numbers in incorrect format or logical errors where the parent is born after its progeny, or errors where parents were not recorded in the studbook as an individual by itself. These parents were added as individuals with no pedigree information. In the Døle, the number of added animals was 430, whereas the respective number in the Nordland/Lyngen was four animals. In Døle, these were mainly Norwegian cold-blooded trotters (the light version of Døle; see www.ansi.okstate.edu/breeds/horse), but also some individuals of North-Swedish horse, with a Døle origin. Of these, 108 were added with unknown birth year (90 Norwegian cold-blooded trotters and 18 North-Swedish horses). In Nordland/Lyngen, three of the four animals were of other origin (Fjord horse). The last of the four is a fictitious animal. At the end of this procedure, the total number of records generated for the two breeds were 31,142 and 1973, respectively (Table I).

The reference populations were defined as horses born between 1990 and 1998, with a total of 1535 horses in Døle, while the corresponding number for the Nordland/Lyngen was 1050 horses (Table II).

All ancestors of the animals in the reference populations were then traced back to the founders,

in an accumulation procedure (individual by individual), also to reveal the completeness of the data. The founders of these populations were defined as horses with unknown parents. When an animal had only one parent known, the unknown parent was considered as a founder (Boichard et al., 1997). In Nordland/Lyngen one Finn horse import existed, Viri (Rian, 2003), and was made a founder by deleting its pedigree information. This pedigree information was originally included only because five generations of pedigree information is required in the Norwegian studbook. In Døle, the pedigree of the imports was not deleted in the same manner as described for Nordland/Lyngen, because the number of founders originating from Norwegian cold-blooded trotters was too high, and the relationship between the two breeds is significant. However, proper identification of trotters would require access to both studbooks.

From the accumulated files, the maximum number of generations in the pedigree (g_{\max}) was derived as was the percentage of known ancestors per generation, as well as the complete generation equivalent (CGE):

$$CGE = \frac{1}{N} \sum_{j=1}^N \sum_{i=1}^{n_j} \frac{1}{2^{g_{ij}}},$$

where N is the number of individuals in the reference population, n_j is the number of ancestors generated for animal j and g_{ij} is the number of generations between individual j and its ancestor i (Boichard et al., 1997). CGE can be interpreted as the number of generations in a comparable complete pedigree, illustrating the depth of the pedigree data.

To describe the genetic variation in a population, several measures can be used. To quantify the rate of genetic drift the rate of inbreeding is most frequently used (Boichard et al., 1997). Unfortunately, the coefficients of inbreeding are very sensitive to incomplete data. A complementary approach, which is more robust, is to analyse the probabilities of gene origin. Parameters from probabilities of gene origin, i.e. the effective number of founders (f_e), effective number of ancestors (f_a) and effective number of founder genomes (f_g), are also affected by incomplete pedigree information, but to a smaller extent than parameters derived from inbreeding coefficients (Boichard et al., 1997).

Effective population size

Individual inbreeding coefficients were calculated using the Quaas–Henderson algorithm (Henderson, 1976; Quaas, 1976). The effective population size is defined as the number of individuals in an idealised

Table I. The number of animals in the total pedigree in the Døle and in the Nordland/Lyngen, respectively.

	Døle			Nordland/Lyngen		
	Stallions	Mares	Total	Stallions	Mares	Total
Number of animals	3739	27,403	31,142	720	1253	1973

population, which would give rise to the same inbreeding rate (ΔF) as observed in the real population (Falconer & Mackay, 1996). The effective population size (N_e) in generation t , unconditional of becoming selected as parents or not in generation t , was computed according to the formula:

$$N_{e_t} = \frac{1}{2\Delta F_t},$$

where the rate of inbreeding, ΔF_t , is:

$$\Delta F_t = \frac{1}{N} \sum_{i=1}^N \frac{F_i - \frac{1}{2}(F_{sire_i} - F_{dam_i})}{1 - \frac{1}{2}(F_{sire_i} - F_{dam_i})}$$

with F_i being the individual coefficient of inbreeding for animal i in generation t (defined reference population), and F_{sire_i} and F_{dam_i} are the individual inbreeding coefficient of the parents in generation $t-1$.

Effective number of founders

Effective number of founders was defined as the number of equally contributing founders that would produce the same genetic diversity as in the reference population under study (Lacy, 1989):

$$f_e = \frac{1}{\sum_{k=1}^f q_k^2},$$

where q_k is the genetic contribution of founder k to the population's gene pool. The balance of the founder contributions measures the preservation of the genetic diversity from the founders to the present population. If each founder contributes the same, the effective number of founders equals the actual number of founders. In any other situation, the effective number is smaller than the actual number of founders. An important limitation with this approach is that it ignores potential bottlenecks in the pedigree (Boichard et al., 1997). Computation is detailed in Olsen et al., (2005).

Effective number of ancestors

The effective number of ancestors describes the minimum number of ancestors (they may or may not be founders) required to explain the complete genetic diversity of the population under study (Boichard et al., 1997):

$$f_a = \frac{1}{\sum_{k=1}^f p_k^2},$$

where

$$p_k = q_k \left(1 - \sum_{i=1}^{n-1} a_i \right)$$

and p_k is the marginal contribution of ancestor k to the population's gene pool, which is q_k adjusted for the genetic contribution a_i from the $n-1$ already

Table II. The number of registered stallions and mares in the reference population in the Døle and in the Nordland/Lyngen, respectively.

Birth year	Døle			Nordland/Lyngen		
	Stallions	Mares	Total	Stallions	Mares	Total
1990	67	91	158	42	60	102
1991	64	95	159	56	53	109
1992	58	74	132	42	64	106
1993	64	87	151	58	51	109
1994	78	89	167	39	51	90
1995	99	100	199	65	58	123
1996	96	88	184	63	68	131
1997	115	100	215	83	65	148
1998	93	77	170	64	68	132
Total	734	801	1535	512	538	1050

selected ancestors. Initially, the largest contributing ancestor was determined as the one having the largest q -value. When determined, the contribution taken out by that ancestor should not be assigned to more remote ancestors. This is taken account of by eliminating its pedigree information, leaving the animal as a “pseudo founder”. The animal’s descendants also need to get their contribution adjusted for the contributions from this “pseudo founder”, leading to marginal contributions, i.e. contributions not explained by other ancestors. In this approach, p_k is q_k adjusted for the genetic contribution a_i from the $n-1$ already selected ancestors to individual k , and for each major ancestor found, its pedigree information is deleted, i.e. the sire and dam information is set to zero, eliminating redundancies. For calculations, see Olsen et al. (2005). In the Nordland/Lyngen, all ancestors with a non-zero contribution to the reference population (horses born 1990–1998) were determined. In the Døle, we stopped the procedure with the 100 highest contributing ancestors, as these individuals covered more than 97% of the contribution to the reference population. The effective number of ancestor accounts for potential bottlenecks, but still the probability of gene loss by drift needs to be considered.

Effective number of founder genomes

To account for loss of genetic variability by genetic drift, Lacy (1989, 1995) and Ballou and Lacy (1995) introduced the concept of effective number of founder genomes, or founder genome equivalent. This measures how many founder genes are maintained in the population for a given locus, and how balanced their frequency is in the offspring of the reference population. Originally, this parameter was calculated by probability calculations (Lacy, 1989) or by gene dropping analysis (MacCluer et al., 1986). Instead, Lacy (1995) proposed to stay with a definition of the founder genome equivalent as half the inverse of average kinship in generation t (the reference population):

$$f_g = \frac{1}{2\bar{f}_t}$$

This is supported, especially from a practical point of view, by Caballero and Toro (2000) and Zechner et al. (2002). For details in computation, see Olsen et al. (2005).

Effective population size from kinship

As the inbreeding coefficient describes the similarity between two alleles in each animal and hence

describes effects of past decisions, it is in addition interesting to look at the average pair wise similarity between the animals in the reference population, i.e. the coefficient of kinship (Falconer & Mackay, 1996). This gives us an indication of current bottlenecks. The coefficient of kinship between two animals equals the inbreeding coefficient of their progeny (Falconer & Mackay, 1996). Therefore, we used the average pair wise kinship to predict an approximate rate of F in the next generation (ΔF_{t+1}), given no selection:

$$\Delta F_{t+1} = \frac{f_t - F_t}{1 - F_t}$$

where f_t is the average pair wise kinship in the reference population, independent of sex and including selfing, and F_t is the corresponding average inbreeding coefficient. The calculation was done repeatedly, each year including the last three-year classes of the reference population.

Mating strategies

Kinship calculation (excluding selfing and intrasex mating) was also used to test whether systematic favour of inbreeding is practised as a mating strategy. This was done by calculating the difference between the average inbreeding coefficients in generation $t(F_t)$ and the average pair wise kinship of their parents in generation $t-1(f_{t-1})$, compared with a two-sample t -test.

Results

In the Døle, animals in the reference population had an average number of 22,464 ancestors (Table III). The corresponding number in the Nordland/Lyngen was 495. Still, both breeds had a pedigree completeness of more than 98.5% in Generation 4 (Table III). Including more generations in calculations, a more complete pedigree was demonstrated in the Døle than in the Nordland/Lyngen. In Table III, the quality of the pedigree information is also indicated by the CGE. In the Døle the pedigree can be traced back a maximum of 25 generations, where CGE is 10.5. In the Nordland/Lyngen, CGE was 7.2, with tracing of pedigree for a maximum of 13 generations.

Figure 1 shows that inbreeding levels in the two breeds evolved somewhat differently throughout the past 100 years. The average inbreeding coefficient for the Døle increased to about 7% in the 1940s, after which it fell significantly before rising again towards the end of the 1970s. During the past few years, the inbreeding coefficient has been around 12%. For the Nordland/Lyngen, the average

Table III. Calculated parameters describing pedigree completeness of the reference population; in the Døle and in the Nordland/Lyngen, respectively.

		Døle	Nordland/Lyngen
Number of animals in reference population		1535	1050
Average number of ancestors per animal		22,464	495
Percentage (%) of known ancestors in generation	1	99.97	99.95
	2	99.89	99.95
	4	98.64	99.83
	6	90.51	91.09
	8	78.66	44.41
	10	66.76	6.00
Maximum number of generations generated (g_{\max})		25	13
Complete generation equivalent (CGE)		10.5	7.2

inbreeding coefficient has been high throughout most of the period. At present, the average lies at the same level of inbreeding one gets with half-sib mating (12.5%).

In the reference population of the Døle, the average inbreeding coefficient was 11.75% (Table IV), while it was 12.77% in Nordland/Lyngen. The corresponding effective population sizes calculated by use of the classical formula were 152 in the Døle and 42 in the Nordland/Lyngen (Table IV).

The number of founders in Døle and Nordland/Lyngen was 770 and 42, respectively (Table IV). In both breeds, there was a larger number of female than male founders (Table V). In Døle, the founders spread out over a long-time span, while in Nordland/Lyngen the majority of the founders were born in the first half of the twentieth century (Table V).

The effective number of founders, f_e , was 48 for the Døle and 14 for the Nordland/Lyngen (Table IV). The corresponding values for effective number of ancestors, f_a , were 12 for the Døle and seven for Nordland/Lyngen, while the effective number of founder genomes, f_g , was calculated as 5.4 for the Døle and 3.7 for Nordland/Lyngen (Table IV).

Table VI shows the rate of inbreeding in generation $t+1$ over a period of 7 years (1992–1998), that results from mating of the last three-year classes, continuously. The resulting trends were in opposite

in the two breeds; with an increasing trend of the rate of inbreeding in generation $t+1$ in the Døle, while it was decreasing in Nordland/Lyngen.

Table VII demonstrates a significantly higher inbreeding coefficient of offspring in the Døle, than the average, pair wise kinship of parents, indicating a mating strategy favouring inbreeding. The tendency of the opposite; a mating strategy avoiding inbreeding, was seen in the Nordland/Lyngen, although not statistically significant (Table VII).

Table VIII shows, as a curiosity, the five most important ancestors in the pedigrees of the reference populations, as represented by their marginal contributions. Together, these five contributed more than 50 and 70% to the gene pools of the reference population in the Døle and in the Nordland/Lyngen, respectively (Table VIII).

Discussion

In Norway, the Norwegian Equine Centre has been responsible for registration of the Døle and the Nordland/Lyngen, since 1988 (Norsk Hestesenter, 1988). From 1990, the rules for registration were changed, so that all horses being born could be registered (Norsk Hestesenter, 2003). Still, there is a lack of registration, probably because it is voluntary and there are some expenses with the registration

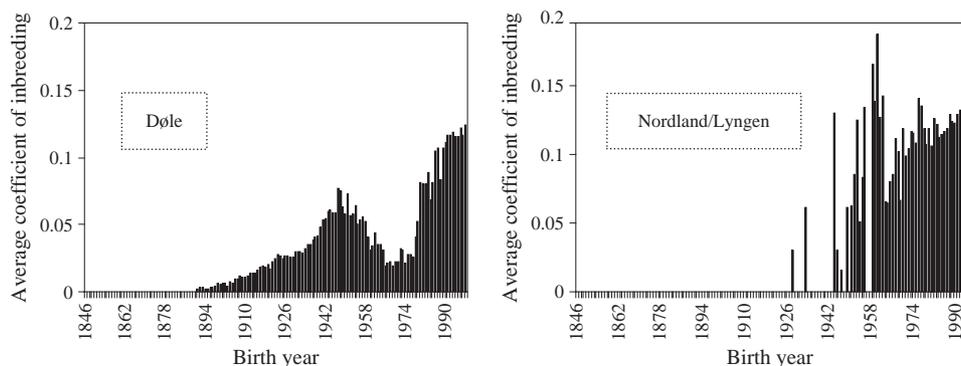


Figure 1. Average coefficient of inbreeding per year of birth; in the Døle and in the Nordland/Lyngen, respectively.

Table IV. Calculated parameters describing genetic diversity of the reference population in the Døle and in the Nordland/Lyngen, respectively.

	Døle	Nordland/ Lyngen
Average coefficient of inbreeding (F_I)	0.1175	0.1277
Rate of inbreeding ($\Delta F_I \pm SE$)	0.0033 ± 0.0455	0.0119 ± 0.0014
Effective population size (N_e)	152	42
Number of founders (f)	770	42
Effective number of founders (f_e)	48	14
Effective number of ancestors (f_a)	12	7
Effective number of founder genomes (f_g)	5.4	3.7

papers, and if a horse is not to be used in breeding it may not be registered. Both breeds have developed breeding plans since 1995 (Vangen, 1996). The breeding goals include weighting of conformation, performance, temperament and durability, and selection is on phenotype. The stallions must be licensed for use in the breeding (e.g. Klemetsdal, 1998).

In the Nordland/Lyngen, the reference population accounted for almost 53% of the total pedigree. By comparison, the corresponding number in the Døle was only 5%. The reason for this can be found in a retrospective set up of the first studbook in Nordland/Lyngen in 1969 (Statens Stambokkontor, 1969), after years of struggle to keep the breed alive,

while the first studbook in Døle was published back in 1902 (Det kgl. selskab for Norges Vel 1902). With a more retrospective set up, the number of animals in the pedigree became restricted, explaining most of the differences in the percentages that the reference population made up of all animals in the pedigree. Additionally, Nordland/Lyngen was nearly extinct in the 1950s, and so went through a narrow bottleneck (Roaldsøy, 1969). Many individuals of the Nordland/Lyngen were lost during the Second World War and the mechanisation in agriculture made the working horses redundant (Roaldsøy, 1969), and so the systematising of the breeding was a last attempt to save the breed. They started with very few animals in the studbook, and these became the important founders of today's current population (Statens Stambokkontor, 1969).

The proportion of founders in the pedigree of the Døle was quite high. As mentioned, the breeding of the Døle was systematised at the end of the nineteenth century, and the first studbook for the Døle included stallions approved from 1859 onwards, with unknown missing values assigned to parents that themselves were not registered in the studbook. For mares, the same registration practise was followed from 1878 onwards (Borchgrevink, 1906). With basically an open studbook as in Døle, it is obvious that the number of founders will be high, spreading out over a long-time span. Furthermore, with more female than male founders, the likelihood of missing parental information will be smallest among males, resulting in more male founders born far back in time. This result was most likely

Table V. Number of founders by sex and decade in the Døle and in the Nordland/Lyngen, respectively.

Decade of birth	Døle			Nordland/Lyngen		
	Stallions	Mares	Total	Stallions	Mares	Total
0	26	82	108	1	3	4
1840	3	0	3	–	–	–
1850	5	0	5	–	–	–
1860	6	0	6	–	–	–
1870	25	5	30	–	–	–
1880	42	61	103	–	–	–
1890	20	91	111	–	–	–
1900	12	47	59	2	4	6
1910	9	32	41	3	4	7
1920	21	31	52	2	3	5
1930	25	57	82	5	8	13
1940	29	41	70	0	1	1
1950	22	32	54	0	1	1
1960	11	19	30	1	1	2
1970	4	7	11	0	2	2
1980	3	1	4	0	0	0
1990	0	1	1	1	0	1
Total	263	507	770	15	27	42

Table VI. Rate of inbreeding in generation $t+1$, calculated each year from average, pair wise kinship (allowing selfing and intrasex mating) and inbreeding coefficients, including the last three-year classes of the reference population in the Døle and in the Nordland/Lyngen, respectively.

Year class	Døle	Nordland/Lyngen
1990–1992	0.0079	0.0157
1991–1993	0.0082	0.0148
1992–1994	0.0101	0.0149
1993–1995	0.0122	0.0129
1994–1996	0.0133	0.0091
1995–1997	0.0137	0.0101
1996–1998	0.0133	0.0121

strengthened from studbook recording being initiated later in females than in males.

From 1990 onwards, registration of horses born in Norway required, if possible, a five-generation pedigree for all horses (Seterlund et al., 1989). This is also reflected in the results for Døle and Nordland/Lyngen, as the reference populations had 98.6 and 99.8% of the ancestors in the fourth generation, respectively. Digitalisation was based on the data used by Vangen (1983), but animals only known as parents in the studbook were deleted together with their pedigrees. This led to a considerable loss of data, which could have been avoided by splitting the studbook in two. In Nordland/Lyngen, the data were very complete in the first six to eight generations of the reference population, despite data only going back to 1906. As mentioned, recording for this studbook was originally done retrospectively, restricting the number of animals in the pedigree, as well as the number of founders. A consequence of the current registration practise was that one rather recent founder was found, e.g. the one mare founder born during the 1990s (Table V), likely with an incomplete pedigree.

Two different approaches were used to quantify the amount of genetic variation; the probability of gene origin measures and the effective population size, from the rate of inbreeding. In essence, the former calculates the probability of gene loss, which by definition will change over generations (Boichard et al., 1997). In contrast, the rate of inbreeding relates to the fractional loss of heterozygosity from generation t to $t+1$, being the same in all generations, for the same breeding structure

(Boichard et al., 1997). In consequence, the probability of gene loss measures is not directly comparable between breeds with different pedigree depth, as was the situation with Døle and Nordland/Lyngen. Furthermore, with the shift in registration practice in 1990, the base generation will not be precisely defined, further reducing the value of these measures. In contrast, with an almost complete pedigree for five generations in both breeds, the asymptotic rate of inbreeding will be estimated without much error (for the asymptotic rate, see e.g. Klemetsdal, 1999). With our data, this gives a preference to the rate of inbreeding and the effective population size over the probability measures. Besides, the rate of inbreeding is only slightly affected by the base definition. An improved calculation of the level of inbreeding and kinship coefficients could have been done by adopting the method of VanRaden (1992), assuming founding trotters as unrelated (although these also have a Døle origin, see e.g. Klemetsdal, 1998), and the remaining founders being related to the contemporary animals, as determined by the level of inbreeding.

The effective number of founders, f_e , only corrects for unbalanced founder contributions in the base generation. At later stages of a breeding programme, the contributions of the founders cannot be affected. Only contributions from animals in the current to the next generation can be influenced. This is possible by the use of the theory of inbreeding. The effective number of ancestors (f_a) giving the marginally most contributing ancestors (the bottlenecks) suffers from the same deficiency as the effective number of founders. The contributions of remote ancestors (Table VII) are not possible to affect at present, and thus they are only contributing through drift. Actually, the speed of drift (the rate of inbreeding) relates to the squared contributions of all ancestors (Woolliams & Thompson, 1994), not only the ones with largest marginal contributions. The value of knowing the most important marginal ancestors in our data is thus more of a cultural-historical one. The effective number of founder genomes (f_g) was calculated from the kinship in the reference population, which is sensitive to pedigree depth, while the rate of inbreeding from kinship was not. Assuming that F increases linearly with time, an

Table VII. Testing the existence of avoidance of inbreeding as a mating strategy from the difference between the average inbreeding coefficients of animals in the reference population and the average pair wise kinship between parents (f_{t-1}) (excluding selfing and intrasex mating) with a two-sample t -test.

	F_t	f_{t-1}	$ t $	p
Døle	0.1175 ± 0.0013	0.1115 ± 0.0003	4.46	0.01
Nordland/Lyngen	0.1277 ± 0.0011	0.1289 ± 0.0003	1.11	n.s.

Table VIII. The five ancestors with the largest marginal contribution to the reference population; in the Døle and in the Nordland/Lyngen, respectively.

Registration number	Name	Sex	Birth year	Marginal contribution
Døle				
1185DH	Gjestar	M	1919	0.21673
0825DH	Brimin	M	1908	0.15894
1752DH	Tuftar	M	1946	0.06595
0613DH	Draupner	M	1898	0.04514
0130DH	Dovre	M	1914	0.03684
Sum				0.52360
Nordland/Lyngen				
0002NH	Rimfakse	M	1935	0.26125
0010NH	Torgrim	M	1946	0.14834
00009N	Mona	F	1935	0.13819
00003N	Bruna	F	1930	0.12669
0009NH	Bamse	M	1945	0.06071
Sum				0.73518

interrelation exists between the two measures; $f_g \approx \frac{N_e}{t}$ (Meuwissen & Woolliams, 2000). The rate of inbreeding from kinship measures the genetic variation among the mated individuals, when ignoring the generation interval and assuming that all animals in the reference population contribute equally to the next generation. If the measure is calculated standardised, e.g. on basis of the last year classes that is not yet reproductive (here: the last three), consistent changes in size of annual values may indicate a loss of genetic variance, as observed in the Døle. Actually, the rate of inbreeding from kinship allows current rather than retrospective rates of inbreeding and effective population sizes to be calculated. Hence, the two inbreeding rates may complement each other when drawing inference with respect to loss of heterozygosity.

The level of inbreeding was high in both breeds, with an average that is equivalent to half-sib matings. However, the level of inbreeding was relatively higher in the Nordland/Lyngen where the base was only 7.3 generations away, compared with 10.5 generations in the Døle. To reduce inbreeding in Døle, crossing with trotters was as mentioned practised from the 1950s onwards. Actually, stallions of the Norwegian cold-blooded trotter were allowed to breed in the Døle, for the first time in 1953 (Gaustad, 1953), to cope with possible inbreeding depression, especially with respect to reduced fertility (Gaustad, 1951). During the 1970s the breeding policies changed, showed by the steep increase in inbreeding, probably because the use of trotters in the Døle produced lighter horses. As a consequence, the cold-blooded trotter was no longer preferred, and within 5–10 years the level of inbreeding was higher than ever. During the past decade the inbreeding coefficient has been around 12%. From

2000, a North-Swedish breed (Nordsvensk brukshäst) was temporarily used in the Døle to cope with this problem (Kvam, 2003), but this is not a good long-term solution. Instead, it will be more important to gain control over the selection with optimal contribution. This will minimise the average coancestry of parents and thus also the level of inbreeding of future generations (Sonesson & Meuwissen, 2001).

The Nordland/Lyngen has, as mentioned, had difficult periods during the past, where ensuring survival of the breed has been the priority. Back in the late 1930s three men initiated conservation of the Nordland/Lyngen (Roaldsøy, 1969). They looked for purebred individuals, and found one stallion in 1939; *Rimfakse*. He was used as the starting point for further breeding, contributing 26% to the reference population (Table VIII). The Nordland/Lyngen has had a relatively high level of inbreeding for many years, with no sudden changes during the past 40–50 years. According to Ehiobu et al. (1989) slower rates of inbreeding cause less inbreeding depression than more rapid rates of inbreeding. There are no recent reports of serious health damages in the two breeds, but such data are still not systematically recorded in these breeds. It would be important to record fertility data to survey possible reductions in fitness over time. This requires that the Norwegian Equine Centre has as a first priority to utilise their newly established database, and to begin to monitor these breeds by recording health traits.

Over the last generation there has not been any significant change in the level of inbreeding in either of the two populations, resulting in a larger effective population size in the Døle (152) than in the Nordland/Lyngen (42). During the 1990s, the rate of inbreeding from kinship indicates a likely reduction of genetic variance in the Døle (Table VI),

but with somewhat improved situation in the Nordland/Lyngen. These changes are a consequence of the lack of control of the general build-up of relationships in the population.

In Døle, the build-up of relationships in the population is due to selection (also not preferring cold-blooded trotters) as well as a mating strategy favouring inbreeding. In Nordland/Lyngen, the mating strategy is somewhat in favour of less inbreeding in the offspring, reflecting the breed's problems with inbreeding in the past.

Conclusion

In Nordland/Lyngen, the loss of heterozygosity has been considerable. The population is managed with a tendency towards avoidance of inbreeding in the offspring. In the study period heterozygosity has improved, most likely from improved selection and sire usage. In the Døle, the genetic variance has been higher, due to cross-breeding with cold-blooded trotters in the past. However, heterozygosity has become reduced due to active elimination of the cold-blooded impact, resulting in a mating strategy that actually favours inbreeding. In both populations we recommend that future management is carried out with optimal contribution selection (Sonesson & Meuwissen, 2001), with no need for additional cross-breeding.

Acknowledgements

Thanks to the Norwegian Equine Centre for help with completion and correction of the data material, to Morten Svendsen for useful help with programming, to Inger Anne Boman for carefully investigating the calculation of the inbreeding coefficients and to the referees of the article, with comments of great value.

References

Ballou, J. D. & Lacy, R. C. (1995). Identifying genetically important individuals for management of genetic variation in pedigreed populations. In J. D. Ballou, M. Gilpin and T. J. Foose (Eds.), *Population Management for Survival and Recovery*. New York: Columbia University Press, pp. 76–111.

Boichard, D., Maignel, L., & Verrier, É. (1997). The value of using probabilities of gene origin to measure genetic variability in a population. *Genetics, Selection, Evolution*, 29, 5–23.

Borchgrevink, O. (1906). *Stambok over heste av gudbrandsdalsk rase, bind 3* [Stud book for horses with Gudbrandsdal origin, volume 3]. Grøndahl & Søn, Oslo, 485 pp. (In Norwegian)

Caballero, A. & Toro, M. A. (2000). Interrelations between effective population size and other pedigree tools for the management of conserved populations. *Genetical Research (Cambridge)*, 75, 331–343.

Det kgl. selskab for Norges Vel. (1902). *Stambog over heste af gudbrandsdalsk rase, 1. bind* [Stud book for horses with

Gudbrandsdal origin, volume 1]. Grøndahl & Søn, Oslo, 153 pp. (In Norwegian)

Ehiobu, N. G., Goddard, M. E., & Taylor, J. F. (1989). Effect of rate of inbreeding on inbreeding depression in *Drosophila melanogaster*. *Theoretical and Applied Genetics*, 77, 123–127.

Falconer, D. S. & Mackay, T. F. C. (1996). *Introduction to Quantitative Genetics* (4th ed). Essex, UK: Longman Group, 464 pp.

Gaustad, M. (1951). Noen betraktninger om årets hingsteutstilling på Bjerke og om hesteavlen i sin alminnelighet [Considerations about the annual horse shows at Bjerke and about horse breeding in general]. In *Våre Hester Sommernummer*, Landbruksforlaget, Oslo, pp. 6–7. (In Norwegian)

Gaustad, M. (1953). Statens utstilling for dølehingster. Dølehestavlen. In S. Rohdin (ed.) *Våre Hester Julenummer* [Official horse show for stallions of the Døle breed], Landbruksforlaget, Oslo, pp. 14–17. (In Norwegian)

Henderson, C. R. (1976). A simple method for computing the inverse of a numerator relationship matrix used in prediction of breeding values. *Biometrics*, 32, 69–83.

Klemetsdal, G. (1998). A genetic study of the Norwegian cold-blooded trotter population. Dr. Agriculture Thesis, Department of animal science, Agricultural University of Norway, Ås.

Klemetsdal, G. (1999). Stochastic simulation of sire selection strategies in North-Swedish and Norwegian cold-blooded trotters. *Livestock Production Science*, 57, 219–229.

Kvam, T. (2003). Tiltak for å begrense innavlsøkning i dølehestpopulasjonen [Initiatives to limit the rate of inbreeding in the Døle horse]. In *Riksstambok over norske hester*. Norsk Hestesenter, Eds., Lena, Norsk Hestesenter, p. 7. (In Norwegian)

Lacy, R. C. (1989). Analysis of founder representation in pedigrees: Founder equivalents and founder genome equivalents. *Zoo Biology*, 8, 111–123.

Lacy, R. C. (1995). Clarification of terms and their use in the management of captive populations. *Zoo Biology*, 14, 565–578.

MacCluer, J. W., VandeBerg, J. L., Read, B., & Ryder, O. (1986). Pedigree analysis by computer simulation. *Zoo Biology*, 5, 147–160.

Meuwissen, T. H. E. & Woolliams, J. (2000). Genetic variance, gain and inbreeding in selection schemes. *Lecture Notes*, Horst, The Netherlands: IPC, 56 pp.

Norsk Hestesenter. (1988). *Årsmelding og regnskap 1988*. Norsk Hestesenter [Annual report and accounts 1988], Lena, Norsk Hestesenter, 38 pp. (In Norwegian)

Norsk Hestesenter. (2003). *Avlsplan for fjordhest* [Breeding plan for the Fjord horse]. Norsk Hestesenter, Lenar, Norsk Hestesenter, 35 pp. (In Norwegian)

Olsen, H. F., Klemetsdal, G., Ruane, J., & Helfjord, T. (2005). Use of probabilities of gene origin to describe genetic variation in two endangered Norwegian horse breeds. In *Conservation genetics of endangered horse breeds*. L. Bodó, B. Langlois and L. Alderson (eds.) EAAP Publication No. 116 Wageningen, Wageningen Academic Publishers, pp. 137–146.

Quaas, R. L. (1976). Computing the diagonal elements and inverse of a large numerator relationship matrix. *Biometrics*, 32, 949–953.

Rian, A. (2003). Kjente hingster. In O. Grøvdahl, G. Böhler, A. Rian and T. Boysen (eds.) *Rasehåndbok for Nordlandshest/Lyngshest* [Well-known stallions]. Landslaget for Nordlandshest/Lyngshest, pp. 39–48. (In Norwegian)

Roadsø, N. (1969). Nordlandshesten [The Nordland horse]. In *Stambok over norske hester*. Oslo: Statens Stambokkontor, pp. 241–254. (In Norwegian)

- Ruane, J. (2000). A framework for prioritizing domestic animal breeds for conservation purposes at the national level: A Norwegian case study. *Conservation Biology*, 14, 1385–1393.
- Seterlund, W., Rolstad, A., Kristiansen, O., & Flatla, L. (1989). Nye regler og rutiner for registrering og stambokføring av hest [New rules and routines when registering horses in the stud book]. Statens fagtjenester for landbruket, Oslo, 33 pp. (In Norwegian)
- Sonesson, A. K. & Meuwissen, T. H. E. (2001). Minimization of rate of inbreeding for small populations with overlapping generations. *Genetical Research (Cambridge)*, 77, 285–292.
- Statens Stambokkontor (1969). *Stambok over norske hester, 1. bind* [Stud book for the Norwegian horse breeds, volume 1] (Oslo: Statens Stambokkontor), pp. 231–309. (In Norwegian)
- Vangen, O. (1983). The use of relationship matrices to avoid inbreeding in small horse populations. *Z. Tierzüchtg Züchtbiol*, 100, 48–54.
- Vangen, O. (1996). Breeding goals and testing systems in Norwegian horse breeding. In *47th Annual Meeting of the European Association for Animal Production*, 26–29 August 1996, Wageningen Academic Publishers, Lillehammer, Norway.
- VanRaden, P. M. (1992). Accounting for inbreeding and crossbreeding in genetic evaluation of large populations. *Journal of Dairy Science*, 75, 3136–3144.
- Woolliams, J. & Thompson, R. (1994). A theory of genetic contributions. In *Proceedings of the Fifth World Congress on Genetics Applied to Livestock Production*. Guelph, Ontario, 7–12 August 1994. (vol. 19) Canada: University of Guelph, pp. 127–134.
- Zechner, P., Sölkner, J., Druml, T., Baumung, R., Achmann, R., Bodo, I., Marti, E., Habe, F., & Brem, G. (2002). Analysis of diversity and population structure in the Lipizzan horse breed based on pedigrees information. *Livestock Production Science*, 77, 137–146.