

Analysis of Inbreeding in a Closed Population of Crossbred Sheep

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(ABSTRACT)

Genetic diversity and the effect of lamb and dam inbreeding on multiple traits were analyzed in an 11-yr closed population of sheep established in 1983 and remained closed after 1987, with 50% Dorset, 25% Rambouillet, and 25% Finnsheep breeding to determine selection response for spring fertility. The population had been randomly divided in 1987 into a fall-lambing selection line (S) of 125 ewes and 10 rams, fall-lambing environmental control line (E) of 55 ewes and 5 rams, and a spring-lambing genetic control line (G) of 45 ewes and 5 rams. Inbreeding effects were estimated from 2678 lambs and 556 dams present after the creation of the respective lines. The traits assessed were ewe spring-fertility, lambing date, lamb birth, 60 d, and 120 d weight, and lamb survival to 1, 3, and 14 d. Genetic diversity was assessed by estimating change in inbreeding per generation (ΔF) and effective number of breeding animals (N_e), and parameters derived from gene drop simulations and an iterative procedure developed by Boichard et al. (1997); effective number of founders (f_e), effective number of ancestors (f_a), founder genome equivalents (f_g), and two additional measures of genetic diversity (GD_1 , GD_2). In order to estimate the diversity available in S and G, three sets of animals from the end of the study and one set of animals at line formation were considered in each line: all lambs born (including dead lambs), all matings (including potential offspring, even if a lamb was not born), and all rams and ewes available at the end of the study and at line formation.

At the time of line formation, most of the loss in diversity was due to unequal founder representation. The smaller population of G, as compared to S, caused a greater decrease in diversity due to bottlenecks at line formation. Very little diversity was lost due to additional drift by the time of line formation because selection had not occurred and a random mixing of founders was the goal. Allelic diversity decreased moderately; of the 322 founder alleles, there were 71% in S and 58% in G of rams and ewes (RE) that appeared in at least 50 runs of gene drop. By the end of the study in 1998, the amount of allelic diversity had decreased substantially. Of the alleles possible in RE at the end of the study in S and G, only 6 and 8 %, respectively, appeared in greater than 50 simulations of gene drop. The measures of f_e , f_a , and f_g revealed there was not much additional loss in diversity from the line founders to the end of the study due to unequal founder representation, but there was a larger amount of loss due to bottlenecks and additional drift. The diversity loss was similar, which was the goal of the selection study, when values for RE were compared in S and G.

The effects of lamb and dam inbreeding were estimated from REML analysis. Effects of lamb or dam inbreeding were negative but not significant for lambing date or survival to 1, 3, or 14 d. Spring fertility was estimated to decrease by 0.70 ± 0.30 %/% inbreeding of the ewe ($P < 0.01$), which seems even greater since the average spring fertility was only 47.5 %. Effects of

lamb inbreeding on birth, 60-d, and 120-d weights were -0.012 ± 0.006 ($P < 0.05$), -0.045 ± 0.020 ($P < 0.05$), and -0.130 ± 0.034 kg/% ($P < 0.01$), respectively. Dam inbreeding had smaller effects on birth, 60-d, and 120-d weights of -0.008 ± 0.010 (ns), -0.033 ± 0.034 (ns), and -0.087 ± 0.056 ($P < 0.1$) kg/%, respectively.

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INTRODUCTION

The unavoidable mating of related animals in a closed population leads to accumulation of inbreeding and decreased genetic diversity (Falconer and MacKay, 1996). Most programs involving domestic animals may try to minimize accumulation of inbreeding and quantify the increase by calculating the change in inbreeding per generation (ΔF) (Boichard et al., 1997). However, cumulative effects of inbreeding and drift, as well as change in allelic diversity and the time of diversity loss may not be reflected in ΔF . To better analyze the population, more descriptive measures derived by Lacy (1989, 1995) and Boichard et al. (1997) allow populations to be characterized with respect to founder effects, bottlenecks, genetic drift, and the most influential ancestors. Each generation these values can be estimated to identify and potentially correct the causes of losses in diversity. Important ancestors may also be evaluated for trait(s) under selection.

Heterozygosity and allelic diversity can be lost from small, closed, selected populations at a rapid rate. The loss of diversity and resulting increase in homozygosity may result in decreased production and/or fitness of inbred animals. This inbreeding depression has been well documented in many populations for a variety of traits (Lamberson and Thomas, 1984). It is apparent that different breeds and populations, as well as different traits vary in their response to inbreeding. Some populations may show a very pronounced effect of increased inbreeding for a trait, whereas others may not demonstrate much of an effect (Analla et al., 1998). It is important to account for effects of inbreeding in populations undergoing selection to properly adjust the breeding process for the potential reduction in performance.

Thus the objectives of this thesis were to 1) thoroughly describe the loss in heterozygosity and allelic diversity in closed selection and a control lines of crossbred sheep, 2) compare the two lines to show the impact of selection on diversity loss, and 3) to evaluate effects of lamb and ewe inbreeding on ewe spring-fertility, lamb survival to 1, 3, and 14 d, lamb weights at birth, 60 d, and 120 d, and fall lambing date.

CHAPTER I.

LITERATURE REVIEW

Defining Inbreeding

Inbreeding is the mating of related individuals, and results in some loci bearing alleles that are identical-by-descent (IBD). Identity by descent occurs because alleles from one common ancestor may flow through multiple offspring. When descendants of the common ancestor breed, copies of the same gene can be passed on to their offspring, who is said to be inbred. The proportion of genes that are IBD, typically designated as the coefficient of inbreeding, or F , can be calculated through knowledge of the pedigree or estimated by determining allele frequencies after detection of genetic polymorphisms. In order to estimate F , a base population must be established. Since the number of ancestors in a pedigree increases per generation by 2^n , where n is the number of generations, the pedigree increases exponentially and, eventually, all individuals are related. The base population is made up of founders, animals whose parents are assumed to be unknown and who are, therefore, treated as though they are unrelated and non-inbred (Falconer and Mackay, 1996).

Founders contain all the genetic variation available for transmission to their descendants. However, founders selected from a larger population only contain a portion of the genetic variability and heterozygosity in that population, even if the selection is random. Small founder numbers and small family sizes in later generations can cause an increase in homozygosity, including alleles that are IBD or identical-in-state (IIS) (i.e., identical alleles that have not descended from a common ancestor) due to inbreeding and random genetic drift. Random genetic drift is a process used to describe the likelihood an allele is lost by chance, causing a change in gene frequency. Large founder populations and/or large family sizes tend to maintain genetic diversity and, over time, may actually result in gains in genetic diversity and increased heterozygosity through mutation of common alleles into new unique alleles. Mutation and migration are the only processes to increase genetic diversity within a population (Lacy, 1989). The rate of inbreeding can change based on number of offspring, population size, and mating system, so it is customary to look at the change in inbreeding per generation, ΔF , for a more descriptive view of increased homozygosity.

It is important to acknowledge that the quantity F is a probability. The inbreeding coefficient is the probability that for any given locus, two alleles are identical by descent. It is also the percent of the genome that is, on average, IBD. Alleles can also be identical and thereby contribute to homozygosity without inbreeding (IIS) since there are not an infinite number of alleles for any one locus. Therefore, the amount of homozygosity is always greater than F , especially in populations or breeds that started with only a few animals, and for loci with only a small number of alleles.

Methods of measuring inbreeding

Individual inbreeding coefficients in pedigreed populations are calculated from the standard formula:

$$F_x = \sum \left(\frac{1}{2} \right)^n (1 + F_a)$$

where n is the number of animals in any path of relationship connecting the parents of individual X to a common ancestor, and where F_a is the inbreeding of the common ancestor. Inbreeding cannot be mentioned without thinking about the complementary measures, heterozygosity or allelic diversity. Maintenance of heterozygosity and allelic diversity present in the founding population is generally desired due to the detrimental effects of inbreeding depression on fitness. The anticipated decrease in heterozygosity associated with closed populations is usually expressed as the expected increase in homozygosity. By calculating F , a measure of the amount of genetic diversity that has been lost can be obtained. In order to measure inbreeding on a more constant scale, ΔF can be estimated by regressing individual inbreeding coefficients on generation number. The change in inbreeding per generation can then be used to estimate the effective number of breeding animals (N_e), where:

$$\Delta F = 1 / (2 N_e)$$

The purpose of the inbreeding effective number, N_e , is to estimate the number of animals that would produce the observed rate of inbreeding if bred under ideal conditions in the current generation (Lacy, 1995). Characteristics of an ideal population include: equal variance in family size, large number of breeders, random mating, equal sex ratio, absence of selection, mutation, or migration, and discrete generations (Falconer and Mackay, 1996). Deviations from these ideal conditions may be quantified by comparing N_e to the actual census number of breeding individuals in the population. Expressions exist to estimate the impact of unequal sex ratio, unequal family size, and genetic bottlenecks on N_e (Hallerman, 2003). Although N_e accounts for most genetic processes, when N_e is calculated from the average ΔF over all generations, it does not account for the loss in diversity due to drift from sampling selected parents.

To account for unequal founder representation, Lacy (1989) estimated the effective number of founders (f_e) as:

$$f_e = 1 / \sum_{i=1}^n p_i^2$$

where p_i is the expected proportional genetic contribution of founder i , calculated by the average relationship of the founder to each animal in the current population, and n is the total number of founders. The parameter f_e indicates the number of equally contributing founders that would produce the same level of genetic diversity as that observed in the current population. When random mating is practiced in a closed population and assuming there is no genetic drift, p_i 's will be very similar and f_e will approach a relatively constant value in a few generations, because founder contributions will remain the same. Selection, on the other hand, causes p_i 's between founders to fluctuate, if multiple traits are selected with superior alleles for each attributed to different ancestors, or to decrease, if there is only one ancestor with the best additive gene effect for the selected trait (Caballero and Toro, 2000). Even in the presence of selection, f_e will approach a constant value as long as new founders are not added to the population. A similar concept, GD_1 , is the measure of heterozygosity:

$$GD_1 = 1 - \frac{1}{2f_e} \approx 1 - \sum_{k=1}^{2n} q_k^2$$

where q_k is the frequency of founder allele k averaged over all individuals of the cohort and all replicates in a gene drop simulation and n is the number of founder animals (Rodríguez et al., 1998). Gene drop is a computer simulation procedure that is performed by giving each founder a

pair of unique alleles and randomly transmitting the alleles through the pedigree (MacCluer et al., 1986). The procedure simulates segregation of alleles and how some alleles will be lost by chance, while others are retained.

A concept similar to f_e was used by Boichard et al. (1997) to determine the effective number of ancestors (f_a):

$$f_a = 1 / \sum_{i=1}^m a_i^2$$

where a_i is the marginal contribution of each ancestor (i.e., any animal in the pedigree except for those animals in the current generation), as opposed to each founder, to the current generation, and m is the total number of contributing ancestors. The marginal contribution of all ancestors should sum to one, and the effective number of ancestors is always smaller than or equal to the effective number of founders. Individual contributions to the effective number of ancestors can be used to find the most influential ancestors. The numbers obtained from the calculation of f_a are essentially the same as f_e , but take into account the decrease in genetic variation in populations that have passed through a bottleneck. It is the animal that passes on the most genes that makes the highest contribution. Even though an influential ancestor (e.g. say a son of a founder) passes on most of his genes through many offspring, he only has half of the genes from his founding father. Animals in the current population under study are given a value of one and marginal contributions are obtained by processing the pedigree from youngest to oldest. When an important ancestor is identified (an animal with the most relationships to the current population), their sire and dam information is removed from the pedigree, so contributions to the current population are not double-counted. An algorithm must be created to rerun the calculations each time an ancestor is removed, so that marginal contributions not due to the ancestor already selected are the only ones measured (Boichard et al., 1997). From the previous example, if the son was selected as an important ancestor, the founding father would not get credit for his contributions through his influential son in the next iterate. There is a discrepancy that may occur when f_a is estimated through the previous equation. Since animals are selected based on their marginal contribution, if multiple animals have the same marginal contribution within one iteration, then the number of effective ancestors may change depending on which one is chosen. Large populations are not affected greatly, but in small populations there could be an increased effect on f_a , since marginal contributions have the potential to be larger. The f_a accounts for bottlenecks in the pedigree, but does not account for genetic drift. The calculation is useful for identifying the most influential ancestors, which may be of importance in selected populations.

Bottlenecks, unequal founder contributions, and genetic drift, which have a greater impact in small populations, can be quantified using the founder genome equivalent (f_g):

$$f_g = \sum_{i=1}^c (p_i^2 / r_i)$$

where r_i is the expected proportion of founder i 's alleles that remain in the current population and can take on a value of 0.5 if one allele is present or 1.0 if two alleles are present, and c is the

number of contributing founders (Lacy, 1989). The value for r_i weights the proportionate contribution of the founder by the number of ways the founders' alleles can be transmitted down the pedigree. The number of founder genomes shows the number of founders needed to create the same diversity in the current population, if they all contributed equally and no alleles were lost by genetic drift. The effective number of genomes is always less than N_e , and also less than f_e and f_a . The estimate of f_g from the above equation must be calculated using gene drop analysis. Multiple simulations give a proportion of founder alleles in the current population that account for all deviations from an ideal population. Lacy (1995) showed that the relationship between the current population's mean coancestry and f_g was:

$$f_g = 1 / (2 \bar{c})$$

where \bar{c} is half the average relationship between all individuals including the relationship of each individual to itself in the current population. The value shows that as the population becomes more related, as would happen with any closed population, f_g decreases. The formula derived by Lacy (1995) is easily calculated from the relationship matrix. Founder genomes do not account for mutation or migration, and, if mutations occur, a slightly higher amount of heterozygosity could be present. Eventually, f_g would go to one-half, showing only one allele present at every locus. To equate the increase in homozygosity to a loss in heterozygosity, Rodríguez et al. (1998) defined the following measure of genetic diversity:

$$GD_2 = 1 - \sum_{j=1}^t \left[\sum_{k=1}^{2n} \bar{q}_{kj}^2 \right] / t$$

where \bar{q}_{kj} is the frequency of founder gene k for replicate j , t is the number of replicates, and n is the number of founders. The measure of genetic diversity is also calculated through gene drop, but can be determined using coancestry. The second term of the equation is simply the mean coancestry of the current population.

The previous measures can be used to determine loss of genetic diversity in a variety of ways. These values can be used to provide information on recent changes in breeding structure in natural and domestic populations that may pose a problem to maintaining genetic variation (Boichard et al., 1997).

Application of alternative measurements of inbreeding and relationship

Boichard et al. (1997) used the measurements derived by Lacy (1989) and created a new method to estimate f_a to identify important ancestors and to estimate the effective number of ancestors. The authors demonstrated how the measurements are calculated and applied them to three French cattle populations and one simulated population. The authors also analyzed the effect of missing pedigree information on the simulation calculations. They found that when 10% of male and 10% of female parental information was removed, the values for N_e , f_e , f_a , and f_g increased. When 20% of female information was removed, there was an even greater overestimation of these measurements. With missing information, divergence between the

estimates and the true values in a complete pedigree increased as the number of generations from the base population also increased.

The three breeds of cattle that were analyzed in the study were Abondance, a relatively small population (106,520 animals in the pedigree); Normande, a relatively large population (2,338,305 animals in the pedigree); and Limousin, a moderate-sized population (919,561 animals in the pedigree) that has less artificial insemination (AI) than the other two breeds. The pedigrees for all three breeds were incomplete, with the Abondance breed being the least complete. The population of study was defined as females born in a 3-yr period with known sires and dams. Normande cattle had the largest increase in inbreeding in the last generation (1.07%) as compared to Abondance (0.47%) and Limousin (-0.05%), which actually saw a decrease in inbreeding, perhaps due to grade-up animals being registered and limited AI use. The measures of f_e , f_a , and f_g better quantified the diversity in the Abondance (69, 25, and 17), Normande (132, 40, and 22), and Limousin (790, 360, and 206) breeds than measures of ΔF . The large impact of AI on a population can be seen when comparing the Limousin to the other two breeds. The Normande breed had values close to that of the Abondance even though the population size was almost 22 times larger. The reason for this could be attributed to increased selection intensity in sires, shorter generation intervals, and a longer pedigree with more generations in the Normande cattle. An f_g of at least 20 would be adequate, if mated randomly, to obtain 97.5% of the variation present from the original population in which the animals were sampled (Lacy, 1989).

Rodríguez et al. (1998) investigated diversity measures in a population of Large White pigs maintained as a closed herd for 55 yr and selected based on visual appraisal to preserve phenotypic variability. Fourteen animals made up the base population, with equal numbers of each sex. Animals from all years were divided into 19 cohorts according to their year of birth, and the inbreeding in the last cohort averaged 41.2%. The population was divided into four lines at the start and bred within these subpopulations. In 1956, the groups were mixed and matings were made between animals with minimum relationships. Between the founders and the next cohort, the f_e dropped to 9.76, indicating unequal representation of the founders, and the f_e value leveled out at 9.2 within the next few generations. Founder genome equivalents decreased from 7.51 in the first cohort to 1.80 in the last cohort. The decrease was relatively small in the last nine cohorts, indicating that the breeding schemes were effective in maintaining remaining diversity. A new parameter was determined using the r_i values obtained from the gene drop simulation, to calculate expected number of founder alleles surviving from each founder, as $2r_i$, and from all founders as $\sum 2r_i$. The first cohort had 20.19 founder alleles. This value decreased to 3.63 in the last cohort, which corresponds to 13% of the founder alleles. The pig population experienced most of the decrease in diversity within the first few years due to unequal founder representation and genetic drift, but the population seemed to be relatively stable thereafter, with only small decreases in heterozygosity and allelic diversity due to the small population size.

Effects of inbreeding in sheep

Inbreeding has been shown to adversely affect many mammalian populations (Ercanbrack and Knight, 1981, 1991; Lamberson et al., 1982; Lamberson and Thomas, 1984; Rodríguez et al., 1998; Mandal et al., 2002). Originally, it was believed that inbred lines may

produce animals that are superior for a trait that is under selection. Different lines could then be crossed to produce offspring with greater merit than either of their parents (Ercanbrack and Knight, 1981; Lamberson and Thomas, 1984). However, Ercanbrack and Knight (1981) found that inbred lines selected for a certain trait sometimes equaled, but often were inferior to non-inbred selected lines. Many studies have found that traits of low heritability are more adversely affected by inbreeding than highly heritable traits, which are easily improved through selection upon additive genetic effects (Ercanbrack and Knight, 1981; Lamberson and Thomas, 1984). Commonly, negative inbreeding effects, or inbreeding depression, are thought to most frequently occur because of an increase in frequencies of recessive alleles that adversely affect the traits of interest. The increased frequency of recessive alleles leads to a larger number of individuals that are homozygous for the recessive alleles, whereas in non-inbred populations, the recessive allele would more frequently be masked by an advantageous dominant allele (Lamberson and Thomas, 1984; McDaniel, 2001). The degree to which inbreeding depression occurs is dependent on how many deleterious alleles are present in the founder population. Highly heritable traits are usually primarily affected by additive genetic effects, whereas dominance effects are the main contributor to genetic variation in less heritable traits (Falconer and MacKay, 1996). Even though low heritability traits are usually most directly affected by inbreeding, rates of improvement in highly heritable traits also can decrease due to a loss in reproductive performance, resulting in fewer animals available for selection (Ercanbrack and Knight, 1981, 1991; Lamberson et al., 1982). Inbreeding, although once thought to be a useful tool to improve sheep breeds by increasing the frequency of desired alleles, was shown to cause economic losses of \$17.00/ewe when dam inbreeding is 20% and lamb inbreeding is 25% (Ercanbrack and Knight, 1991). The potential for great losses indicates the importance of finding the effects of inbreeding.

Although inbreeding depression is usually thought to be the result of an increased frequency of deleterious recessive alleles, there are alternative hypotheses. The overdominance hypothesis regards inbreeding depression as the result of losses in heterozygosity per se, not because of the increased homozygosity for deleterious recessive alleles. The hypothesis is sometimes invoked to explain the observed advantages of crossbreeding and hybrid vigor. There are not many well designed studies to prove the overdominance hypothesis. A higher level of heterozygosity and allelic diversity proving advantageous is apparent in regards to the major histocompatibility locus (O'Brien et al., 1985). More generally, increased variability may give an animal more ability to adapt to a changing environment (Ralls et al., 1986). The effect may have a greater impact on natural populations than on domestic populations because of human management.

Growth traits

Birth weight. An overview of most studies before 1984 on inbreeding in sheep found that, on average, there was a decrease in birth weight (BW) of 0.013 kg/% increase in lamb inbreeding in multiple flocks of Ossimi, Hampshire, Merino, and No-tail sheep breeds (Lamberson and Thomas, 1984). The lamb inbreeding effect ranged from 0.0022 to -0.029 kg / %F. The only significant effects were reported in a Merino flock with an effect of -0.015 kg / %F. Average inbreeding in this Merino flock was only 1.8%, which may not permit accurate extrapolation to higher levels of inbreeding. Inbreeding of the dam decreased BW by an average of 0.013 kg /

%F, with a range of 0.001 to -0.020 kg / %F in these studies. Significant effects of inbreeding of the dam were noted in the Merino (-0.016 kg / %F) and No-tail (-0.020 kg / %F) breeds (Lamberson and Thomas, 1984). Ercanbrack and Knight (1991) showed a similar decrease in BW in Rambouillet, Targhee, and Columbia sheep due to inbreeding of the lamb (-0.008 ± 0.002 , -0.014 ± 0.002 , and -0.002 ± 0.003 kg / %F, respectively) and a smaller decrease due to inbreeding of the dam (-0.004 ± 0.001 , -0.001 ± 0.002 , and 0.001 ± 0.003 kg / %F, respectively). Rambouillet lines had been outcrossed periodically to decrease inbreeding, which may increase the effects of future inbreeding by introducing new undesired alleles. Targhee lines were a few generations younger than Rambouillet and Columbia lines when measurements were taken, so assuming that natural and artificial selection decreases frequencies of deleterious alleles, these lines have had less time for their removal.

Weiner et al. (1992) found that BW in hill sheep (Scottish blackface, Cheviot, and Welsh mountain) breeds and their crosses decreased due to lamb inbreeding up to an inbreeding level of 37.5%, and then positive effects were seen at inbreeding levels of 37.5% to 59%. The positive effects seen at higher levels of inbreeding may have been due to purging of deleterious alleles in previous generations. At an inbreeding level of 12.5%, the regression coefficient of BW on F in this study was -0.016 kg / %F. The effects were not fitted with quadratic or cubic terms, so only a slope at one point was given and may be compared only to results found in other studies at the same level of inbreeding. This study is one of the few to estimate performance effects of high levels of rapid inbreeding. Dam inbreeding had a significant linear effect of -0.0057 kg / %F, which is near the average of the values found in previous studies.

Boujenane and Chami (1997) reported contrasting results in one of two Moroccan sheep breeds. Sardi sheep showed no significant effects of lamb inbreeding (0.0001 kg / %F) and a significant negative effect of dam inbreeding (-0.0053 kg / %F). The average inbreeding of lambs was 8.07% with a maximum of 32%, so a low level of inbreeding cannot explain these results. This breed may have few undesirable genes for BW, in association with their small genetic base. The Beni Guil breed had effects similar to those seen in other breeds, with declines of -0.0061 kg / %F of the lamb and -0.0222 kg / %F of the dam. The Spanish Merino also showed effects for lamb inbreeding that were similar to previous results: -0.011 kg / %F (Analla et al., 1998). Analla et al. (1999) also tested linear and quadratic effects of inbreeding of the lamb on BW in the Merino breed and found that both were significant with linear and quadratic regression coefficients of -0.017 and 0.010 kg / %F, respectively. Significant effects of inbreeding of the lamb (-0.010 ± 0.003 kg / %F) in an Indian breed of sheep, Muzaffarnagari, were reported by Mandal et al. (2002). Effects of dam inbreeding on BW were not significant in this study. Causes of variation in inbreeding effects could be due to differences among breeds in alleles segregating, amount of genetic variation in the base population, location, management, and diversity of the founders in the flocks tested. Across studies, there is a greater effect of inbreeding of the lamb on BW, averaging -0.0112 kg / %F, than of inbreeding of the dam, averaging -0.00815 kg / %F.

Weaning weight. Weight at weaning also tends to be more affected by the inbreeding of the lamb than the inbreeding of the dam. Lamberson and Thomas (1984) found average effects of lamb inbreeding on weaning weight (WW) from all studies of Rambouillet, Whiteface, Ossimi, Hampshire, Merino, No-tail, Targhee, and Columbia of -0.111 kg / %F; dam effects

were $-0.072 \text{ kg} / \%F$. The significant effects for lamb and dam inbreeding averaged -0.131 and $-0.107 \text{ kg} / \%F$, respectively. The range for lamb and dam inbreeding was 0.036 to $-0.177 \text{ kg} / \%F$ and 0.000 to $-0.107 \text{ kg} / \%F$, respectively. Results of this summary tended to indicate a greater effect on single births than on twins, and on females than males, but these values were not tested for significance. Ercanbrack and Knight (1991) found that WW decreased significantly (by -0.114 , -0.116 , and $-0.087 \text{ kg} / \%F$ in Rambouillet, Targhee, and Columbia sheep) due to inbreeding of the lamb. The Columbia breed had a smaller effect of inbreeding on weight traits, which may be due to fewer alleles present that would decrease weight. The effect of inbreeding of the dam on WW was only significant in Rambouillet ewes ($-0.033 \text{ kg} / \%F$), and was smaller than that found in other studies. Both values for the other two breeds were also negative.

A non-linear trend in effects of inbreeding on WW in hill sheep breeds was presented in a study by Weiner et al. (1992). There was a negative effect for inbreeding of the lamb at inbreeding coefficients less than or equal to 37.5%, and then positive effects for inbreeding coefficients between 37.5% and 59%. At lamb inbreeding of 12.5%, the regression coefficient was -0.073 , a little lower than values found previously. The effect of the dam inbreeding on WW was significant ($-0.0764 \text{ kg} / \%F$) and relatively linear, except for a slight increase at F of 50% to 59%. Boujenane and Chami (1997) found non-significant effects of lamb inbreeding ($0.0056 \text{ kg} / \%F$) and significant negative effects of inbreeding of the dam ($-0.0364 \text{ kg} / \%F$) in the Sardi breed. The Beni Guil had significant negative effects due to the lamb inbreeding ($-0.0469 \text{ kg} / \%F$) and non-significant effects of dam inbreeding ($-0.0168 \text{ kg} / \%F$). The slightly smaller result may be due to earlier weaning, and to the lower mature weight of this breed. Spanish Merinos had a similar reduction due to lamb inbreeding in two studies (-0.076 and $-0.065 \text{ kg} / \%F$) (Analla et al., 1998, 1999). When a quadratic effect was added to the model, it was not significant ($-0.120 \text{ kg} / \%F^2$); the linear effect was significant in the presence of the quadratic and was more negative ($-0.111 \text{ kg} / \%F$). Mandal et al. (2002) reported that at 3 mo of age (around weaning), the effect of lamb inbreeding on WW was $-0.046 \pm 0.020 \text{ kg} / \%F$ in Muzaffarnagari sheep, which was less than the effects found in other sheep breeds. Dam effects were not significantly different from zero. Inbreeding of the lamb thus had a greater effect on the weight of the animal than the inbreeding of its dam, but breed variation in effects of inbreeding still exist.

Fitness Traits

Low heritability of fitness traits and high impact on economic traits make it important to consider inbreeding effects, since fitness and production related performance traits usually decline with increased inbreeding. Fertility, prolificacy, and lamb viability, have been shown to decrease significantly with inbreeding of the dam and/or lamb in individual studies and breeds. The effect of lamb inbreeding was usually found to have a greater effect than dam inbreeding on lamb survival (Lamberson et al., 1982; Lamberson and Thomas, 1984; Ercanbrack and Knight, 1991). The effect of inbreeding on reproduction has an obvious impact through fertility, pregnancy, and lambing, resulting in fewer lambs per flock.

Fertility. Ewe fertility is a very important factor in sheep operations. Improvement of fertility through selection can be difficult, since it has low heritability. Lamberson et al. (1982)

found a decrease in fertility of 1.2 %/F in Hampshire ewes. Lamb inbreeding may affect the apparent fertility of the ewe by decreasing lamb survival in-utero, but effects were not significant in this study (-0.3 %/F). Ercanbrack and Knight (1991) found significant effects of lamb inbreeding in the Targhee breed (-0.301 %/F), but values were not significant in the Rambouillet and Columbia breeds (-0.001 and -0.017 %/F, respectively). Dam inbreeding effects were significant in Rambouillet ewes (-0.230 %/F), which is one-sixth of the value reported previously in Hampshires, but not in Targhee and Columbia ewes (-0.012 and -0.088 %/F, respectively). Although there are inconsistent data to determine whether inbreeding of lamb or of dam has a greater effect on fertility, it is clear that inbreeding generally decreases the fertility of matings. Further studies in other breeds may be useful to help quantify any effect of inbreeding on fertility.

Prolificacy. The number of lambs produced per ewe lambing was not significantly affected by inbreeding of lamb or dam in Hampshire sheep (Lamberson et al., 1982). In another study, lamb inbreeding (-0.00292 lambs/F) and dam inbreeding (-0.00385 lambs/F) significantly affected prolificacy in the Rambouillet breed (Ercanbrack and Knight, 1991). Significant effects of dam inbreeding also were noted in the Targhee breed (-0.00476 lambs/F) with non-significant effects due to the lamb inbreeding. Columbia sheep did not show any significant effects of inbreeding on prolificacy. The increased effects seen in Rambouillet ewes may be due to higher variability in lamb numbers or a different genetic base for the breed. In the Sardi and Beni Guil breeds, lamb inbreeding (0.0007 and -0.0018 lambs/F) and dam inbreeding (0.0004 and -0.0035 lambs/F) did not have significant effects on the number born (Boujenane and Chami, 1997). Inbreeding of lambs did not affect prolificacy in Spanish Merinos in two studies (-0.002 and -0.004 lambs/F) (Analla et al., 1998, 1999). Although inbreeding does not seem to significantly affect prolificacy in sheep, which may be due to the lower number of lambs per litter in the sheep breeds studied, significant effects have been reported in swine (Rodrigañez et al., 1998). In a closed population of Large White pigs litter size decreased due to inbreeding of the sow (-0.02 piglets/F) and litter (-0.03 piglets/F), but only inbreeding of the litter was found to be significant. Most studies on sheep have found that fertility is negatively affected by inbreeding, but prolificacy is not. It seems that as long as the ewe becomes pregnant, the viability of the fetus and the number of ovulations are not significantly affected.

Lamb survival. Lamberson and Thomas (1984) reviewed eight studies and found that lamb survival to 90 d decreased with inbreeding. The average effects of lamb and dam inbreeding were -0.0278 and -0.0124 lambs / F, respectively. Hampshires and Merinos had significant negative effects due to lamb inbreeding (-0.013 and -0.017 lambs / F, respectively); all other values for other breeds and studies were negative but not significant. Effects of dam inbreeding varied from 0.055 to -0.12 lambs / F, none of which were significant. Some studies have seen increased survival with highly inbred dams (Lamberson et al., 1982). Galal et al. (1981) studied the effect on lamb survival of varying levels of inbreeding in two Egyptian sheep breeds and in Merinos and Merino crosses. Lambs and dams were grouped by their inbreeding coefficient in intervals of 5%. Inbreeding of the lamb had negative effects for ages 7 to 180 d for inbreeding coefficients of 5 to 15% and 20 to 25%. Lambs with inbreeding coefficients between 15 and 20% and $\geq 25\%$ had positive effects of inbreeding on survival for all ages. Lambs with inbreeding coefficients below 5% showed varying effects of inbreeding that were positive for 7,

30, and 180 d, but negative for survival at 120 d. Values at 120 and 180 d were not significant. Dam inbreeding effects were not significant for any inbreeding group or age, but tended to be negative with increasing inbreeding. This study did not show regression coefficients or tests of significance, but variation in effects at different levels of inbreeding is important and deserves further analysis. The increased survival of more highly inbred lambs may be due to elimination of less fit animals by natural selection at lower inbreeding. Ercanbrack and Knight (1991) found similar significant effects of lamb inbreeding on survival to weaning in Rambouillet, Targhee, and Columbia breeds (-0.00241, -0.00458, and -0.00444 lambs/%F). The study showed negative effects of dam inbreeding in all breeds, but the effect was significant only for Rambouillet ewes (-0.00332 lambs/%F). A study that measured inbreeding effects in the two breeds of Moroccan sheep found very small, mostly positive and no significant effects of dam and lamb inbreeding on survival (Boujenane and Chami, 1997). Overall, there is a large variation in lamb survival due to the effect of inbreeding, perhaps resulting from the many causes of lamb death, inaccurate record keeping, or real differences among breeds, lines, or environments. Most studies measured survival to 90 d, whereas a better analysis would look at survival within the first few days or weeks of life since this is when most genetic death occurs (Templeton and Read, 1984).

Wool traits

Twelve-month fleece weight. A review by Lamberson et al. (1984) showed little effect of inbreeding on fleece weight, but effects were more prominent in Merinos than in other range sheep. The average decrease in greasy fleece weight was 0.017 kg/%F of the lamb, with a range of -0.006 to -0.029 kg/%F. Similar results were found by Ercanbrack and Knight (1991) in Rambouillet, Targhee, and Columbia breeds with an average effect of -0.0127 kg/%F. Mandal et al. (2002) showed smaller effects in an Indian coarse-wool sheep breed, -0.0029 kg/%F for ewe inbreeding and non-significant values due to lamb inbreeding. Inbreeding appears to have a small effect on 12-mo wool yield, but in highly inbred sheep, the effect could become significant.

Other wool traits. Lamberson et al. (1984) reviewed several studies on staple length over various days of growth and sheep ages and found that the average reduction due to inbreeding of the lamb was -0.008 cm/%F. The effect was more prominent in range sheep than in Merinos. Dam inbreeding resulted in an increase in staple length, by an average of 0.002 cm/%F, but none of the values were significant. Fiber diameter, fibres/mm, and crimp/cm showed variable effects of lamb or dam inbreeding, and none of the results were significantly different from zero. It seems that inbreeding does not have a large effect on the quality of the fleece, but may slightly decrease fleece weight.

Managing Inbreeding

The undesired effects of inbreeding usually lead breeders to try to avoid matings among relatives. There are problems that can occur as a result of avoiding inbreeding, including rapid accumulation of inbreeding in later generations and submaximal selection response. To maintain a selected group of animals with constant or increasing production and fitness, inbreeding must be controlled and a specific time period has to be established in which ΔF is to be held below a threshold level. The target time period is relevant because inbreeding cannot be avoided or

limited indefinitely, especially in small populations, and at some point all animals will be closely related and inbreeding will inevitably increase. Mutation can make up for some of the loss in heterozygosity, but this comes at a very slow pace. Inbreeding is always accumulating, but the rate of inbreeding needs to be limited in order to maintain diversity at an acceptable level so that genetic variation will still be present and animals can respond to changes in environment and to selection. Without genetic variation, animals cannot adapt to these changes.

An additional concern is depression in fitness due to outcrossing. Some populations form coadapted gene complexes through selection that provides a benefit or high fitness in the environment in which they reside. These coadapted gene complexes have remained intact because of continued selective pressure and low migration into the population. When animals from different populations that would not normally come in contact with one another breed, they can produce offspring in which coadapted gene complexes are disrupted resulting in a decreased fitness level (Templeton and Read, 1984). Outbreeding depression is usually first observed in the second generation of animals, because offspring of the first mating have coadapted gene complexes still intact (i.e., recombination has not occurred) and hybrid vigor results in increased fitness. Outbreeding depression can sometimes be confused with inbreeding depression if inbreeding occurs in the first few years of matings of animals from different areas. In order to determine what type of depression is being expressed, the change in performance over increased 'mixing' of the different local populations (i.e., contributions of founders from different locations) at the same level of inbreeding can be analyzed. A decrease in performance with increased founder contribution indicates that outbreeding depression has caused at least part of the decrease in performance (Templeton and Read, 1984).

Maintaining genetic gains

The process of selection in livestock has become relatively straightforward and more efficient due to genetic prediction methods, such as BLUP (best linear unbiased prediction). BLUP can be used to obtain estimated breeding values (EBV), a prediction of the additive genetic merit of an animal, using a combination of individual and family information. Due to the use of family information, groups of relatives often have similar EBV's and therefore tend to be selected or culled as a group, thereby increasing relatedness among the breed. In addition to the use of BLUP, high selection intensities, embryo transfer, and AI also have increased relatedness within some breeds by reducing the effective number of parents. The problem has become apparent in Holstein cattle, when it was found that of 2500 widely-used bulls, only three Y chromosomes were found with only 46 maternal grandsires (McDaniel, 2001). The expected result of high selection intensity is high homozygosity, resulting in decreased future selection response and decreased fitness (Weigel, 2001).

The most studied method for management of inbreeding in livestock populations has been to attempt to maximize selection response at an acceptable level of ΔF (Weigel, 2001; Klieve et al., 1994; Quinton et al., 1992). An increase of less than 1% F per generation is usually deemed acceptable for most populations, but the magnitude of ΔF is usually greater than this in most simulation studies (McDaniel, 2001; Quinton et al., 1992). Klieve et al. (1994) studied animal selection based on BLUP or phenotype and at mating strategies that could be used to minimize inbreeding. These were compared to random mating with heterozygosity decreasing in

varying amounts due to inbreeding. They found that there was little decrease in EBV's after 20 generations from BLUP selection with random mating or from mate selection in the presence of up to a 10% decrease in attainable genetic response due to inbreeding (MS10). The average inbreeding was significantly reduced in the 20-generation simulation (42.25% for BLUP with random mating versus 32.68% for MS10). The values were similar but less drastic at one and five generations, and a slight decrease in EBV for mate selection was found with a 25% decrease in attainable genetic response at a decreased level of inbreeding. The study only used a small population and moderate heritability ($h^2 = 0.20$), which may result in increased effects as compared with larger populations and higher heritabilities. Mate selection to minimize inbreeding is applicable to other breeding programs, and the study showed that constraints on inbreeding can be used without greatly affecting genetic gains.

It is obvious that the number of parents and their relatedness impact the rate of inbreeding per generation. Quinton et al. (1992) compared selection methods at the same level of inbreeding. The reduction in genetic variance and response due to inbreeding was accounted for in the model but inbreeding depression was not. The number of sires used included one and 2 to 40 by intervals of two; with selection using phenotypes or BLUP was simulated. Results of the study showed that for an acceptable ΔF of 1% per generation, phenotypic selection, with 16 of 100 sires and 50 of 100 dams at h^2 of 0.25 for 10 generations had a greater selection response (2.33 versus 2.21 SD) with half the sires (8) used in BLUP selection. As expected, BLUP selection gives the greatest response with relatively few sires (6 sires, 2.99 SD of genetic response), but at an inbreeding level of 3.5% per generation. In order to maintain genetic variation for future selection response, inbreeding must be reduced and BLUP selection may not be the optimum method to achieve the maximum response at a fixed level of ΔF .

Weigel (2001) reviewed multiple studies summarizing current information on control of inbreeding in livestock programs. Simulations indicated that reductions in family size and increased number of selected males and females decreased inbreeding with only small reductions in genetic gain. The creation of sublines was discussed, suggesting that complete isolation was ineffective, but inbreeding was decreased when sublines were crossed in multiple years, similar to movement of animals each year or every few years. The impact of different mating programs on inbreeding was substantial. Many studies showed that computerized mate selection decreased inbreeding with only small decreases in genetic gain, but with increases in lifetime profit. These programs tended to decrease inbreeding in the next generation, but may not decrease inbreeding over many generations.

The addition of inbreeding information to BLUP selection and reduction of emphasis on family merit in EBV calculations (by artificially increased heritability to increase the weighting of the animals' records) increased long term genetic response with a lower ΔF and minimal loss of selection response. A problem with the simulation studies is that all pedigree information is assumed to be known. Some studies have addressed this issue, assuming animals with unknown parents are as inbred as the average inbreeding of similarly-aged animals (Boichard et al., 1997). The assumptions tend to work well for populations in which less than 20% of the pedigree information is missing. The main problem with methods to reduce inbreeding is that breeders may not realize an immediate advantage. They may favor short-term increase in profits through increased selection intensity over long-term increases in genetic variation and response to

selection. Once breeders can be convinced of the importance of controlling inbreeding, then long-term improvements may be attainable.

Founder contributions

The main goal in most captive wildlife populations is to maintain the highest level of genetic diversity, whereas this is not the case in domestic populations. The rate of inbreeding is usually quantified by N_e , but this does not give an accurate description of the level of homozygosity over time in populations that are not ideal. The new methods used by Lacy (1989), including f_c and f_g , give a more accurate description of the amount of diversity left in a population. Lacy (1995) stated that to maximize heterozygosity in future generations, animals with the lowest mean kinship (MK) or relationship to the population should be preferentially bred. Simulations show that this method of breeding is more beneficial than breeding animals to produce the lowest inbreeding. Livestock populations can use such information in selection decisions by breeding animals with the lowest MK within the selected group of parents. Gene drop techniques, developed by MacCluer et al. (1986), are useful in determining genetic measurements, in addition to determining which founders are the most influential and quantifying their contribution. The application of these techniques in domestic populations allows the determination of the most influential ancestors, but also allows assessment of founders that may have had an adverse effect on the population by contributing undesired alleles (Rodrigañez et al., 1998). Concepts used by wildlife managers to measure changes in genetic diversity could have an important use in livestock to determine not only diversity, but also founder effects and ancestor contributions.

The information gathered on the effective numbers of founders, ancestors, and genomes can be used in domestic populations to determine changes in breeding structure before the change is expressed in ΔF . Boichard et al. (1997) studied three French cattle breeds and found that the Abondance breed had an increase in ΔF of only 0.47% in the last generation, but the changes in f_c (-16%), f_a (-48%), and f_g (-55%) were larger, documenting effects of changes in breeding structure on the population. Observed changes were mostly due to bottlenecks and were apparent from the large change in f_a . The larger population of the Normande had in a smaller decrease in f_c (-2.6%). The decline for f_a (-28%) and the difference between f_c and f_a showed that the breed had been experiencing a significant effect from bottlenecks that probably resulted from high selection intensity on sires and use of AI. The population experienced a large amount of drift as well, apparent by the 31% difference between f_a and f_g (-59%). The Limousin breed, with limited use of AI, had smaller decreases in allelic diversity than the other two breeds, as was apparent in the decreases in f_c (-1.7%), f_a (-3.4%), and f_g (-8.1%). Most of the reduction was observed in f_g , indicating that drift was the major cause of diversity loss. In a population that has limited use of AI, there are usually relatively small family sizes for both sexes. All the genes in the parents cannot be passed to the offspring, as they can in sires used by AI, with hundreds of offspring. These calculations reveal why a population is losing diversity, and, since information is available before inbreeding is actually expressed, there may still be time to recover some of the losses while older, less related animals are still present.

Roughsedge et al. (1999) used the concepts of Lacy (1989) and Boichard et al. (1997) to determine the contribution of the American Holstein to the U.K. Holstein-Friesian population.

Random samples of 2000 cows from specific years were taken and ancestry traced from the five million animals in the U.K. database, which included imported cows with three generations of their pedigree, if available. There were 10 samples from 1997 births and four additional samples taken at 5-year (one generation) intervals prior to the 1997 sample. The average inbreeding in 1997 was $0.43 \pm 0.057\%$ with a base population of 1960. Inbreeding in 1960 was estimated to be around 2.28% according to additional pedigree information, which would increase the value for 1997. The trend since 1967 was a rapid increase in inbreeding from 1967 (0.05%) to 1972 (0.55%) and 1977 (0.82%), then a decrease due to importation of foreign sires until 1992 (0.38%) and a slight increase in 1997 (0.43%), when there were more U.S. Holstein genes in the population.

Since the population is open, both non-registered U.K. and imported U.S. animals can become founders, so f_e and f_a have not stabilized. Even with increased numbers of founders, f_e and f_a continued to decrease from 1967 to 1997, from 1353 to 93 for f_e , and from 702 to 93 for f_a , respectively. The f_e decreased by at least 30% between all periods except 1982 to 1987. The reason for these large reductions is probably because grade-up females had significantly smaller representation compared to sires used in AI. The f_a had decreases of greater than 30% in three of the six time periods. For the other three periods, decreases in values of f_a were relatively low between 1977 and 1992 (3.4, 3.6, and 12%, respectively), in contrast to values of f_e for these years. The difference in values showed the highly-contributing animals for these years were not the founders, and the important contributing ancestors were more equally represented. The percentage of female and male British genes in the population decreased from 72% and 24%, respectively, in 1967, to only 13% and 11% in 1997. The large increase in foreign animals will soon make the U.K. population essentially the same as the U.S. population. The usefulness of f_e and f_a is evident by providing a better visualization of genetic losses in the population, even though the inbreeding decreased through the 1980's and early 1990's.

Rodríguez et al. (1998) used the transmission of founder alleles in pigs in a unique way to determine each founder's contribution and its effect on the population. They found that alleles from two sows and two boars of 14 founders were virtually eliminated by the last generation of the study (<0.033 alleles surviving for each of the four founders). They also determined that the four lowly contributing founders' alleles were reduced to very low levels (<0.05) between 1958 and 1960 for three of the founders, and between 1973 and 1975 for one sow. Their extinction occurred about midway through the population's existence when four subdivided lines were mixed, which probably is an indication of a small bottleneck at this point. Three of the 3.63 alleles expected in the last cohort were from seven of the 14 founders. Knowing who the founders were could be beneficial if frozen semen or ova were available and could be used to bring back under-represented animals.

Regression analysis was used by Rodríguez et al. (1998) to determine whether founder alleles contributed to inbreeding depression. Five founder lineages had a correlation between the partial inbreeding coefficient of the litter due to a founder and inbreeding depression on litter size of at least 0.60. These five lineages corresponded to the four subdivided populations; the other lineage was a sow introduced in 1935, lineage E. Lineage B had a positive but non-significant effect of inbreeding on number of pigs born (0.0072 pigs/ %F) and born alive (0.00134 pigs / %F), whereas lineage E had the highest significant negative effect of inbreeding

on number born (-0.095 pigs / %F) and born alive (-0.12 pigs / %F). The other lineages had smaller negative effects on both traits; the value for lineage D was significant. These results show that specific animals contributed alleles that were more detrimental to the traits of interest than other founder alleles. The information can be useful if founder alleles can be reintroduced into the current population. It also supports the approach of purging alleles that have negative effects on the population.

Purging and mixing of founder genomes

There have not been designed experiments to study the potential to purge deleterious recessive alleles from inbred domestic populations, but purging is thought to have occurred in the dairy population and other livestock populations where breeding of sires to their daughters has been used to identify deleterious alleles. The concept of identification and removal of deleterious recessive alleles in inbred populations can be visualized easily, but its effectiveness depends on the nature of the undesired alleles and, especially, on the number of these alleles, or genetic load, that the founding animals possess. Inbreeding animals increases the probability of homozygosity, so there is increased potential for recessive undesired alleles to come together and to cause a decrease in fitness or some other trait of interest. To effectively purge deleterious recessive alleles, they need to be 100% distinguishable in homozygous form; alleles with 50% mortality as homozygotes would still potentially be able to pass through a pedigree with even high levels of inbreeding. Also, alleles that have a heterozygous advantage could not be purged even if they are quite undesirable when homozygous (Kalinowski et al., 2000). Templeton and Read (1984) implemented a breeding plan based on the idea of purging deleterious alleles by breeding inbred parents to produce inbred offspring in addition to equalizing founder contributions and increasing recombination in inbred offspring. The inclusion of equalizing and mixing the genome was thought to give inbred offspring of noninbred parents more gene combinations to allow them to adapt to the increase in inbreeding.

A captive population of Speke's gazelle was established with one male and three females, so inbreeding could not be avoided, and a program to reduce the effects of inbreeding on viability had to be created. In order to determine whether the animals showed a decrease in inbreeding depression under this breeding program, the number of lethal equivalents, or genes that would cause death, was calculated before and after the program was started and estimated by the formula:

$$\ln [(x_i + 1) / (n_i + 2)] = -A - Bf_i$$

where n_i is the number of animals with a given inbreeding coefficient f_i , x_i is the number of animals that survived to a given age (30 d), A is indicative of the deaths not due to inbreeding, and B is the number of lethal equivalents per gamete. The left side of the equation is the viability of animals with a certain inbreeding coefficient, with a correction for small sample size. The proportion of the parents' genome derived from different founders was defined by the hybridity coefficient:

$$h = [(1 - D_m) + (1 - D_f)] / 2$$

where D is equal to one in the founders, all other animals have D equal to zero, and D_m and D_f are values for the sire and dam, respectively. The hybridity coefficient measures the amount of opportunity for recombination that has occurred in the offspring. Another measure calculated by

Templeton and Read (1984) is the ancestry coefficient, a_c , determining the extent of equalization of founder genes and defined in a more general form as:

$$a_c = R \left\{ \sum_{i=1}^R [g_i - (1/R)]^2 \right\} / (R - 1)$$

where g_i is the proportion of genes from ancestor i , and R is the number of founders. With complete equalization, a_c is zero and with only one founder contributing all the genes, a_c is one.

The effect of inbreeding on survival was first estimated to determine if inbreeding depression was a problem in the population. Regression analysis revealed there was 21% death rate due to causes other than inbreeding and 2.62 lethal equivalents, showing that inbreeding did decrease survival. Three groups of animals were created, group A, noninbred parents with noninbred offspring ($n = 28$), group B, noninbred parents with inbred offspring ($n = 36$), and group C, including at least one inbred parent with inbred offspring ($n = 59$). Ideally, group C should have been divided into two groups, one with only one inbred parent and the other with both parents being inbred to show more significant results, but the small numbers in the study probably did not allow further separation. The inbreeding range was 0 to 0.3125 among all animals. If Templeton and Read (1984) are correct, then inbreeding depression should be apparent in group B but significantly less in group C. It is important to note that animals are usually divided into inbred and non-inbred groups for further analysis. By creating two distinct groups, the results contain some bias. An animal with an inbreeding coefficient of 0.0625 is more like a non-inbred animal than an animal with an inbreeding coefficient of 0.25, so there should be a continuous scale, which could not be used because of small numbers, rather than discrete groups. Also, the study involved only small sample sizes, and conclusions should be taken with caution.

Templeton and Read (1984) showed that by regressing survival on inbreeding in groups B and C with a pooled intercept, there were significantly fewer lethal equivalents in C (1.59) than in B (2.97). However, the results are not quite as simple as they appear. Kalinowski et al. (2000) pointed out that there was an effect of year of birth on survival of offspring of noninbred parents. They used a less biased approach to measure lethal equivalents per gamete, but the method was still similar to the calculation used by Templeton and Read (1984). Most of the births of groups A and B occurred in the first half of the study, whereas the births of group C occurred mostly in the latter half of the study. Animals in group B born between 1970 to 1975 had 6.56 lethal equivalents whereas those born between 1976 to 1982 had only 1.35 lethal equivalents. The decrease shows that something in addition to selection was acting to decrease the number of lethal equivalents, most likely better management and medical assistance. Group C still showed a decrease in lethal equivalents to 0.57, showing that purging may still be somewhat effective.

Templeton and Read (1984) also tested the effects of equalizing ancestry and of hybridization on inbreeding depression. They hypothesized that a more thoroughly mixed genome, with an increase in gene combinations, will enhance the animals' abilities to respond to changing environments. When the viability was regressed on the hybridity coefficient, the result was not significantly different than zero in group A and B combined or group C. However, inbreeding and hybridity have a correlation of 0.66, so viability is expected to decrease with increasing hybridity if there was no effect other than that associated with inbreeding (Templeton

and Read, 1984). The results obtained again seem to be confounded with year of birth and impact of husbandry (Kalinowski et al., 2000). Templeton and Read (1984) stated that the ancestry coefficient has a significant, positive influence on viability. For a fixed level of low mixed ancestry, inbreeding had a larger effect. In animals with $F > 0$, inbred animals with a highly mixed ancestry ($a_c < 1/3$) had higher viability than inbred animals with poorly mixed ancestry. Kalinowski et al. (2000) pointed out that animals with poorly mixed ancestry had an average inbreeding coefficient of 0.25, whereas animals with highly mixed ancestry had an average inbreeding coefficient of 0.125. As stated previously, Templeton and Read (1984) have divided the animals into inbred and non-inbred individuals, making it hard to determine accurate and unbiased results. Even though the study of the Speke's gazelle leaves questions about the validity of results, Templeton and Read (1984) did what they could with the animals they had and the results they obtained. The study brought forth interesting methods of controlling inbreeding depression that seem to be of conceptual value, but need to be further analyzed for practical situations.

Summary

The mating of relatives has long been associated with inbreeding depression, but the extent to which depression is seen in measured traits depends on the breed of animal, the trait, the genetic load of the founding animals, and the environment. In sheep, inbreeding depression has been shown to have significant effects on growth traits and fertility, with a smaller effect on lamb survival and wool traits. Without maintaining heterozygosity in the population, genetic gains are expected to plateau, and animals will become less adaptable to changes in the environment and less responsive to selection.

The most common description for assessing the expected inbreeding in a population is N_e , but the methods that have been used by conservationists in natural populations are useful in identifying inbreeding when there is still a chance to address the problem. Once real or potential problems of inbreeding have been identified, there are methods to limit inbreeding without a drastic decrease in genetic gains. BLUP selection, although used by many breeders, may not be the best method for limiting inbreeding, but can be altered to include inbreeding or the effects of inbreeding in the calculation of animal merit to make it a better selection tool. The impact of founder contribution, ancestral mixing, and loss of heterozygosity can be assessed to determine the diversity in the population and make breeding decisions resulting in less inbreeding depression based on the information. The current thought on decreasing ΔF in the next generation and in the future is by mating animals with minimum coancestry. Breeders can incorporate this into their breeding scheme by using mass selection and then breeding these animals based on minimum coancestry to limit increases in inbreeding. Breeders may be able to remove some undesired or deleterious alleles in a population through purging, so that inbreeding depression is decreased or removed in populations where inbreeding cannot be avoided. Inbreeding in domestic and natural populations has always been a problem and cannot be ignored, so inbreeding needs to be managed in a way to maintain diversity and/or remove undesired or deleterious alleles in a population to allow animals to survive and remain able to adapt to changing conditions.

Literature Cited

- Analla, M., J. M. Montilla, and J. M. Serradilla. 1998. Analyses of lamb weight and ewe litter size in various lines of Spanish Merino sheep. *Small Ruminant Res.* 29:255-259.
- Analla, M., J. M. Montilla, and J. M. Serradilla. 1999. Study of the variability of the response to inbreeding for meat production in Merino sheep. *J. Anim. Breed. Genet.* 116:481-488.
- Boichard, D., L. Maignel, and E. Verrier. 1997. The value of using probabilities of gene origin to measure genetic variability in a population. *Genet. Sel. Evol.* 29:5-23.
- Boujenane, I., and A. Chami. 1997. Effects of inbreeding on reproduction, weights, and survival of Sardi and Beni Guil sheep. *J. Anim. Breed. Genet.* 114:23-31.
- Caballero, A., and M. A. Toro. 2000. Interrelations between effective population size and other pedigree tools for the management of conserved populations. *Genet. Res. Camb.* 75:331-343.
- Ercanbrack, S. K., and A. D. Knight. 1991. Effects of inbreeding on reproduction and wool production of Rambouillet, Targhee, and Columbia ewes. *J. Anim. Sci.* 69:4734-4744.
- Ercanbrack, S. K., and A. D. Knight. 1981. Weaning trait comparison among inbred lines and selected noninbred and randomly bred control groups of Rambouillet, Targhee, and Columbia sheep. *J. Anim. Sci.* 52:977-988.
- Falconer, D. S., and T. F. C. Mackay. 1996. *Introduction to Quantitative Genetics*. 4th Ed. Longman Group, LTD., Harlow, Essex, UK.
- Galal, E. S. E., E. A. Afifi, I. S. El-Kimary, I. A. Ahmad, and A. F. Shawar. 1981. Lamb survival as affected by inbreeding and cross-breeding. *J. Agric. Sci. Camb.* 96:1-5.
- Hallerman, E. M. 2003. Random genetic drift. In: *Population Genetics: Principles and Applications for Fisheries Scientists*. p 197-214. Am. Fisheries Soc. Bethesda, MD.
- Kalinowski, S. T., P. W. Hedrick, and P. S. Miller. 2000. Inbreeding depression in the Speke's gazelle captive breeding program. *Conserv. Biol.* 14:1375-1384.
- Klieve, H. M., B. P. Kinghorn, and S. A. Barwick. 1994. The joint regulation of genetic gain and inbreeding under mate selection. *J. Anim. Breed. Genet.* 111:81-88.
- Lacy, R. C. 1989. Analysis of founder representation in pedigrees: founder equivalents and founder genome equivalents. *Zoo Biol.* 8:111-123.
- Lacy, R. C. 1995. Clarification of genetic terms and their use in the management of captive populations. *Zoo Biol.* 14:565-578.
- Lamberson, W. R., and D. L. Thomas. 1984. Effects of inbreeding in sheep: a review. *Anim. Breed. Abstr.* 52:287-297.
- Lamberson, W. R., D. L. Thomas, and K. E. Rowe. 1982. The effects of inbreeding in a flock of Hampshire sheep. *J. Anim. Sci.* 55:780-786.
- MacCluer, J. W., J. L. Van de Berg, B. Read, and O. A. Ryder. 1986. Pedigree analysis by computer simulation. *Zoo Biol.* 5:147-160.
- Mandal, A., K., P. Pant, P. K. Rout, S. A. Singh, and R. Roy. 2002. Influence of inbreeding on growth traits of Muzaffarnagari sheep. *Indian J. Anim. Sci.* 72:988-990.
- McDaniel, B. T. 2001. Uncontrolled inbreeding. *J. Dairy Sci.* 84:E185-E186.
- O'Brien S. J., M. E. Roelke, L. Marker, A. Newman, C. A. Winkler, D. Meltzer, L. Colly, J. F. Evermann, M. Bush, and D. E. Wildt. 1995. Genetic basis for species vulnerability in the cheetah. *Science.* 227:1428-1434.
- Quinton, M., C. Smith, and M. E. Goddard. 1992. Comparison of selection methods at the same level of inbreeding. *J. Anim. Sci.* 70:1060-1067.

- Ralls, K., P. H. Harvey, and A. M. Lyles. 1986. Inbreeding in natural populations of birds and mammals. In: M.E. Soule ad. Conservation Biol.: The Science of Scarcity and Diversity. p 35-66. Sinauer Associates, Sunderland, MA.
- Rodrigañez, J., M. A. Toro, M. C. Rodriguez, and L. Silio. 1998. Effect of founder allele survival and inbreeding depression on litter size in a closed line of Large White pigs. Anim. Sci. 67:573-582.
- Roughsedge, T., S. Brotherstone, and P. M. Visscher. 1999. Quantifying genetic contributions to a dairy cattle population using pedigree analysis. Livestock Prod. Sci. 60:359-369.
- Templeton, A. R, and B. Read. 1984. Factors eliminating inbreeding depression in a captive herd of Speke's gazelle (*Gazella spekei*). Zoo Biol. 3:177-199.
- Weigel, K. A. 2001. Controlling inbreeding in modern breeding programs. J. Dairy Sci. 84: E177-E184.
- Weiner, G., G. J. Lee, and J. A. Woolliams. 1992. Effects of rapid inbreeding and of crossing of inbred lines on the body weight growth of sheep. Anim. Prod. 55:89-99.

Chapter II.

The effect of population structure on the rate and degree of diversity loss in a crossbred sheep flock

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ABSTRACT: This study analyzed changes in genetic diversity in small, closed selection and control lines of sheep established in 1983 from 50% Dorset, 25% Rambouillet, and 25% Finnsheep breeding. Founder animals of the three breeds were mated to create three-way crosses. In 1987, descendents of the 161 founders were divided into a fall-lambing selection line (S) of 125 ewes and 10 rams, a fall-lambing environmental control line (E) of 55 ewes and 5 rams, and a spring-lambing genetic control line (G) of 45 ewes and 5 rams. In each line, measures of genetic diversity were calculated for all rams and ewes available at the time of line formation (REL) and at the end of the study for three sets of animals: all lambs born including dead lambs (L), a single offspring of all matings including potential offspring of ewes that did not lamb (M), and all rams and ewes available (RE). The average generation interval was 2.65 yr in S and 4.28 yr in G. Values used to quantify diversity were the change in inbreeding per generation (ΔF), effective number of breeding animals (N_e), effective number of founders (f_c), effective number of ancestors (f_a), founder genome equivalents (f_g), and two additional measures of heterozygosity (GD_1 , GD_2), calculated using the additive relationship matrix and(or) 100 gene drop simulations. The averages of N_e for S and G lines were 33.78 and 29.07; values of N_e were most similar between S and G for RE. The values for f_c , f_a , and f_g , calculated from the additive relationship matrix for REL were 40.25, 38.42, and 24.03, respectively for S and 39.59, 33.95, and 20.50, respectively for G. At the end of the study values for f_c , f_a , and f_g for RE were 31.46, 20.85, and 7.65, respectively for S and 30.80, 19.57, and 10.04, respectively for G. The differences between the actual number of founders and f_c shows the loss of diversity due to unequal founder representation, differences between f_c and f_a are due to bottlenecks, and differences between f_a and f_g are due to additional drift. Larger decreases in diversity were seen in M and L in both S and G, but particularly in G, where all available breeding rams were not used in all years. For most calculations, values obtained from gene drop and the relationship matrix were very close. The exception was f_g in M of G, apparently due to non-random sampling of sires in previous generations that resulted in increased diversity. Gene drop analysis allowed allelic diversity and survival to be estimated. Of the 322 unique alleles assumed present in the founders, 71% in S and 58% in G survived at line formation; all appeared in over 50% of the runs. Of the alleles possible in RE of S and G, only 65% of 254 alleles and 70% of 192 alleles, respectively, survived in any single run, and only 6% and 8% of the possible alleles were present in more than 50 of 100 runs. The number of alleles and heterozygosity lost demonstrates the impact of a closed, small population on diversity. The values obtained also show that RE of S and G were relatively similar in diversity, as desired to determine genetic change in a selection study relative to an appropriate baseline.

Key Words: Allelic diversity, Bottlenecks, Heterozygosity, Inbreeding, Selection, Sheep

Introduction

Small, closed, selected populations can rapidly lose heterozygosity and allelic diversity. Most breeding programs and studies involving domestic animals acknowledge that inbreeding occurs, and may try to minimize inbreeding or to quantify the increase by calculating the change in inbreeding per generation (ΔF) (Boichard et al., 1997). However, calculation of ΔF may not reflect the cumulative effects of inbreeding and random genetic drift on allelic diversity or specify when or how such diversity was lost. Methods more commonly used by conservation geneticists may provide more informative measures of changes in genetic diversity. Procedures derived by Lacy (1989, 1995) and Boichard et al. (1997) allow analysis of founder effects, bottlenecks, and genetic drift. The calculation derived by Boichard et al. (1997) also can be used to identify the most influential ancestors in a pedigree. If populations are evaluated at a single point or in each generation by these methods, the causes of losses in diversity can be identified and potentially corrected, and important ancestors may be evaluated for the trait(s) under selection.

It is also important in a selection experiment to be able to compare genetic change relative to an unselected control line otherwise similar in population structure and original composition. After a breeding structure has been created and implemented, retrospective analysis of rates of inbreeding and of remaining diversity in the selection and control lines can assess the adequacy of the breeding design. The experiment described in this paper was designed to quantify diversity present in a population of sheep and identify which subset of animals provides a better estimate of that diversity. We also compared the genetic control line to the selection line to discuss the impacts of selection and restricted population size, and to determine if the two lines were adequate for comparison in the selection study.

Materials and Methods

An experimental sheep flock was established in 1983 from 50% Dorset, 25% Rambouillet, and 25% Finnsheep breeding. By 1987, all lambs were progeny of three-way crosses, after which the population remained closed. After the 1987 lambing, 45 of 225 ewes were randomly chosen to form a genetic control line (G) that remained on a spring-lambing system. The remaining ewes were randomly divided into a selection line (S) with 125 ewes and an environmental control line (E) with 55 ewes. Both the S and E ewes then were selectively bred to lamb in the fall. Ten rams were used in each year in S, and each ram was exposed to about 13 ewes. The E and G lines used about five rams each, which were bred to approximately 11 and 9 ewes each, respectively (Al-Shorepy and Notter, 1997).

S line. Selection in the S line for fertility in spring (May and June) began in 1988 (Al-Shorepy and Notter, 1997). Up to one-third of the ewes in S were replaced annually as older ewes were culled for declining fertility and unsoundness. All healthy ewe lambs were exposed in the spring at 7 mo of age to selected rams. Pregnant ewe lambs were retained, and additional ewe lambs were selected, as needed, based on their estimated breeding value for fertility. Ewe lambs that had been selected but did not lamb in the first year were given a second chance to lamb before being culled. Four to seven rams were replaced each year. No more than two rams per sire were chosen in any given year. Most rams were used for only 1 or 2 yr; only two of 37

rams were used in a third year (Al-Shorepy and Notter, 1997). The entire S line, from 1987 until the end of the study in 1998, consisted of 1190 lambs (including dead lambs), 297 dams, and 67 sires.

E line. The E line was maintained with approximately the same ewe age distribution as S. Replacement rams and ewes were randomly chosen from the G line, to avoid inadvertent selection for fall-lambing due to a fall-birth. Because of the movement of G animals to E, the breeding stock in E was a sub-sample of the G line. Again, approximately one-third of ewes and one-half of rams were replaced each year. No more than one ram per sire/yr was randomly chosen for use in E. Ewes were culled at random within year of birth. Ewe lambs were randomly chosen, but only one ewe lamb/ewe could be chosen each year. For the purpose of the population analysis, the E line was not used because E rams and ewes did not produce replacement breeding stock. The entire E line consisted of 492 lambs (including dead lambs), 129 dams, and 54 sires.

G Line. Animals in the G line were used as a source of replacements for E in the fall lambing study. In order to minimize genetic drift, ewes in G were only culled for unsoundness or if they failed to produce in two consecutive years. Ewe lambs were randomly chosen with the restriction of one ewe lamb per ewe/yr. An average of 7.5 replacement ewe lambs were used in G each year. Almost all of the G line ewe lambs were retained for use in either E or G. The G line included 922 lambs (including dead lambs), 115 ewes, and 32 sires.

Sire families were identified for G in 1987. Whenever possible, two sons were maintained from each sire family, even if they were not used. When a sire died, a son of the sire was kept, when possible. The maintenance of sire families was used to limit inbreeding and drift in the small G line. Of the 14 sire families present in 1987, 12 were still available in 1993 (Al-Shorepy and Notter, 1997), and 11 were available in 1998. Usually, five sires representing different sire families were used each year. Three new sires were used the following year in order to eventually sample all sire families and maintain genetic connections between years. In the last year of the study, only three rams were used. Rams were culled only for physical unsoundness.

Pedigree Analysis

Founders were defined as the 161 animals that created the flock, starting in 1983, whose parents were assumed to be unknown. Crossbred animals used in 1983 that were from Virginia Tech flocks had their parents included in the pedigree; all other animals were assumed to have unknown parents. A few animals with one or two unknown parents that were not founders were removed from the pedigree. The entire pedigree consisted of 5041 animals, but animals produced in other studies peripheral to the main selection were removed from the study, resulting in a pedigree that contained 4257 animals.

Generation codes (g) were calculated for all animals as, $g = \frac{1}{2} (g_s + g_d) + 1$, where g_s is the generation code of the sire, g_d is the generation code of the dam, and line founder animals were given a generation code of zero. Generation intervals were determined by regressing generation number on year of birth. Inbreeding coefficients and additive relationships were

calculated using the pedigree to form the additive relationship matrix. Changes in ΔF in each line were obtained by regressing individual inbreeding coefficients on generation number. The effective number of breeding animals (N_e) calculated as:

$$N_e = 1 / (2\Delta F)$$

Indicates the number of breeding animals that would have produced the observed rate of inbreeding if bred under ideal conditions in one generation.

Genetic diversity was analyzed using the additive relationship matrix, gene drop methods (MacCluer et al., 1986), and an iterative procedure to estimate the effective number of ancestors (Boichard et al., 1997). Gene drop is a computer simulation procedure that is performed by giving each founder a pair of unique alleles and randomly transmitting the alleles through the pedigree (MacCluer et al., 1986). In order to obtain the probability that founder alleles would survive and estimate standard deviations on measures of genetic diversity, the gene drop procedure was repeated 100 times.

Additional measures of genetic diversity. The effective number of founders (f_e) indicates the number of equally contributing founders that would have been expected to result in the same level of genetic diversity as that observed in the current population (Lacy, 1989). The f_e is calculated as:

$$f_e = 1 / \sum_{i=1}^n p_i^2$$

where p_i is the expected proportional genetic contribution of founder i , obtained from the average relationship of the founder to each animal in the current population, and n is the total number of founders (Lacy, 1989). A complimentary measure, genetic diversity (GD_1), approximates the amount of heterozygosity still available in the population after reductions due to unequal founder contributions and can be calculated from the additive relationship matrix as:

$$GD_1 = 1 - 1/2f_e$$

or from gene drop simulations as:

$$GD_1 = 1 - \sum_{k=1}^{2n} \bar{q}_k^2$$

where \bar{q}_k is the frequency of founder gene k averaged over all individuals in the current generation and all replicates in a gene drop simulation, and n is the number of founder animals (Rodrigañez et al., 1998).

A method created by Boichard et al. (1997) was used to estimate the effective number of ancestors (f_a) and to identify animals that have the highest relationship to the current population:

$$f_a = 1 / \sum_{i=1}^m a_i^2$$

where a_i is the marginal contribution of each ancestor to the current generation, and m is the total number of contributing ancestors. The calculation can be used to identify the most influential ancestors, which may or may not be founders.

The number of founder genome equivalents (f_g) was used to estimate the joint effects of unequal founder contributions, bottlenecks, and genetic drift:

$$f_g = 1 / \sum_{i=1}^c (p_i^2 / r_i) \approx t / (2 \sum_{j=1}^t (\sum_{k=1}^{2n} \bar{q}_{kj}^2))$$

where p_i is the expected proportional genetic contribution of founder i , r_i takes values of 0.5 or 1 and is the expected proportion of founder i 's alleles that remain in the current population, and c is the total number of contributing founders (Lacy, 1989), \bar{q}_{kj} is the frequency of founder gene k for replicate j , t is the number of gene drop replicates, and n is the total number of founders (Rodríguez et al., 1998). The effective number of genomes also can be calculated using the additive relationship matrix as:

$$f_g = 1 / 2\bar{r}$$

where \bar{r} is half the average relationship (mean coancestry) between all individuals in the current population, including the relationship of each individual to itself (Lacy, 1995). To equate the increase in homozygosity to a loss in heterozygosity due to unequal founder contribution and genetic drift, the following measure of genetic diversity (GD_2) was calculated from gene drop:

$$GD_2 = 1 - \sum_{j=1}^t [\sum_{k=1}^{2n} \bar{q}_{kj}^2] / t$$

where \bar{q}_{kj} is the frequency of founder gene k for replicate j , t is the number of replicates, and n is the number of founders (Rodríguez et al., 1998). The second term of the equation is simply the mean coancestry of the current population, giving an alternative calculation for GD_2 .

Determination of informative population statistics is complicated by overlapping generations, selection, and the maintenance of sire families that are not sampled in all years. In order to analyze the diversity available in S and G, one set of animals at line formation and three sets of animals from the end of the study were considered in each line: all rams and ewes available at the time of line formation, all lambs born (including dead lambs), one offspring of all matings (including potential offspring of ewes that did not lamb), and all rams and ewes available at the end of the study. The final population of available breeding animals is particularly important in G, where rams were retained but not used in all years and also in S where all ewes may not reproduce in a given year. It is also important to note that the measures in this study were calculated with a complete pedigree, and that missing information may result in a substantial apparent increase in f_e , f_a , f_g , GD_1 , and GD_2 .

Results and Discussion

Measures of inbreeding and heterozygosity

The rate of inbreeding in the current rams and ewes of S and G were comparable (Table 2.1). Even though G had fewer breeding animals in each year, the longer generation interval, decreased culling, and maintenance of sire lines limited rates of inbreeding relative to those observed in the larger S line. Differences in ΔF and N_e among the last three groups in line S are small. One problem with the N_e metric is that the value indicates the number of breeding

animals needed to produce the average ΔF and does not quantify the cumulative decrease in allelic diversity or changes in breeding structure from year to year. Hence, the value obtained for N_e is not comparable to measures of f_e , f_a , and f_g . The change in inbreeding per generation in the current available breeding animals of G is similar to ΔF for the S line due to the increased generation interval and retention of sire lines. Lambs and matings had a slightly higher ΔF in G than in S. The differences observed in both lines between lambs and matings are not large, and are probably not biologically significant.

It is apparent from Figures 2.1 and 2.2 that more matings with inbreeding coefficients greater than or equal to 0.25 resulted in lambs in G than in S. This result may reflect the higher mean fertility in fall-matings compared to spring-matings (88.2% vs. 47.5%). Lambs with inbreeding coefficients greater than or equal to 0.25 generally did not become parents in either line (Figure 2.3). The lack of rams and ewes with inbreeding coefficients greater than or equal to 0.25 in S could be attributed to decreased survival and selection against decreased fertility in spring. In G, the lack of animals with higher inbreeding coefficients could only be due to decreased survival or to animals that were too sickly or small to be bred.

The difference between the actual number of founders (161) and f_e shows that unequal founder representation has a large impact on genetic diversity (Tables 2.2, 2.3, and 2.4), but most of the loss in diversity had occurred by 1987, when the lines were formed (Table 2.2). There was a smaller loss in diversity from line formation to the end of the study, showing that there was either a thorough mixing of founders, which was the goal, and(or) that all line founder genomes were adequately maintained. Similar values for f_e were found in the current lambs, matings, and available breeding animals for S (Table 2.3), showing that f_e has become relatively constant. In a closed ideal population, f_e is expected to plateau when founders are no longer present, because founder representation will eventually be relatively stable among all animals (Caballero and Toro, 2000). The f_e is still decreasing in G from available breeding animals to lambs because of the longer average generation interval of 4.6 yr/generation in G, as compared to 2.6 yr/generation in S (Table 2.4). Although f_e in the current animals is less than the actual number of founders by at least 129, the amount of heterozygosity, calculated by GD_1 , was still relatively high at the start of the lines and in the final animals (Table 2.2, 2.3, and 2.4). There was only a negligible loss in heterozygosity due to founder representation from the beginning of the lines until 1998. It is important to note that the measures of GD are relative to the founding population, which is only a sample of the diversity present in the breed or species. The values obtained for GD_1 , calculated using the relationship of founders to the current population and gene drop, are comparable showing that founder representation is adequately captured using the relationship matrix.

When compared to f_e , f_a shows the impact of bottlenecks on the population (Table 2.2, 2.3, and 2.4). It is apparent from the small decrease in rams and ewes present at the formation of the respective line that there were very few bottleneck events that had occurred up to that point, but the G line rams and ewes showed a larger decrease than S due to fewer animals being used at line formation causing a greater potential for bottleneck events (Table 2.2). Since there was no selection occurring in the matings made to create three-way crosses, it was expected that limited drift due to bottlenecks would occur by the time of line formation.

The difference in f_a between the final lambs and matings of S indicates that matings that had resulted in lambs had a more equal distribution of ancestors. It may be that lambs with low contributing ancestors had a better chance of surviving, because they were less likely to be inbred than lambs with prominent ancestors. The slightly larger f_a noted in the final available breeding animals of S is due to older animals being present with more diverse relationships to older ancestors. Bottlenecks had some effect in S, but had an even greater effect in the final lambs and matings of G due to the small population size and only one discrete year being represented. The value for f_a obtained from the final available breeding animals of G, when compared to lambs and matings of G, shows the beneficial impact of overlapping generations on maintaining diversity by limiting bottlenecks and increasing the possible number of offspring per parent.

Marginal contributions of all prominent ancestors were obtained by calculation of f_a , identifying the most influential ancestors in each population (Figure 2.4 and 2.5). The three most influential ancestors, making over 50% of the genetic contribution, in the final lambs and matings of G were the three sires used in the last breeding (data not shown). If G had discrete generations, then these would be bottleneck sires, but the retention and use of additional sires spread the diversity of the population over multiple years. Sixteen animals made up about 80% of the contribution to the current available breeding animals of G (Figure 2.5). Of these animals, only one was born after formation of the G line, six were older sires, two were older dams, six were foundation sires, and one was a foundation dam. Since the most important animals occur before or shortly after formation of G, the currently available breeding animals in G have not experienced narrow bottlenecks, which is also apparent from the change in f_e to f_a in currently available breeding animals as compared to lambs or matings of G (Table 2.4).

The most important ancestor in all three sets of animals in S was a sire born in 1990 (Figure 2.4). This sire may have contributed genes with a particularly significant positive effect on out-of-season lambing. Seven, six, and nine animals in S made up about 50% of the contributions in final lambs, matings, and available breeding animals, respectively. Only one of the seven most important ancestors of the current lamb crop was born before the formation of S. Of the remaining six animals, three were sires of the last lamb crop, one was an older bottleneck sire, and the remaining two were bottleneck dams. Since the only lamb crop analyzed was the current one and there were overlapping generations, there is the potential for the animals that contributed most to the current lamb crop change each year. There was only one animal in common between the significant ancestors of lambs and of final available breeding animals of S (i.e., in ancestors making up the top 50% of contributions). The 20 influential animals making up the top 80% of the contribution to the currently available breeding animals in S consisted of five sires born in S, four dams and four sires born before line formation and one dam and six sires that were foundation animals. Since most of the high contributors to final available breeding animals of S were older animals, the reason for the differences between lambs and available breeding animals of S is that the parents would be bottleneck animals with discrete generations, but this bottleneck was circumvented by retention of breeding animals from multiple generations. Hence, the influential animals born and used in S may have had a considerable impact on the trait being selected.

When all losses of diversity were accounted for in f_g , the impact of a closed population becomes apparent. The numbers obtained from calculation of f_g show how many animals would

be needed to produce the same genetic diversity if all founders contributed equally and no founder alleles were lost through drift under random mating (Lacy, 1989). The values for f_g were estimated through gene drop and the additive relationship matrix (Table 2.2, 2.3, and 2.4). It is apparent from the rams and ewes present at the formation of the lines (Table 2.2) that there was not much diversity loss due to drift and that most of the loss in diversity up to this point was due to unequal founder representation. The G line founder rams and ewes had a slightly smaller number of founder genomes than the S line due to the smaller size of G. The founder genome equivalents estimated through gene drop and the relationship matrix are equivalent as are the measures for GD_2 (Table 2.2).

The final populations of lambs and final available breeding animals have similar values for f_g in both lines using gene drop or the relationship matrix, but in matings of S and particularly of G, the f_g was higher when calculated using gene drop simulation analysis (Table 2.3 and 2.4). It is the difference between values for f_g in lambs and matings of G that is of concern. The average difference in f_g between lambs and matings in G for all simulations of gene drop was 0.28 with a standard deviation of 0.23. A difference of 0.75 genomes or more between f_g in lambs and matings in G had a probability of occurrence of 0.03 in the 100 gene drop simulations. The highest difference observed between f_g in lambs versus matings in the gene drop simulations was 1.13 genomes; the second highest difference was 0.80. The discrepancy between lambs and matings in the gene drop and relationship matrix calculations is not out of the range of probability, but the difference in the relationship matrix calculation seems high and may indicate that our pedigree resulted in a higher amount of diversity than other pedigrees with the same relationships.

The f_g calculated using the relationship matrix in S and G for lambs is larger than that for matings, because more than one animal was produced in some matings, resulting in less genetic drift. The almost two-fold difference of lambs and(or) matings to final available breeding animals of G is due to the maintenance of sire lines and reduced culling. When all four values of f_g in lambs and matings of S and G from gene drop analysis are compared, they are all similar, which is the goal for a genetic control line.

The amount of heterozygosity calculated by GD_2 , as a function of f_g , quantifies the decrease from the founding population (Table 2.3 and 2.4). In S and G, lambs are predicted to retain 92.19% and 90.34% of the original heterozygosity in the founders, respectively. The difference in GD_2 between final available breeding animals and lambs of S shows that there has been an estimated reduction of 1.3% heterozygosity from parents to offspring. The difference in the loss of heterozygosity between lambs and available breeding animals of G is larger (4.68%) than S, due to a smaller population.

Allelic diversity

One of the unique benefits of gene drop analysis is to be able to estimate the quantity and change in allelic diversity. Heterozygosity is an indication of the presence of more than one allele at a locus, and populations can still have high levels of heterozygosity if only two alleles are present. The more alleles that are present, the higher the allelic diversity, and thus potential selection response, and(or) adaptability. The gene drop procedure simulates the most limiting

case of allelic loss at neutral loci, because it assumes that each founder had two unique alleles. Obviously, the quantity of allelic diversity that appears to have been lost will be much larger than in almost all populations, but it gives a point of reference for determining the amount of loss.

The information in Table 2.5 shows the simulated loss of allelic diversity if all founders had had two unique alleles. There were large proportions of alleles that had the chance to pass through the pedigree at line formation. However, some alleles were lost due to the formation of the lines dropping the number of possible founder alleles from 322 to 254 in S and 192 in G. For rams and ewes present at the end of the study in S and G, more than half of the possible alleles were present in at least one of the 100 simulations of gene drop. Within each simulation there was an average of 84 and 65 founder alleles present in S and G rams and ewes that founded the respective lines (Table 2.6). The average numbers of alleles per run dwindled to an average of 35 and 33 alleles in the final rams and ewes in S and G, respectively, which were only 14% of the possible 254 founder alleles in S and 17% of the 192 possible founder alleles in G. Based on the 161 total founders, an average of 10% of their alleles passed through to the end of the pedigree in any one simulation in the S line or the G line (Table 2.6). In S and G about half and a little less than half of the founder alleles, respectively, passed through to the end of the pedigree in at least one simulation of gene drop, most alleles did not have a high frequency of appearance across simulations (Table 2.5).

The distribution of the occurrence of each individual allele (Figure 2.6 and 2.7) shows that of the 166 and 135 alleles in the final rams and ewes of S and G, respectively, only 8% of the alleles in S and 12% of the alleles in G had a 50% or greater probability of survival. Alleles present in the founders had an 89% and a 90% chance of loss through the pedigree in final rams and ewes of S and G, respectively. The large amount of loss in allelic variation is expected in small, closed, selected populations. For the purpose of the out-of-season breeding study, the loss should have been as similar as possible between lines, even though the breeding structure differs.

Implications

The breeding structure of the G line was shown to be adequate to sustain genetic diversity approximately comparable to that maintained in the S line, even though it had a smaller census population. Losses of diversity were determined through calculation of f_e , f_a , and f_g . Had animals been available from prior generations, a higher amount of the diversity could have been recovered. The rate of loss of allelic diversity apparent through gene drop analysis supports the loss in diversity seen in calculations of f_e , f_a , and f_g . Influential ancestors, selected by the estimation of f_a , specify animals that have a high contribution to the current population. These animals can then be reexamined in order to determine if they were better than average in their contemporary group, indicating they possess beneficial alleles for the selected trait. Gene drop and the iterative procedure developed by Boichard et al. (1997) may be used in S to indicate if frequencies of particular alleles increased because they were associated with or transmitted by an animal that had high genetic merit for out-of-season breeding.

If maintenance of current diversity is the only goal, there is more diversity available in the rams and ewes present in G as compared to S, and an even larger amount could be retained if

the two lines were combined. The analysis of only lambs does not adequately describe the variation present in the lines, but is a close approximation for lines with short generation intervals and without retention of surplus breeding animals (e.g., of rams in G). If adequate information is available, it is better to analyze the available breeding animals to determine the amount of diversity in the population. With the creation of programs such as GENES (Lacy, 1994), any pedigree could be easily entered into the program and a diversity analysis performed.

Literature Cited

- Al-Shorepy, S. A., and D. R. Notter. 1997. Response to selection for fertility in a fall-lambing sheep flock. *J. Anim. Sci.* 75:2033-2040.
- Boichard, D., L. Maignel, and E. Verrier. 1997. The value of using probabilities of gene origin to measure genetic variability in a population. *Genet. Sel. Evol.* 29:5-23.
- Caballero, A., and M. A. Toro. 2000. Interrelations between effective population size and other pedigree tools for the management of conserved populations. *Genet. Res. Camb.* 75:331-343.
- Lacy, R. 1989. Analysis of founder representation in pedigrees: Founder equivalents and founder genome equivalents. *Zoo Biol.* 8:111-123.
- Lacy, R. C. 1994. GENES: A computer program for pedigree analysis and population management. Chicago: Chicago Zoological Soc.
- Lacy, R. C. 1995. Clarification of genetic terms and their use in the management of captive populations. *Zoo Biol.* 14:565-578.
- MacCluer, J. W., J. L. Van de Berg, B. Read, and O. A. Ryder. 1986. Pedigree analysis by computer simulation. *Zoo Biol.* 5:147-160.
- Rodríguez, J., M. A. Toro, M. C. Rodríguez, and L. Silio. 1998. Effect of founder allele survival and inbreeding depression on litter size in a closed line of Large White pigs. *Anim. Sci.* 67:573-582.

Table 2.1. Generation interval, change in inbreeding per generation (ΔF), and effective number of breeding animals (N_e) for lambs, matings, and final rams and ewes in selection (S) and genetic control (G) lines

	S	G
Generation interval:		
Lambs	2.68	4.51
Matings	2.56	4.32
Rams and Ewes	2.71	4.00
ΔF :		
Lambs	0.0141 ± 0.0004	0.0191 ± 0.0010
Matings	0.0156 ± 0.0004	0.0182 ± 0.0013
Rams and Ewes	0.0146 ± 0.0006	0.0142 ± 0.0015
N_e :		
Lambs	35.39	26.18
Matings	32.07	27.49
Rams and Ewes	34.29	35.21

Table 2.2. Effective number of founders (f_e), effective number of ancestors (f_a), effective number of genomes (f_g), and gene diversity measures one and two (GD_1 , GD_2) in the selection (S) line and the genetic control (G) line at the time of line formation calculated from the relationship matrix (A) and gene drop simulation (D)

	Rams and ewes	
	S	G
f_e	40.25	39.59
GD_1 - A	0.988	0.987
GD_1 - D	0.9971 ± 0.0006	0.9860 ± 0.0003
f_a	38.42	33.95
f_g - A	24.03	20.50
f_g - D	24.08 ± 2.38	19.88 ± 2.46
GD_2 - A	0.979	0.976
GD_2 - D	0.979 ± 0.024	0.975 ± 0.021

Table 2.3. Effective number of founders (f_e), effective number of ancestors (f_a), effective number of genomes (f_g), and gene diversity measures one and two (GD_1 , GD_2) in the final animals of the selection (S) line calculated from the relationship matrix (A) and gene drop simulation (D)

	S		
	Lambs	Matings	Rams and ewes
f_e	31.42	31.30	31.46
GD_1 - A	0.984	0.984	0.984
GD_1 - D	0.9839 ± 0.0004	0.9838 ± 0.0004	0.9844 ± 0.0003
f_a	19.39	16.82	20.85
f_g - A	6.40	5.76	7.65
f_g - D	6.30 ± 1.27	5.98 ± 1.23	7.66 ± 1.39
GD_2 - A	0.922	0.913	0.935
GD_2 - D	0.921 ± 0.013	0.917 ± 0.012	0.935 ± 0.014

Table 2.4. Effective number of founders (f_e), effective number of ancestors (f_a), effective number of genomes (f_g), and gene diversity measures one and two (GD_1 , GD_2) in the final animals of the genetic control (G) line calculated from the relationship matrix (A) and gene drop simulation (D)

	G		
	Lambs	Matings	Rams and ewes
f_e	25.17	26.20	30.80
GD_1 - A	0.980	0.981	0.984
GD_1 - D	0.9785 ± 0.0004	0.9794 ± 0.0004	0.9845 ± 0.0003
f_a	9.34	9.14	19.57
f_g - A	5.18	4.43	10.04
f_g - D	5.07 ± 0.93	5.12 ± 0.92	9.85 ± 1.61
GD_2 - A	0.903	0.887	0.950
GD_2 - D	0.901 ± 0.009	0.902 ± 0.009	0.949 ± 0.016

Table 2.5. Proportions of possible founder alleles that appeared in any single simulation of gene drop or in ≥ 50 simulations at line formation and at the end of the study in the selection (S) and genetic control (G) lines

	Alleles in any run ^a		Alleles in ≥ 50 runs	
	S	G	S	G
Line founder rams and ewes ^b	0.71	0.58	0.71	0.58
Final lambs ^c	0.65	0.69	0.05	0.06
Final matings ^c	0.65	0.69	0.05	0.06
Final rams and ewes ^c	0.65	0.70	0.06	0.08

^a 322 founder alleles possible at line formation, 254 possible in S, 192 possible in G.

^bRams and ewes present at the formation of the respective line.

^cLambs, matings, and rams and ewes present at the end of the study.

Table 2.6. Average number of alleles present per simulation of gene drop for selection (S) and genetic control (G) lines and the average percent of alleles appearing out of all possible alleles

	Avg number of alleles per run		% of possible alleles ^a	
	S	G	S	G
Final lambs ^b	31 ± 3	26 ± 3	12	14
Final matings ^b	30 ± 3	23 ± 2	12	12
Final rams and ewes ^b	35 ± 3	33 ± 3	14	17
Line founder rams and ewes ^c	84 ± 4	65 ± 4	26	20

^a 322 possible alleles at line formation, 254 possible in S, 192 possible in G.

^b Lambs, matings, and rams and ewes present at the end of the study.

^c Rams and ewes present at the formation of the respective line.

Figure 2.1. Matings change in inbreeding per generation in selection and genetic control lines

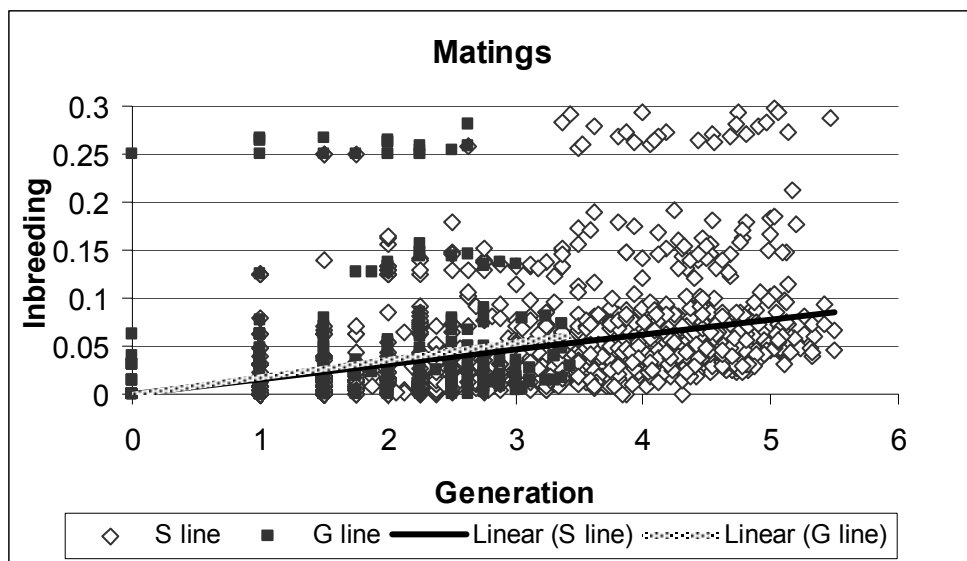


Figure 2.2. Lambs change in inbreeding per generation in selection and genetic control lines

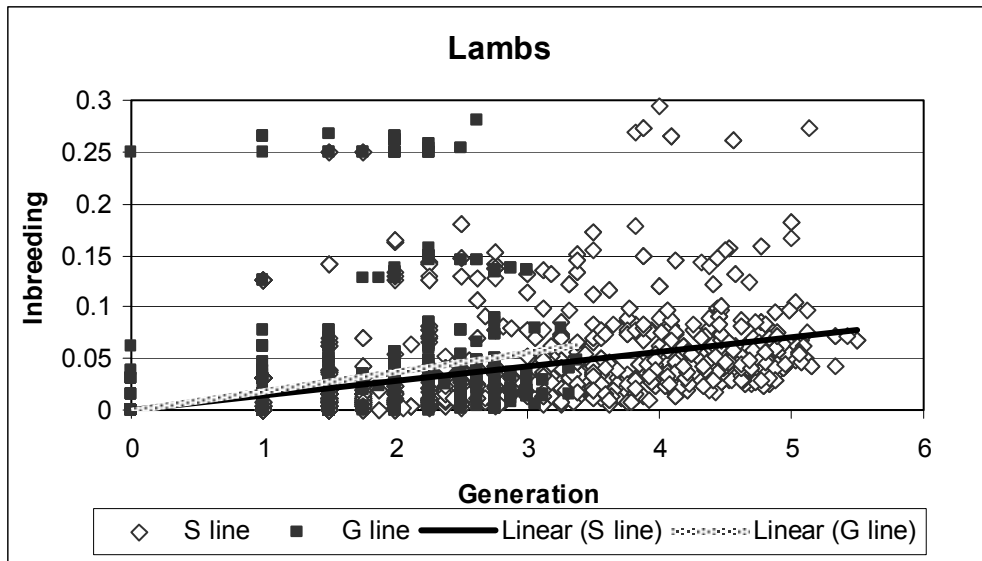


Figure 2.3. Rams and ewes present at the end of the study change in inbreeding per generation in selection and genetic control lines

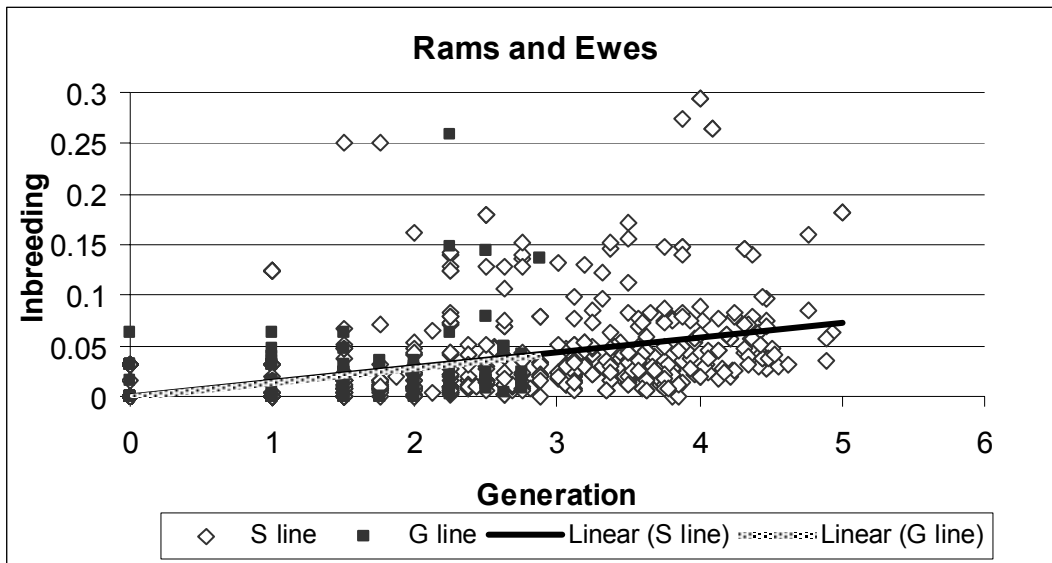


Figure 2.4. The marginal and total contribution of the ancestors of rams and ewes present at the end of the study in the selection line

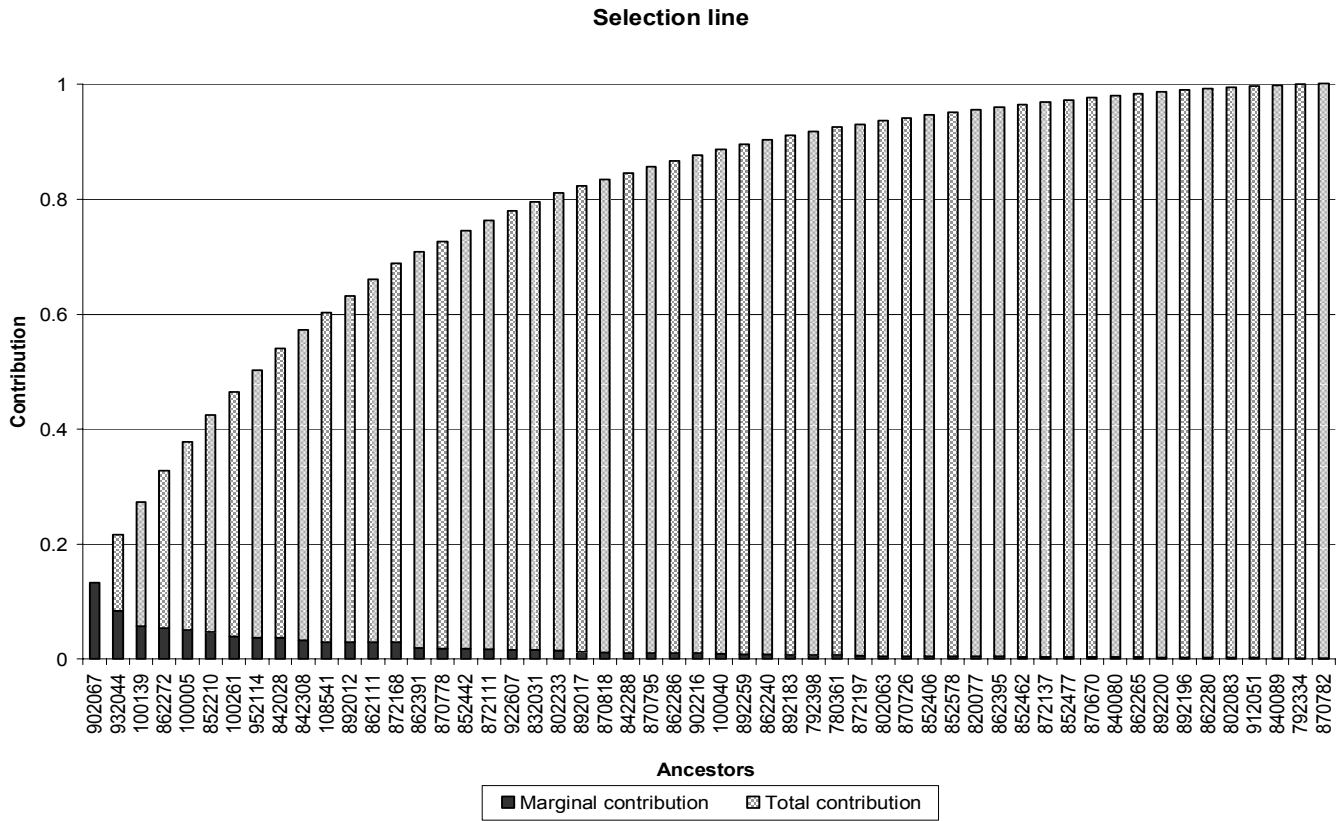


Figure 2.5. The marginal and total contribution of the ancestors of rams and ewes present at the end of the study in the genetic control line

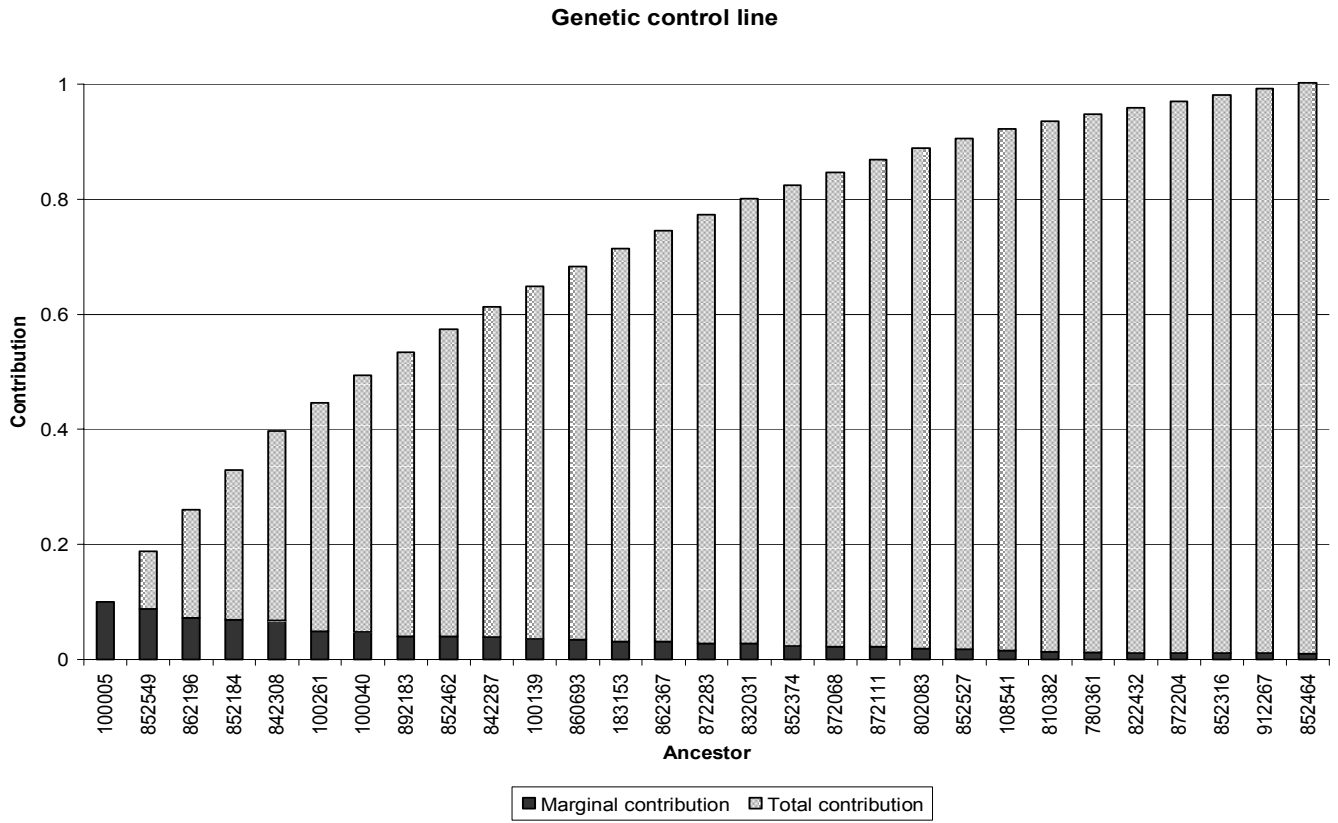
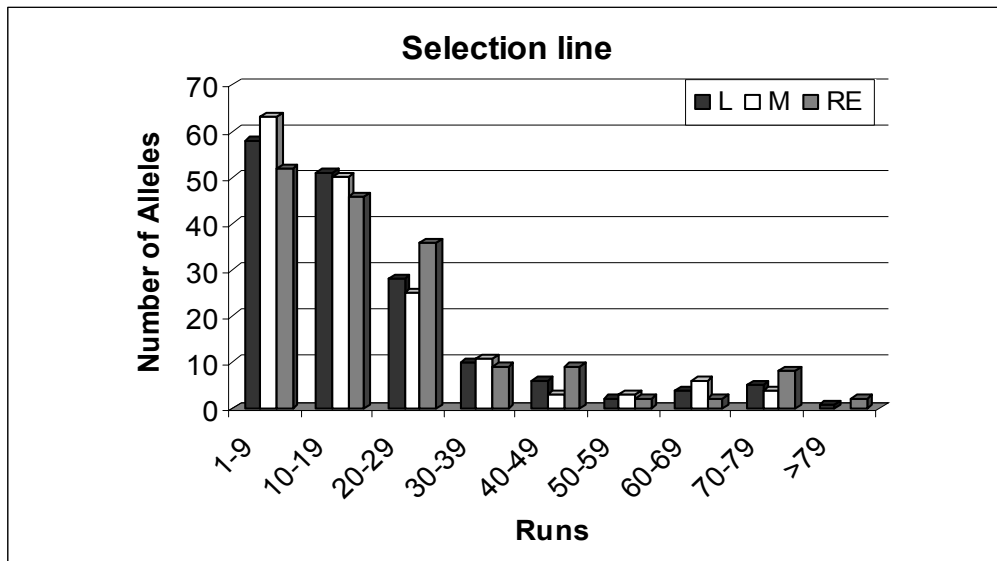
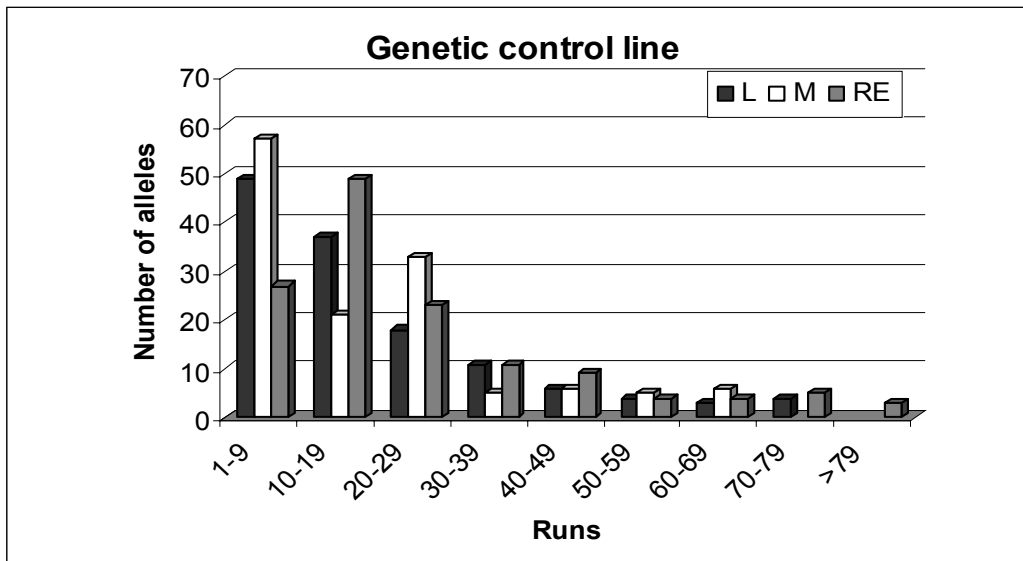


Figure 2.6. Number of alleles and the number of runs in which they appear out of 100 runs of gene drop for final lambs (L), matings (M), and rams and ewes (RE) in the selection line^a



^a Maximum number of alleles for L, M, and RE are 165, 165, and 166, respectively

Figure 2.7. Number of alleles and the number of runs in which they appear out of 100 runs of gene drop for final lambs (L), matings (M), and rams and ewes (RE) in the genetic control line^a



^aMaximum number of alleles for L, M, and RE are 132, 133, and 135, respectively

Chapter III.

Effects of inbreeding on fertility, lambing date, survival and weights in a crossbred sheep flock

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ABSTRACT: This study analyzed the effects of inbreeding on reproductive and production traits in a sheep flock started in 1983 with 50% Dorset, 25% Rambouillet, and 25% Finnsheep breeding. Inbreeding coefficients were calculated from a pedigree containing 4383 animals present since formation of the population in 1983. Three-way crosses were created at that time, and the flock subsequently remained closed. In fall 1987, the flock was divided into fall-lambing selection, fall-lambing environmental control, and spring-lambing genetic control lines. Inbreeding effects were estimated from 2678 lambs and 556 dams present after the creation of the respective lines. The mean, SD, and range of inbreeding coefficients were 3.8, 4.5, and 0 to 29%, respectively. The effects of lamb inbreeding on birth, 60-d, and 120-d weights were -0.012 ± 0.006 ($P < 0.05$), -0.045 ± 0.020 ($P < 0.05$), and -0.130 ± 0.034 kg/% ($P < 0.01$). Dam inbreeding had non-significant smaller effects on birth, 60-d, and 120-d weights of -0.008 ± 0.010 , -0.033 ± 0.034 , and -0.087 ± 0.056 kg/%, respectively. Spring-fertility decreased by 0.70 ± 0.30 %/% ($P < 0.01$) inbreeding of the ewe. Effects of lamb or dam inbreeding were not significantly different from zero for lambing date or survival to 1, 3, or 14 d.

Key Words: Inbreeding, Fertility, Survival, Body weight, Sheep

Introduction

Inbreeding accumulates in any closed population because of the mating of related individuals (Falconer and Mackay, 1996). In most populations of animals, inbreeding results in inbreeding depression, which in domestic animals can lead to a decrease in selection response and in potential genetic gains in economic traits. Measurement of the effect of inbreeding on these traits is important in order to estimate the magnitude of change associated with increases in inbreeding. Once the impact of inbreeding has been estimated, its economic impact upon a particular trait can be determined.

This study was designed to analyze the effects of inbreeding in a closed population of crossbred sheep. The objectives of this study were to estimate the effects of lamb and ewe inbreeding on spring-fertility, lamb survival, lamb weights, and fall-lambing date.

Materials and Methods

Animals, management, and experimental design

Details of the breeding structure were described in Chapter 2. Briefly, an experimental sheep flock was established in 1983 with 50% Dorset, 25% Rambouillet, and 25% Finnsheep

breeding. The population was closed in 1987, and all animals born following that year were progeny of three-way crosses. The flock was divided into selection (S), environmental control (E), and genetic control (G) lines. Of all animals available in 1987, 125 ewes and 10 rams were randomly chosen for the fall-lambing S line, 55 ewes and 5 rams were chosen for the fall-lambing E line, and 45 ewes and 5 rams were chosen for the spring-lambing G line. Each ram within each line was exposed to an approximately equal number of ewes, and selection for spring-fertility continued until the 1998 lambing (Al-Shorepy and Notter, 1997). The entire pedigree, back to the foundation animals present in 1983, consisted of 4383 animals. The flock used in this study, from 1988 to 1998, consisted of 2678 lambs, 556 dams, and 95 sires with 2138 exposures and 1445 lambings.

Ewes in S and E were exposed to rams for breeding in single-sire pens from May 1 to June 30. Ewes that failed to conceive were given a second chance to breed from August 1 to August 30. Ewes in G were given 42 d to breed beginning around October 1 (Al-Shorepy and Notter, 1996). A fertility score of 1 was given to ewes that lambed and a score of 0 to those that did not. An enclosed barn was used for lambing, and lambing date, number of lambs born, sexes of lambs, and lamb birth weights were recorded at parturition. All surviving lambs were weighed at approximately 60 (weaning), 90, and 120 d of age (Al-Shorepy and Notter, 1996). Survival was measured to 1, 3, and 14 d; scores of 1 for a lamb that survived and 0 for a lamb that did not were assigned. A few animals from triplet or larger litters were sold shortly after birth. These lambs were assumed alive up to 3 d but were not included in the 14 d analysis of survival.

Statistical analysis

Effects of dam and lamb inbreeding were estimated using the restricted maximum likelihood software (REML) of Boldman et al. (1993). The traits analyzed were birth, 60-d, and 120-d weights, spring-fertility, lambing date, and survival to 1, 3, and 14 d. For weights, a model was fitted to the data to account for fixed effects for sex of the lamb (for birth and 60-d weight) and contemporary group, and random additive direct, additive maternal, permanent environmental maternal, and litter effects (for birth weight only), as well as continuous effects of inbreeding coefficients of the lamb and dam and lamb age (for 60-d and 120-d weight). Sex of the lamb was included in the contemporary group definition for 120-d weight. Weights were pre-adjusted for type of birth (single, twin, or more), age of dam, and type of rearing (60-d and 120-d weights only) using multiplicative adjustment factors derived from these data. A model for fertility and date of lambing was developed to include fixed effects for ewe age and year, and random additive, permanent environmental (fertility only), and service sire by year (i.e., breeding pasture) effects, as well as continuous effects of inbreeding coefficient of the ewe. For survival, a model was developed with fixed effects for sex of the lamb, type of birth, ewe age, and year, and random additive direct, additive maternal (1-d survival only), permanent environmental (1 and 14-d survival), and litter effects, as well as continuous effects of lamb and dam inbreeding and birth weight (linear and quadratic, 3 and 14-d survival).

Results and Discussion

Inbreeding coefficients ranged from 0 to 29 % in both lambs ($\bar{x} = 3.8$, SD 4.5%) and dams ($\bar{x} = 2.2$, SD 3.2%). The flock had a generation interval of 2.4 ± 0.1 yr with an inbreeding rate of $1.3 \pm 0.1\%$ per generation. The linear partial regression coefficients for weights (Table 3.1) showed a decrease in weight as inbreeding increased in both the lamb and dam. Inbreeding of the lamb significantly affected weight at birth, 60 d, and 120 d. Dam inbreeding effects were negative, but not significantly different from zero for weight of lamb at birth or weaning, but effects of dam inbreeding on lamb weight at 120 d (-0.087 kg/%) approached significance ($P = 0.06$).

Most studies, including this one, have found that inbreeding of the lamb had a greater impact on weight traits than did the inbreeding of the dam. Effects due to inbreeding of the dam on birth weight and 60-d weight reported by Lamberson et al. (1982) were comparable to those estimated from this study, and effects of lamb and dam inbreeding on weight traits (Table 3.1) were very close to the average of studies reviewed by Lamberson and Thomas (1984), and to estimates by Mandal et al. (2002), Analla et al. (1998, 1999), and Weiner et al. (1992). Less negative effects due to inbreeding of the lamb and dam were reported by Boujenane and Chami (1996) for birth weight in two Moroccan sheep breeds, possibly due to the smaller adult size of the breeds. Ercanbrack and Knight (1991) reported an average inbreeding effect on 120-d weight in multiple lines of Rambouillet, Targhee, and Columbia sheep of -0.106 kg/% for lamb inbreeding and -0.015 kg/% for dam inbreeding, which were slightly less negative than those found in this study. Lamberson et al. (1982) reported a significant positive effect of inbreeding of the lamb on birth weight (0.022 kg/%) and positive but non-significant effects of inbreeding on 60-d weight, which they attributed to selection for body size in the Hampshire flock used in that study.

Inbreeding of the ewe decreased spring-fertility by 0.70 ± 0.30 %/% F increase in inbreeding ($P < 0.01$). A larger negative effect of inbreeding of the dam on fall-fertility (-1.2 %/% F) was reported in a study involving Hampshire sheep (Lamberson et al., 1982), although these estimates may not be directly comparable since they analyzed fertility in a different season. The smaller effect of inbreeding on spring-fertility, as compared to that on fall-fertility, may be due to the higher heritability for the former. Spring-fertility was estimated to have a heritability of 0.09 (Al-Shorepy and Notter, 1996) which is higher than most estimates for fall-fertility (Rosati et al., 2002; Matos et al., 1997; Bunge et al., 1990). Since spring-fertility was the trait under selection, frequencies of alleles with a negative effect could have decreased through artificial selection prior to significant inbreeding accumulation, resulting in a reduced amount of inbreeding depression.

The effect of inbreeding of the ewe on lambing date was not significant (Table 3.1), but was slightly negative, which may have contributed to decreased birth weight with increasing inbreeding. This result conflicts with the significant positive estimate of 0.346 d/% found by Lamberson et al. (1982), in a flock of Hampshire sheep, who discovered a later lambing date and a positive effect of inbreeding on birth weight. Both were attributed to the increased gestation interval, but effects on lambing date could have resulted from later conception due to inbreeding.

Survival of the lamb was not significantly affected by lamb or dam inbreeding at 1 d after birth or when birth weight was included as a covariate, at 3 or 14 d (Table 3.1). Birth weight,

fitted as a quadratic, did significantly affect survival at 3 d ($-0.028 + 0.006 \text{ kg/\%}$) and 14 d ($-0.032 + 0.007 \text{ kg/\%}$). Most previous studies measured the effect of inbreeding on survival to 90 d or more. A majority of these studies showed a decrease in survival or a non-significant effect of increasing inbreeding of the lamb and a smaller decrease or non-significant effect due to the inbreeding of the dam (Boujenane and Chami, 1997; Ercanbrack and Knight, 1991; Lamberson et al., 1982; Lamberson and Thomas, 1984). Galal et al. (1981) did not estimate the linear effect of increasing inbreeding, but did report the effect of inbreeding on survival at different inbreeding levels. They found that inbreeding of the dam had no effect on survival up to 7 d. Inbreeding of the lamb had no effect on survival to 7 d up to an inbreeding level of 5%, decreased survival from 5 to 15%, increased survival slightly but not significantly from 15 to 20%, decreased survival from 20 to 25%, and increased survival again when the inbreeding level was greater than 25%. There were relatively many animals in the $F > 25\%$ category, so small numbers cannot explain the result. The non-linear effect of inbreeding could be explained by natural selection removing less fit homozygotes at moderate levels of inbreeding, thereby leaving animals with beneficial or neutral homozygous combinations at higher levels of inbreeding (Templeton and Read, 1984).

Implications

Although survival and lambing date did not seem to be affected significantly by inbreeding, fertility and weights at birth, 60 d, and 120 d showed a detrimental effect. The inbreeding effects of the lamb and dam found in this flock are only estimates, and could vary based on management conditions. For example, the effect of inbreeding on survival was not significant under the management conditions of this flock, but could show an increased effect under less intensive, range conditions. One result that was relatively consistent among studies, including this one, is that the animal's own inbreeding was found to have a greater negative impact on most traits than the inbreeding of the dam. Although the effects of inbreeding may not be apparent when inbreeding is low, the accumulation of inbreeding in a small population like this one may result in significant economic losses and decreased response to selection for fertility.

It is important to acknowledge that a range of values for effects of inbreeding on individual traits have been found in different breeds and different lines within a breed. Although most estimates fall within a limited range (Lamberson and Thomas, 1984), there is still much between-breed and within-breed variation in the effect of inbreeding on a particular trait (Analla et al., 1998, 1999; Boujenane and Chami, 1997; Weiner et al., 1992; Ercanbrack and Knight, 1981). The variation could be attributed to the number of detrimental alleles present in the founding population or line, in addition to differing environmental and management conditions.

Literature Cited

- Al-Shorepy, S. A., and D. R. Notter. 1996. Genetic variation and covariation for ewe reproduction, lamb growth, and lamb scrotal circumference in a fall-lambing sheep flock. *J. Anim. Sci.* 74:1490-1498.
- Al-Shorepy, S. A., and D. R. Notter. 1997. Response to selection for fertility in a fall-lambing sheep flock. *J. Anim. Sci.* 75:2033-2040.
- Analla, M., J. M. Montilla, and J. M. Serradilla. 1998. Analyses of lamb weight and ewe litter size in various lines of Spanish Merino sheep. *Small Ruminant Res.* 29:255-259.
- Analla, M., J. M. Montilla, and J. M. Serradilla. 1999. Study of the variability of the response to inbreeding for meat production in Merino sheep. *J. Anim. Breed. Genet.* 116:481-488.
- Boldman, K. G., L. A. Kriese, and L. D. Van Vleck. 1993. A manual for use of MTDFREML. ARS, USDA, Washington, DC.
- Boujenane, I., and A. Chami. 1997. Effects of inbreeding on reproduction, weights, and survival of Sardi and Beni Guil sheep. *J. Anim. Breed. Genet.* 114:23-31.
- Bunge, R., D. L. Thomas, and J. M. Stookey. 1990. Factors affecting productivity of Rambouillet ewes mated to ram lambs. *J. Anim. Sci.* 68:2253-2262.
- Ercanbrack, S. K., and A. D. Knight. 1981. Weaning trait comparison among inbred lines and selected noninbred and randomly bred control groups of Rambouillet, Targhee, and Columbia sheep. *J. Anim. Sci.* 52:977-988.
- Ercanbrack, S. K., and A. D. Knight. 1991. Effects of inbreeding on reproduction and wool production of Rambouillet, Targhee, and Columbia sheep. *J. Anim. Sci.* 69:4734-4744.
- Falconer, D. S., and T. F. C. Mackay. 1996. Introduction to Quantitative Genetics. 4th Ed. Longman Group, LTD., Harlow, Essex, UK. pp 5764.
- Galal, E. S. E., E. A. Afifi, I. S. El-Kimary, I. A. Ahmad, and A. F. Shawar. 1981. Lamb survival as affected by inbreeding and cross-breeding. *J. Agric. Sci. Camb.* 96:1-5.
- Lamberson, W. R., and D. L. Thomas. 1984. Effects of inbreeding in sheep: a review. *Anim. Breed. Abstr.* 52:287-297.
- Lamberson, W. R., D. L. Thomas, and K. E. Rowe. 1982. The effects of inbreeding in a flock of Hampshire sheep. *J. Anim. Sci.* 55:780-786.
- Mandal, A., K., P. Pant, P. K. Rout, S. A. Singh, and R. Roy. 2002. Influence of inbreeding on growth traits of Muzaffarnagari sheep. *Indian J. Anim. Sci.* 72:988-990.
- Matos, C. A. P., D. L. Thomas, D. Gianola, R. J. Tempelman, and L. D. Young. 1997. Genetic analysis of discrete reproductive traits in sheep using linear and nonlinear models: I. Estimation of genetic parameters. *J. Anim. Sci.* 75:76-87.
- Rosati, A., E. Mousa, L. D. Van Vleck, and L. D. Young. 2002. Genetic parameters of reproductive traits in sheep. *Small Ruminant Res.* 43:65-74.
- Templeton, A. R., and B. Read. 1984. Factors eliminating inbreeding depression in a captive herd of Speke's gazelle (*Gazella spekei*). *Zoo Biol.* 3:177-199.
- Weiner, G., G. J. Lee, and J. A. Woolliams. 1992. Effects of rapid inbreeding and of crossing of inbred lines on the body weight growth of sheep. *Anim. Prod.* 55:89-99.

Table 3.1. Partial regression coefficients for lamb and ewe inbreeding

Trait	Mean	SD	Lamb inbreeding coefficient	Dam inbreeding coefficient
Birth wt (kg)	3.5	1.0	-0.012 ± 0.006 kg/% *	-0.008 ± 0.010 kg/%
60d wt (kg)	20.4	5.1	-0.045 ± 0.020 kg/% *	-0.033 ± 0.034 kg/%
120d wt (kg)	37.9	7.9	-0.130 ± 0.034 kg/% **	-0.087 ± 0.056 kg/% †
Spring fertility (%)	47.5	49.9	—	-0.70 ± 0.30 %/% **
Lambing date (d) ^a	14.32	11.66	—	0.052 ± 0.092 d/%
Survival (%):				
1d	92	27	-0.01 ± 0.20 %/%	-0.05 ± 0.20 %/%
3d	90	30	0.10 ± 0.20 %/%	-0.10 ± 0.20 %/%
14d	87	33	0.10 ± 0.20 %/%	- 0.07 ± 0.20 %/%

^a Days after the first day of lambing.

† P < 0.10

* P < 0.05

** P < 0.01

GENERAL DISCUSSION AND IMPLICATIONS

The experimental design used in this study allowed an in-depth analysis of the effects of inbreeding on performance and genetic diversity in a crossbred population of sheep. With the creation of programs such as GENES (Lacy, 1994), any pedigree could be easily entered into the program and a diversity analysis performed. An evaluation of heterozygosity and allelic diversity would allow future selection studies to more firmly state if the selection and control lines are genetically similar and appropriate for comparison. Any producer or breeder with a small relatively closed population of animals could use a genetic analysis to pinpoint when and where genetic losses are occurring and potentially correct them and better maintain the current diversity present in the breeding stock. The analysis performed in this study could also be useful for maintaining endangered breeds of livestock to preserve as much genetic variation as possible.

Genetic diversity measured in final lambs, matings, and breeding animals available were similar between the three groups in S, but the measures between the groups were quite a bit different in G. These fairly large differences seen in G were due to breeding animals being retained but not bred, a longer generation interval, decreased culling, and a smaller census population. These parameters emphasize the importance of measuring the available breeding animals in the population. Although diversity measures can be taken on the lambs if accurate and complete records for the rams and ewes are not available and if the opposite of the parameters stated previously are in place.

Most estimations of f_e , f_a , and f_g , were comparable when calculated using the additive relationship matrix or simulations of gene drop. Thus, only one method of calculation may be necessary for an evaluation. The values obtained for f_g revealed a small discrepancy between calculation methods; the estimated f_g from gene drop (5.12 ± 0.92) was quite different from the value obtained from the additive relationship matrix (4.43). Lacy (1989) stated that the calculation using the relationship matrix for f_g should be the exact measurement of proportional diversity loss in the population. Upon re-evaluating the differences between f_g in lambs and matings, which should be relatively close, the average difference observed from gene drop was 0.28 genomes. The difference calculated from the relationship matrix was about two standard deviations from the mean difference in gene drop and had only been observed in 3 out of 100 simulations. The exact reason for the discrepancy is not known and may not be of any significance, but non-random mating may play an important role.

Lamb and ewe inbreeding had undesirable effects on birth, weaning, and 120-d weights, and fertility, and non-significant effects on survival and lambing date. Although the effect of lamb and dam inbreeding on weight traits seems to become more negative at higher weights, the effect as a percentage of the mean remains relatively constant. A somewhat stable effect of inbreeding of the animal as a percentage of the mean was also observed from birth weight up to body weight of the breeding ewe in other studies (Weiner et al., 1992; Ercanbrack and Knight, 1991). Inbreeding of the individual may have a similar effect over different ages, but it seems to have a varied effect over different inbreeding levels, sometimes with less negative effects at high levels of inbreeding (Weiner et al., 1992).

Spring-fertility, less heritable than weight, is more negatively affected in this study than birth, 60-d, and 120-d weights (as a percentage of the mean), but was less negatively affected than fall-fertility in other studies (Lamberson et al., 1982). Survival to 1, 3, or 14 d was not significantly affected by lamb or dam inbreeding, but survival in-utero could have attributed to a more significant effect. If inbreeding of the lamb or the dam decreased survival of the lamb in-utero, then the effect should have been attributed to the effect of dam inbreeding on spring-fertility. In order to more accurately determine the effects of inbreeding on survival and fertility, ewes should be checked for pregnancy shortly after breeding.

The values found for the effects of inbreeding on this flock are, for the most part, within the range of values for similar traits found in other studies, but the variety of effects due to inbreeding of the dam and lamb show a fair amount of between-breed and within-breed variation on a particular trait (Analla et al., 1998, 1999; Boujenane and Chami, 1997; Weiner et al., 1992; Ercanbrack and Knight, 1981). The differences observed between lines and(or) breeds could be attributed to the number of detrimental alleles present in the founding population or line as well as different management conditions. Lines or breeds that were found or developed to have a low impact of increased inbreeding on performance could be beneficial to producers and breeders of these animals.

Literature Cited

- Analla, M., J. M. Montilla, and J. M. Serradilla. 1998. Analyses of lamb weight and ewe litter size in various lines of Spanish Merino sheep. *Small Ruminant Res.* 29:255-259.
- Analla, M., J. M. Montilla, and J. M. Serradilla. 1999. Study of the variability of the response to inbreeding for meat production in Merino sheep. *J. Anim. Breed. Genet.* 116:481-488.
- Boujenane, I., and A. Chami. 1997. Effects of inbreeding on reproduction, weights, and survival of Sardi and Beni Guil sheep. *J. Anim. Breed. Genet.* 114:23-31.
- Ercanbrack, S. K., and A. D. Knight. 1981. Weaning trait comparison among inbred lines and selected noninbred and randomly bred control groups of Rambouillet, Targhee, and Columbia sheep. *J. Anim. Sci.* 52:977-988.
- Ercanbrack, S. K., and A. D. Knight. 1991. Effects of inbreeding on reproduction and wool production of Rambouillet, Targhee, and Columbia sheep. *J. Anim. Sci.* 69:4734-4744.
- Lacy, R. C. 1989. Analysis of founder representation in pedigrees: founder equivalents and founder genome equivalents. *Zoo Biol.* 8:111-123.
- Lacy, R. C. 1994. GENES: A computer program for pedigree analysis and population management. Chicago: Chicago Zoological Soc.
- Lamberson, W. R., D. L. Thomas, and K. E. Rowe. 1982. The effects of inbreeding in a flock of Hampshire sheep. *J. Anim. Sci.* 55:780-786.
- Weiner, G., G. J. Lee, and J. A. Woolliams. 1992. Effects of rapid inbreeding and of crossing of inbred lines on the body weight growth of sheep. *Anim. Prod.* 55:89-99.

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