



Genetic aspects on the viability of the Scandinavian wolf population A report from SKANDULV

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This report is a complement to the report on demographic viability of the Scandinavian wolf population from SKANDULV to the Swedish Environmental Protection Agency of the 2nd July 2012.

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Assignment

This assignment from the Swedish Environmental Protection Agency (SEPA) to SKANDULV asks for a genetic complement to the demographic PVA that SKANDULV earlier this year produced and reported to SEPA July 2nd 2012 (Chapron et al. 2012). The complement should be based on the most recent relevant scientific literature and knowledge, and on relevant analyses made in various earlier reports concerning the Scandinavian wolf population. The complement shall clarify the correlations between population size and rate of genetic inflow respectively, with inbreeding levels and retention of genetic variation for the Scandinavian wolf population. The results should be useful for determining the demands for the Scandinavian wolf population to attain so called Favourable Reference Population in accordance with the European Union's Species and Habitat Directive.

The original assignment (in Swedish) is presented in Appendix 1.

Svensk sammanfattning (Summary in Swedish)

Den 2 juni 2012 levererade SKANDULV en demografisk sårbarhetsanalys (PVA) för den skandinaviska vargpopulationen till Naturvårdsverket. Vid mitten av september fick SKANDULV ett nytt uppdrag, att komplettera den demografiska analysen med genetiska aspekter som skulle kunna ligga till grund för fastställande av kraven för att den skandinaviska vargpopulationen ska uppnå s.k. 'Favourable Reference Population', i enlighet med Europeiska Unionens Art- och Habitatdirektiv. Rapporten skulle vara klar 12 oktober. Uppdraget finns i Appendix 1. På grund av den snäva tidsfristen fanns inte tid att göra en fullständig PVA, men vi framför även argument varför en genetisk PVA är problematisk. Argument för detta finner vi i en genomgång av den mest relevanta litteraturen i ämnet de senaste 20 åren. De två viktigaste aspekterna att beakta i en genetisk sårbarhetsanalys är inavelsdepression och förlust av evolutionär potential på grund av reducerad variation i genomet som beror på så kallad genetisk drift. Den genetiska driften är starkare ju mindre populationen är. Problemet med att föra in dessa två processer i en sårbarhetsanalys ligger i att kvantifiera deras effekt på risken för utdöende av populationen. Inavelsdepressionens relation till utdöenderisk går att kvantifiera, men problemet här är att det sällan finns tillräckliga data. Den skandinaviska vargpopulationen är kanske världens bäst undersökta vilda däggdjurspopulation ur ett genetiskt perspektiv. Vi har till och med kvantifierat ett par aspekter av inavelsdepressionen i stammen. Trots detta finns inte tillräckliga data för att fullständigt kvantifiera inavelsdepressionen. Vill man föra in inavelsdepression i en klassisk sårbarhetsanalys blir vi tvungna att göra antaganden om dess omfattning. Pessimistiska antaganden ger en större Minsta Livskraftig Population (MVP), optimistiska antaganden ger en mindre. Ännu värre är det med förlust av genetisk variation genom drift. Här finns överhuvudtaget ingen kvantifierbar relation till utdöenderisk.

När det gäller genetikens betydelse för en populations livskraft är det inte bara populationens storlek som är av betydelse, utan också dess förbindelse med andra populationer, dvs. hur stort inflöde av migranter man har till populationen. Ett alternativ till att bygga sin genetiska PVA på utdöenderisk är då istället att beräkna den kombinerade effekten av populationsstorlek och migration på nivån av inavel och förlust av genetisk variation, och sedan överlåta till beslutsfattaren att avgöra vilken nivå av inavel och förlust av genetisk variation man kan acceptera. Det är denna väg vi valt i denna rapport. Eftersom populationsstorleken i genetiska analyser vanligen uttrycks som s.k. 'genetiskt effektiv populationsstorlek' (N_e), har vi undersökt hur relationen mellan denna och den 'verkliga' populationen (N_c) ser ut. Olika beräkningar för varg i Sverige och Finland har gett värden på kvoten

N_e/N_c i intervallet 0.18 – 1.47. De flesta värdena är från växande populationer vilket ger högre värden för kvoten. En simulering för en stabil population gav värden på 0.24, vilket vi betraktar som något konservativ och därför föreslår användandet av kvoten 0.25.

För att visa på de genetiska effekterna för vargpopulationen av olika nivåer på populationsstorlek och migration, utgår vi från några redan publicerade analyser av relationen mellan populationsstorlek och migration å ena sidan och inavelsnivåer och bevarande av genetisk variation å den andra. De invandringsfrekvenser som nämns här gäller antalet effektiva invandrare (i fortsättningen kallade migranter), dvs. migranter som går in i aveln och bidrar med lika mycket till denna som en genomsnittlig ynglande individ i populationen. En annan förutsättning för att de presenterade sambanden ska gälla är att migranterna kommer från en mycket stor population. Detta innebär t.ex. att den skandinaviska djurparkspopulationen av varg kan bidra med nya individer upp till en viss gräns, därefter krävs tillskott från en större genetisk pool.

I små populationer ökar inaveln med tiden, men förekommer det en viss konstant invandringsfrekvens, till exempel en migrant vartannat år, uppstår efter en viss tid en stabilisering eller jämvikt av inavelsgraden, som är oberoende av populationsstorleken. De två modellerna som använts visade likartade resultat. Det krävs två migranter per varggeneration (ca 5 år) för att nå ned till stabila nivåer på inavelsnivån runt 15 % (inavelskoefficienten $F = 0.15$). För att komma ned till 5 % inavel ($F = 0.05$) krävs minst en migrant varje år. Det tar dock ganska lång tid för att uppnå dessa jämviktsvärden, någonstans mellan 80 och 100 år i båda fallen. Den ena modellen visar dock att denna process kan påskyndas, om man inleder med en stor ”stötdos” av nya individer på kort tid (kan förmodligen endast uppnås genom artificiell inflyttning). Med en stötdos på 10 djur de första åren kan man komma ned till $F = 0.15$ på 50 – 60 år om den fortsatta invandringen är 2 per generation. Med en stötdos på 20 djur kommer man ned till $F = 0.15$ på mindre än 10 år, och ned under $F = 0.1$ på 30 år, om det fortsatta inflödet är en individ per år. Dessa processer är alltså oberoende av den aktuella populationens storlek, men migranterna måste komma från en mycket stor population.

Bevarandet av genetisk variation är dock beroende av både populationens storlek och invandringsfrekvens. Ett vanligt sätt att uttrycka genetisk variation är graden av heterozygoti (som kan variera mellan 0 och 100 %) i populationen. Det finns inte någon självklar nivå eller tröskelvärde här som vetenskapligt har visats vara avgörande för populationens långsiktiga livskraft, men ett, som det verkar allmänt vedertaget, förslag är 95 procents bevarande på 100 år. För att uppnå denna nivå utan invandring krävs uppemot 800 djur ($N_e = 200$). Med 4 migranter per varggeneration kan man precis nå denna nivå även med en så liten population som 100 – 200 djur, medan man med 5 migranter per generation (1 per år) nästan är oberoende av populationsstorlek. Kommer man å andra sidan under 3 migranter per generation är det svårt att nå denna nivå oavsett populationsstorlek.

Vi lämnar inga färdiga förslag till myndigheterna vad som krävs för att populationen ska uppnå genetisk livskraft, eller nivå för 'Favourable Reference Population'. Vi hoppas dock att denna rapport klart visar att kontakt med annan population, ju större ju bättre, är viktigare än populationsstorleken i sig. Detta illustreras av att med en invandringsfrekvens på en migrant per år kan inavelsgraden på sikt reduceras ned till runt 5 % oberoende av populationsstorlek, och 95 procents heterozygoti kan bevaras på 100 år även med en mycket liten populationsstorlek.

Problems with genetic PVA's

This complementing report is not based on a formal population viability analysis (PVA) leading to levels for a minimum viable population (MVP) including genetics. The most important reason for this is that the time frame given for the assignment simply did not allow for a complete analysis. But there are also methodological reasons for a critical attitude towards classical PVAs including genetics. The importance of genetics for extinction risk has been pointed out since the pioneering works by e.g. Franklin (1980) and Shaffer (1981). Although it has been questioned (Lande 1988, Caughley 1994, Caro and Laurenson 1994), today there seems to be little dispute within the scientific community about the link between genetics and extinction risk (e.g. Allendorf and Ryman 2002, O'Grady et al 2006, Jamieson 2007, Frankham 2010). However, there still is little and hard-to-get quantitative data to support this link (Jamieson and Allendorf 2012), especially when compared with possibilities to obtain demographic data for PVAs. Therefore genetic PVAs generally are much more dependent on assumptions. Optimistic assumptions will yield small MVPs, whereas pessimistic assumptions will yield large MVPs. We will here develop a little further on this problem and argue for another approach than estimating a population size needed to minimize the extinction risk.

There are mainly two genetic factors leading to an increased risk for extinction, inbreeding depression and loss of genetic variation (Frankham 1995a, 2005). A third factor is accumulation of deleterious mutations, so called mutational meltdown (Lynch et al. 1995), but it seems to be of less importance in conservation biology (Frankham 2005: "*Thus, the impact of mutational accumulation on extinction risk appears to be less important than the other genetic factors and to take very many generations*"). This was also the opinion of the expert meeting on genetics of the Scandinavian wolves held in Färna 2002 (Liberg 2005). We therefore disregard this aspect in this report.

The mechanism behind inbreeding depression is an increasing degree of homozygosity due to the fact that parents are closely related. This allows for increased expression of recessive deleterious alleles. Fitness of inbred populations will also be affected by decreased heterozygosity at certain loci with so called overdominance (e.g. Allendorf and Ryman 2002), but this is regarded as less important (Charlesworth and Charlesworth 1987) and is rarely accounted for in PVAs, possibly because it is very difficult to quantify, and will therefore be dismissed here. The negative effects of inbreeding in a specific population will depend not only on the actual inbreeding level, but also on how many deleterious alleles there are in the population, and their respective impact, the so called genetic load. The genetic load is often standardized as the mean number of "lethal equivalents" per gamete (Allendorf and Ryman 2002). This is normally the currency for inbreeding depression that is put into PVA models, e.g. VORTEX (Lacy 1993). However, the number of lethal equivalents is rarely known for any specific population, why often default values are used. The recent VORTEX manual (Lacy 2009) suggests 3.14 as a default value, taken from a study of captive populations (Ralls et al. 1988). However, there are other suggestions, e.g. 12 lethal equivalents, taken from Keller (1998) and suggested by Frankham (2005) to be more realistic as a default value. We have earlier calculated a genetic load of 6.04 'litter reducing equivalents' for the Scandinavian wolf population (Liberg et al. 2005), but that was based only on depression of litter sizes, which most likely is only part of the genetic load in this population. There are reported values as low as 0.63 and 0.71 for captive red and Mexican wolf populations (Fredrickson & Hedrick 2002), but in a more recent report a load of 5.19 litter-reducing equivalents were measured for wild Mexican wolves (Fredrickson et al. 2007). There is thus a large variation in this measurement between species and populations, as well as between individuals within a population. Thus, a newly established population, founded by a small number of individuals like the Scandinavian wolf population (five founders), may have a genetic load that

deviates, not only from other species and populations, but also from its own source population, making an assumption on this measurement even more uncertain.

If it is difficult to quantify the effect of inbreeding on extinction risk, it is even harder to do it with loss of genetic variation, if possible at all. Jamieson and Allendorf (2012) are rather categorical on this point: “...*the effects of inbreeding depression on individual fitness and population growth can normally be incorporated in standard PVAs, such as those generated by VORTEX, but the potential harmful effects of loss of adaptability on population viability cannot...*”. The problem with loss of genetic diversity is that it increases extinction risk by adversely affecting the ability of populations to adapt to environmental change, or in other words, it hampers the “evolutionary potential” (Frankham 2005). This is by definition a problem on very long term, which makes it hard for scientists to observe in species with longer generation times than a couple of weeks. The negative effects of this factor of course also is completely dependent on what kind of environmental change the population will meet in the future, something that is just as difficult to predict as future catastrophes.

In spite of the problems with estimates of MVP, there has been quite a few produced during the last decades. Recently, there have been performed three different meta-analyses of estimates of MVP (Reed et al. 2003, Brook et al. 2006, Traill et al. 2007). Using various methods they all tried to standardize the various MVP estimates, covering several hundred of species and a broad variety of taxa, from mammals to plants (Reed et al. 2003 only included vertebrates). A summary review of all three studies (Traill et al. 2010) concluded that there was rather little variation between different taxa and suggested 5000 animals would be a good general ‘rule of thumb’ for an MVP for any species. In a popular summary this figure was even coined ‘a magic number’ that applies to ‘mammals, amphibians, insects, plants and the rest’ (Clabby 2011). This figure actually is in concordance with the upper value of the so called ‘50-500 rule’, suggested already in 1980 (Franklin 1980). The lower value (50) was based on experiences of animal breeders and concerns inbreeding depression, while the upper value (500) was based on the theoretical population size required to balance the loss of additive genetic variation due to genetic drift by the creation of new genetic variation due to mutation (Jamieson and Allendorf 2012). Both values are expressed as ‘genetic effective population sizes’ (N_e). A generally used default ratio between N_e and N_c (actual, or censused, population size) is 0.1 (Frankham 1995b), giving the upper MVP threshold of the 50-500 rule a value of 5000 animals (this default ratio has been questioned, and we will come back to it further down in this report). Actually, the upper value of 500 in the ‘rule’ also has been questioned. It has been argued that the equilibrium between drift and positive mutations rather is in the region of N_e 1000 to 5000, giving MVP thresholds up towards 10 000 to 50 000 animals (Lande 1995), but there does not seem to be a consensus on this (Franklin and Frankham 1998, Lynch and Lande 1998).

The ‘magic number’ of 5000, and the different meta-analyses cited above supporting it, has been heavily criticized on methodological grounds (Flather et al. 2011a, b, Garnet and Zander 2011). Among other things, the critics point out that the lack of variation between taxa is masked by a very large variation within taxa, and even between populations of the same species, strongly cautioning against any generally applicable threshold value for MVP. However, in spite of the dispute over the usefulness of a single universal value for MVP, there seems to be a general consensus about a need for thousands rather than hundreds of individuals for long-term evolutionary persistence (Frankham 2010, Traill et al 2010, Flather et al 2011b, Brook et al 2011, Jamieson and Allendorf 2012, but see Garnet and Zander 2011). However, it has also been pointed out that the need for evolutionary persistence refers to the global rather than the local population as long as there is some gene flow into the latter (Hoegh-Guldberg et al. 2008, Jamieson and Allendorf 2012). This important point that often is overlooked was also stressed by the international expert workshop on wolf genetics at Färna in 2002 (Liberg 2005). It turns the attention away from focus on population size per se, to the demand for

genetic inflow. And this is the approach we have chosen for this genetic supplement to our earlier report on demographic viability of the Scandinavian wolf population. By using earlier analyses for this wolf population, we discuss the consequences of various immigration scenarios for inbreeding levels and for loss of genetic variation. But first the ratio between effective population size (N_e) and the real, or censused (N_c), population size has to be discussed.

Ratio effective population size/censused population size (N_e/N_c)

The concept of an “ideal” or “effective” population is important in genetics and goes back all the way to Sewall Wright and Ronald Fisher, together with John Haldane the “fathers” of theoretical population genetics. It is a kind of standardized measure of population size useful in all kinds of genetic analyses, e.g. when analyzing effect of population size on changes of inbreeding levels or for loss of additive genetic variation in small populations through genetic drift. Often MVP’s that include genetics are expressed in terms of effective population, e.g. ‘the 50-500 rule’. Jamieson and Allendorf (2012) provided this definition: - “an ‘ideal’ (Wright–Fisher) population of N individuals is one of a constant size with non-overlapping generations in which the next generation is produced by drawing $2N$ genes at random from a large gamete pool to which all individuals contribute equally. This model also assumes selective neutrality so that allele frequencies change only by genetic drift” -. The effective population size, usually designated N_e , normally is substantially smaller than the corresponding ‘real’ or ‘censused’ population (N_c). As some of the ‘rules’ or results discussed and presented in this report are expressed in the currency of effective population size, its relation to the real population size, i.e. the ratio N_e/N_c is of great importance. This ratio will vary substantially between species and also between populations of the same species, due to demographic and social characteristics of the population. Frankham (1995b) reported an average ratio of 0.1 from a review of more than 100 natural populations of animals and plants, but also a large variation in the ratio, from 0.00001 in a marine mollusk to greater than 0.30 for some low fecund species.

There has been made several estimates of the N_e/N_c ratio for the Scandinavian wolf population. Andrén (2005) used a method originally suggested by Harris and Allendorf (1989), by using demographic data from the Scandinavian wolf population in a VORTEX simulation, which resulted in a N_e/N_c ratio of 0.34 – 0.35. Bensch et al. (2006) reached a somewhat higher result (0.46) by analyzing variation in microsatellite allele frequencies between cohorts in the program NeEstimator (Peel et al. 2004). Based on the same type of data, but using four different models, Aspi et al. (2006) presented for the Finnish wolf population a ratio of 0.40 – 0.46, depending on the model used. All these estimates were calculated on data from expanding populations, which normally will give higher ratios than those calculated for stable populations or declining populations. This was very clearly illustrated in a more recent analysis by Aspi’s group (Jansson et al. 2012). They measured the ratio during different phases of the development of the Finnish wolf population, which expanded during 1994 – 2006, with the strongest growth during 2004 – 2006, and then had a sudden and strong decline 2007 – 2009. The so called ‘single sample’ methods gave ratios of 0.28 – 0.29 during periods of strong growth in the populations and 0.10 during declines. So called temporal estimates where allele frequencies in four different sub-periods were compared pairwise, gave ratios between 0.71 – 1.47 during the periods with strongest growth and 0.19 – 0.40 for the comparison that include the final decline in the population. However, when dealing with MVP’s, it is of greater interest to know the ratio N_e/N_c in a stable population. This was the case for the simulations made by Forslund (2009), which were made with an individual based model with demographic variables taken from the Scandinavian population. In the simulations the population was kept stable through regulating harvest. Simulations were made for two population levels, 230 and 400 respectively. Both yielded a N_e/N_c

ratio of 0.24. This is rather low compared with the ratios mentioned before, even considering effects of population growth. Still, being the only ratio calculated for a stable wolf population, we think it is relevant to use for the present analysis, but as we consider it conservative we find it justified by use the simpler ratio of 0.25, i.e. N_e will be 4 times larger than N_c .

Effect of immigration on inbreeding levels

We here will present the results of two recent reports on the effects of immigration on the future development of inbreeding levels for the Scandinavian wolf population. Ryman and Laikre (2009) used an analytical 'finite island model', not especially designed for any specific population but based on theoretical population genetic, described in Ryman and Leimar (2008). They ran their model with an effective population size of 50, which with a N_e/N_c ratio of 0.25 corresponds to 200 real individuals. Eight different migration scenarios were tested, running from zero to 20 immigrants per wolf generation. It was assumed that all migrants were "effective" i.e. that their contribution to breeding was the same as that of an average breeder in the population.

In Figure 1a, the population from which the migrants come (the donor population) is infinitely large. It is demonstrated that without immigration, the inbreeding level will continue to increase. However, even with the smallest immigration modeled (1 migrant per generation, corresponding to approximately 5 years for wolves), the average inbreeding coefficient (F) will decrease continuously during the whole 100-year period modeled, although slowly. Any immigration frequency will eventually lead to an equilibrium in the inbreeding level (Hartl and Clark 2007), but for this small immigration it was not reached even within the whole 100 year period. To reach down to the level of average $F = 0.1$ within the 100 year period it would require at least one migrant every second year, and to reach down to $F = 0.05$ it would require at least one migrant per year.

In Figure 1b, the same scenarios have been modeled, but with the difference that the donor population is of the same size as the receiving population ($N_e = 50$). Here we can see that the positive effect of immigration fades after some time, irrespective of the magnitude of immigration. This illustrates that immigration (or translocation) from a limited population, like the Scandinavian zoo population, will be positive for a limited time period, but sooner or later the provision of new genetic material will be exhausted, and inflow from a wider genetic pool will be necessary.

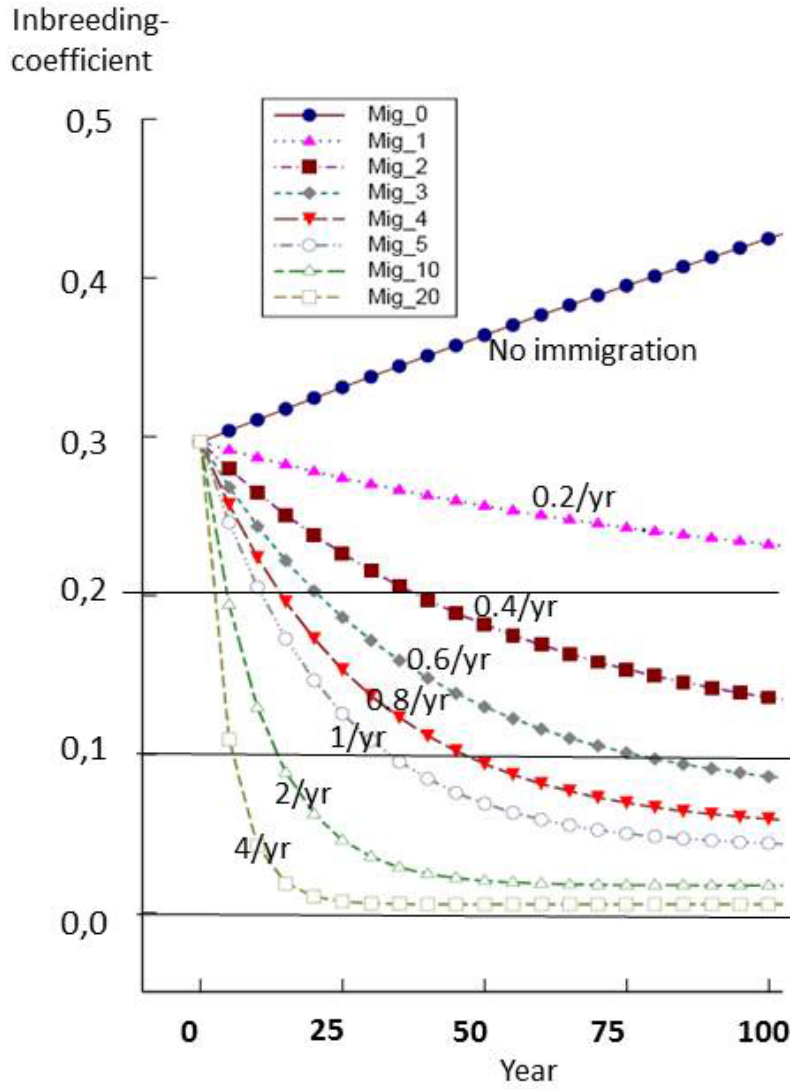


Figure 1a. Change in inbreeding coefficient over 100 years at different immigration rates (0 – 20 immigrants per generation). The donor population is indefinitely large, the receiving population has an effective population size of 50, corresponding to 200 individuals with an N_e/N_c ratio of 0.25. We have translated the time scale on the x-axis from generations in the original figure to years, and number of immigrants in the different scenarios is given as number/year directly on each scenario curve in the graph, assuming a wolf generation is 5 years. In the box the original currency (number of migrants per generation) is kept. (Modified from Ryman and Laikre 2009)

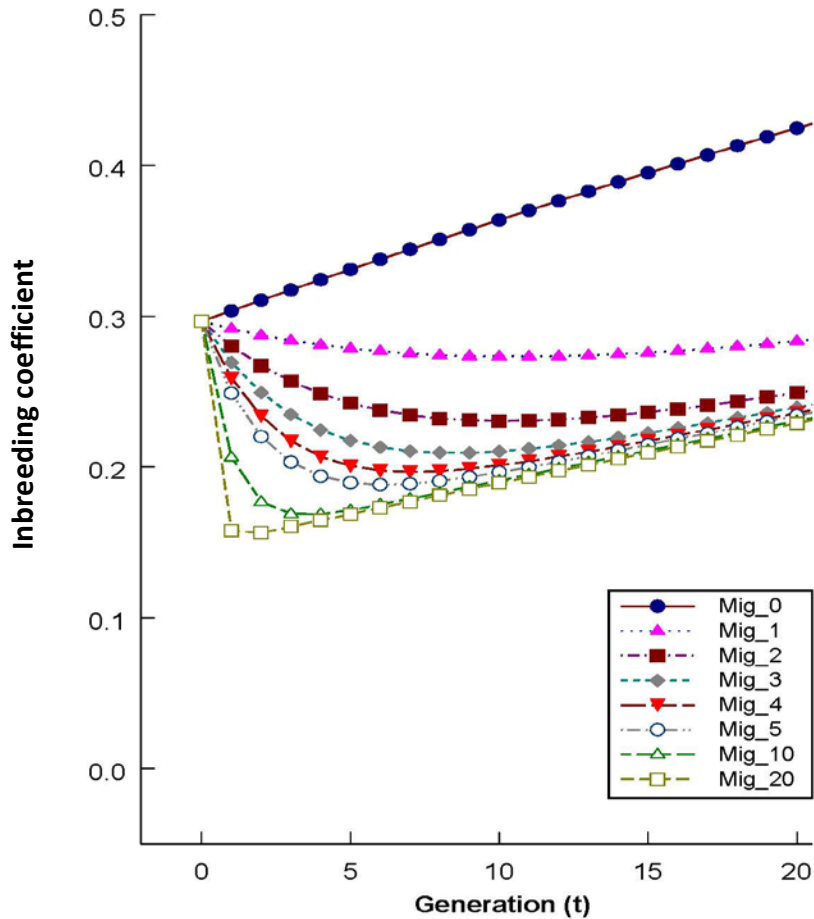
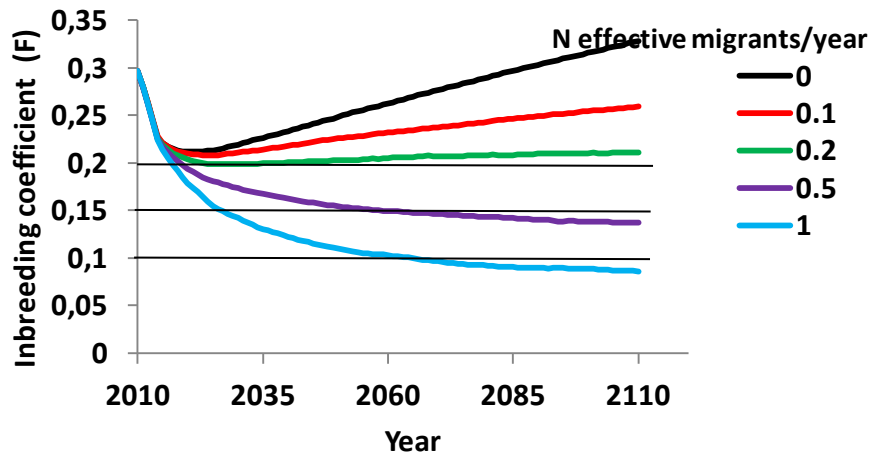


Figure 1b. The same as figure 1a, but here the donor population is of the same size as the receiving population ($N_e = 50$). Here no translation from generation time to years has been performed. This means that the figures given in the box refers to number of migrants per generation. (After Ryman and Laikre 2009)

Forslund (2010) used a simulation model specifically designed after the real Scandinavian wolf population, and based on data from this population, including data from the near complete pedigree established for this population (Liberg et al. 2005). He simulated change of average inbreeding coefficient 100 years into the future under different immigration scenarios, and ran 500 simulations for each immigration scenario. The size of the receiving population was held constant at 240 (real) individuals, and there was no limit on the donor population. Five different immigration scenarios within the range of 0 – 1 migrant per year were simulated. One important difference from Ryman/Laikre's scenarios was that Forslund started all five scenarios with an inflow boost of either 10 or 20 migrants the first four years. The reason for this was that the Swedish government in November 2008 granted permission to the managing agency to translocate up to 20 wolves from other wolf populations into the Swedish population within a time frame of five years.

a)



b)

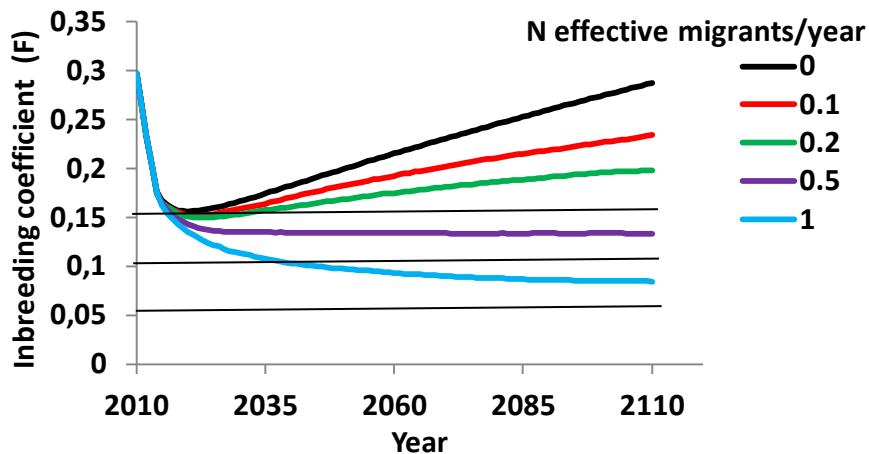


Figure 2. Change of inbreeding coefficient (F) over 100 years at different immigration scenarios, after an initial translocation boost of 10 migrants (a) and 20 migrants (b) the first 4 years, followed by a continuous inflow according to the different scenarios. (After Forslund 2010)

Overall Forslund's simulations support the results of Ryman/Laikre's model. The difference is the initial boost of migrants in Forslund's models. This brings the inbreeding level down to 0.20 within the first few years with a boost of 10 migrants, and down to just below 0.15 with the higher boost of 20 migrants. After this initial effects of the translocation boosts, the development of the different scenarios is similar to those demonstrated by Ryman/Laikre. A continued inflow of one migrant per generation (5 years) will stabilize average F at a level around 0.20, while an inflow of one migrant every second year will stabilize F around 0.13. To further reduce F towards 0.05, at least one migrant per year is needed. The equilibria are independent of the initial boosts, but the time to reach them will differ.

Effect of immigration on loss of genetic variation

Whereas the equilibrium between inbreeding level and immigration rate is independent of population size (Hartl and Clarke 2007), the loss of genetic variation is determined by a combined effect of population size and immigration rate. Ryman and Laikre (2009) investigated this relation, using the same basic model as described above. They investigated two cases, one where the immigrants came from an infinitely large donor population, and one case where the immigrants came from a population of the same size as the receiving population. For each case they plotted the relationship between immigration frequency and remaining genetic variation after 20 generations, under four different population levels in the receiving population (N_e 25 – 200). Genetic variation was expressed as heterozygosity in the population. Heterozygosity is proportional to the amount of genetic variance at loci affecting the quantitative variation, and thus regarded as a good predictor of the potential for population to adapt to environmental changes (Allendorf and Ryman 2002).

Remaining heterozygosity

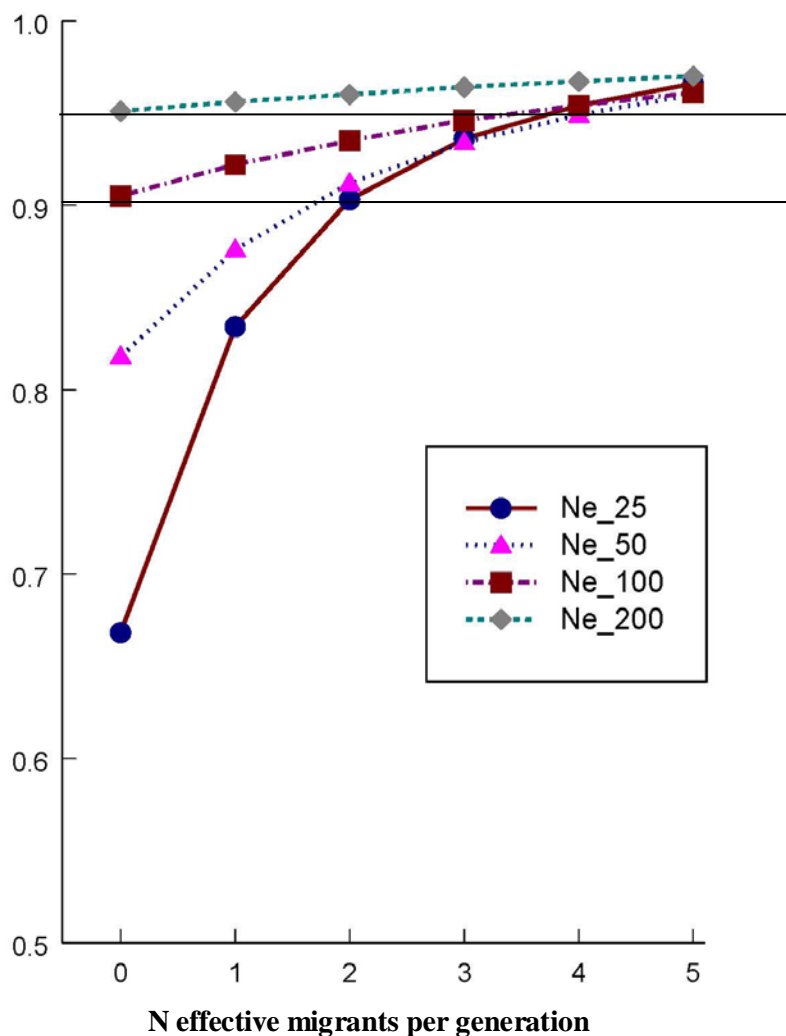


Figure 3a. Remaining degree of heterozygosity $[H(t=20)/H(t=0)]$ after 20 generations (≈ 100 years) at different effective population sizes (N_e) and for different immigration frequencies. Migrants come from an infinitely large population.

There is no broad consensus how much genetic variation should be preserved for a specified time, but Allendorf and Ryman (2002) suggested that 95 % heterozygosity should be retained over 100 years. According to the present model this will be achieved without any immigration at all if the effective population size is above 200, corresponding to 800 wolves in our case (Figure 3a and 3b). At lower populations this is achieved with 4 migrants per generation, and almost achieved with 3 migrants per generation, provided that the migrants come from a large population. With less than 2 migrants per generation, the effective population has to be at least 100 (400 real wolves) to retain more than 90 % of its heterozygosity.

If migrants come from a similar sized population, the receiving population has to be at least N_e 100 to retain 90 % or more of its heterozygosity (Figure 3b). At lower populations, no migration rate will be enough to obtain even 90 % retained heterozygosity.

Remaining heterozygosity

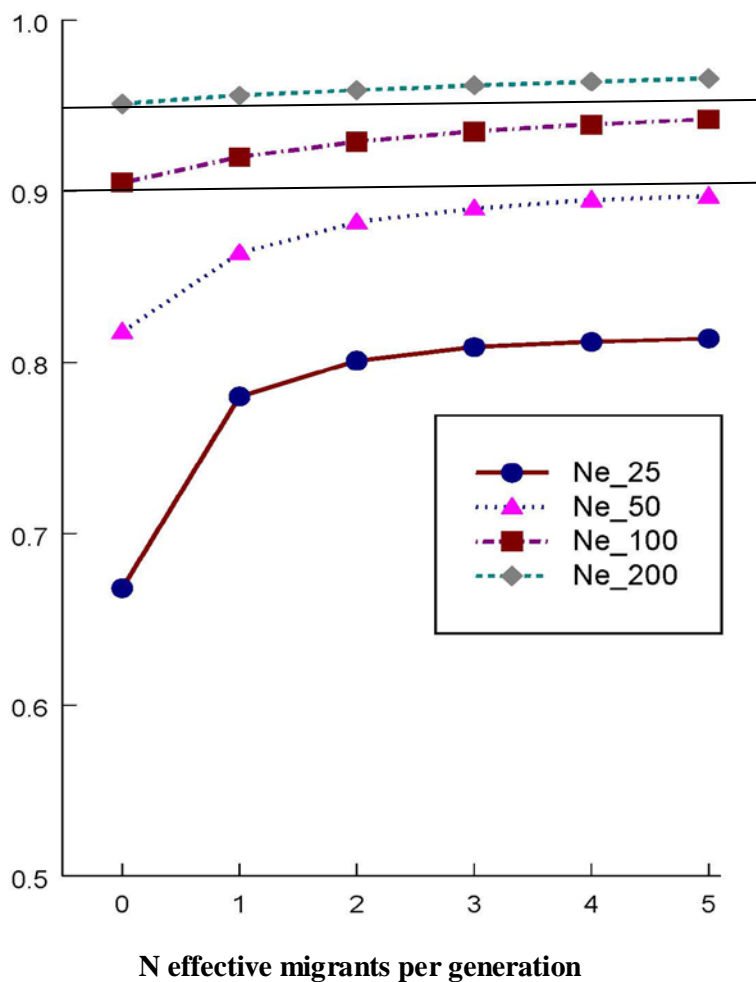


Figure 3b. Remaining degree of heterozygosity $[H(t=20)/H(t=0)]$ after 20 generations (≈ 100 years) at different effective population sizes (N_e) and different immigration frequencies. Migrants come from a population with the same size as the receiving population

Concluding remarks

We will not here make any recommendations as to what ambition or goal there should be for the Scandinavian wolf population, or what population size or frequency of immigration is needed for its viability from a genetic point of view. We believe, however, that this report has made it clear that contact with other populations through migration, the larger the better, is much more important than population size per se. The results of the different models here indicates that, even with a population as low as 200 wolves ($N_e = 50$), a continuous inflow of at least one effective migrant per year will reduce inbreeding levels close to $F = 0.05$, and retain more than 95 % of heterozygosity for the nearest 100 years. Inflow ratios of 2-3 wolves per generation (0.4 – 0.6 per year) will retain between 90 and 95 % heterozygosity even at real population levels of 200 – 400 wolves, and will reduce inbreeding levels below $F = 0.15$. Considering that our present wolf population has showed an average annual growth rate above 20 % the last 10 years with an average inbreeding coefficient around 0.3, a level of 0.15 is not likely to threaten the viability of the population.

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Appendix 1. Assignment

Ärendnr NV-08347-12

Naturvårdsverket uppdrar till SKANDULV Grimsö Forskningsstation, SLU, att göra en genetisk komplettering till den demografiska sårbarhetsanalys som SKANDULV utfört och som rapporterades till Naturvårdsverket 2 juli 2012 (Chapron et al. 2012: Demographic Viability of the Scandinavian Wolf Population. *A report to Naturvårdsverket from SKANDULV.*) Kompletteringen ska bygga på en genomgång av senaste publicerade relevanta vetenskaplig litteratur i ämnet, och på redan utförda analyser. Kompletteringen ska belysa krav på populationsstorlek och grad av kontakt med andra populationer för att nå ned till och stabilisera viss given inavelsnivå hos den nuvarande skandinaviska vargpopulationen och för bevarande av olika grad av genetisk variation i samma population. Resultaten ska kunna användas för att fastsätta kraven för att den skandinaviska vargstammen ska uppnå s.k. Favourable Reference Population, i enlighet med det s.k. Art- och Habitatdirektiv. Rapporten ska innehålla en populärvetenskaplig sammanfattning på svenska.