The correlation between inbreeding and fitness: does allele size matter?

Jérôme Goudet and Laurent Keller

The availability of highly polymorphic genetic markers, in particular microsatellites, has made it possible to test the effect of inbreeding on fitness in the field and in the absence of pedigree information. It has been suggested that the squared difference in allele size at a locus (d^2) might be a better indicator of the level of inbreeding than is heterozygosity. Using an elegant new analytical model, Tsitrone *et al.* now put this idea to the test, and to rest.

Published online: 20 February 2002

Considerable debate remains as to whether inbreeding depression is an important selective force under natural conditions. Correlations between inbreeding and fitness have generally been studied with allozyme markers and the level of inbreeding measured as the level of heterozygosity. However, allozymes have now been almost completely replaced by microsatellites, which are much more variable, and a new measure (d^2) has been developed to quantify the level of inbreeding. d^2 is based on the squared difference in the number of repeats for the two microsatellite alleles at a locus within an individual, and is considered to be a useful new tool for exploring the effect of inbreeding (see Keller and Waller, this issue). Accordingly, new studies have used both heterozygosity and d^2 indiscriminately as a measure of inbreeding (reviewed in [1,2]), and have provided divergent results. Clearly, there has been a strong need for a theoretical investigation of the relative merits of heterozygosity and d^2 as a measure of inbreeding, as well as an assessment of the conditions under which each measure should be used. This is just what Tsitrone *et al.* [1] have now done.

The inbreeding-fitness model

Tsitrone *et al.* [1] constructed models to determine how d^2 and heterozygosity are correlated with fitness under different scenarios. They compared situations in which inbreeding had occurred recently in the pedigree [e.g. when mating occurs between close relatives (close inbreeding)] and deep in the pedigree [e.g. when individuals from isolated populations are brought together (deep inbreeding)]. Close inbreeding was modelled as partial selfing, where a given proportion of the population reproduced by self-fertilization, and the other mated randomly. Deep inbreeding was modelled as a two-population admixture model, in which an ancestral population of large size was split into two smaller subpopulations, which evolved independently during several generations before being reunited. In both models, the fitness was assumed to decrease linearly with the level of inbreeding. The distribution of differences in size among pairs of alleles was obtained within and among individuals under different mutation schemes, in particular under the stepwise mutation model, which assumes that new alleles are generated by the addition or subtraction of one repeat motif (the model characteristic of microsatellites). Analytical results were explored numerically to quantify the effects of (a) mutation rates (from 10^{-6} to 10^{-2} per locus per generation); (b) the sizes of the ancestral and descendant subpopulations (from 10^4 to 10^6 and 10^2 to 10^4 individuals, respectively); and (c) the divergence time between the two subpopulations (between 10^4 and 10^6 generations).

The role of genetic markers

The results are striking. First, the simulations showed that the type of inbreeding does matter with regard to which genetic markers are more suitable for examining inbreeding. Under close inbreeding, the correlation between fitness and heterozygosity was always higher for markers with high mutation rates. Thus, microsatellites are better suited to such studies. However, under deep inbreeding, the correlation between fitness and heterozygosity was highest for rates of mutation of the order of the inverse divergence time between subpopulations (1/number of generations). Thus, if divergence time were short, say a few hundred to a few thousand generations, markers with high mutation rates, such as microsatellites, would be most suited. By contrast, if the divergence time were of the order of a million generations, allozymes or even single nucleotide polymorphisms (which have a lower mutation rate) would be better.

The type of inbreeding matters

Second, the authors found that the type of inbreeding was important with regard to what measure of inbreeding was more closely correlated with fitness. Under close inbreeding, fitness is always more closely correlated with heterozygosity than is d^2. This is especially true for markers with high mutation rates. The situation is more complex under deep inbreeding, because two parameters (the mutation rate μ and the subpopulation size N) influence how correlated fitness is with heterozygosity or with d^2. If the product of these two parameters (which governs how many mutations occur per generation in each of the two subpopulations) is <1, heterozygosity is a better indicator of fitness than is d^2. This is because individuals whose parents were from the same subpopulation will be mainly homozygous at all loci, whereas hybrids will be heterozygous at the loci where different alleles were fixed in the two subpopulations before admixture. Thus, heterozygosity provides more information about the origin of the parents of an individual than does d^2, because accounting for differences in allele sizes in heterozygotes is superfluous and adds unnecessary noise to the estimation of inbreeding.

However, if $N\mu$ is high, most individuals, be they hybrids or not, are heterozygous and heterozygosity is therefore a poor predictor of whether individuals are hybrid. This is when d^2 becomes useful, because, under stepwise mutations, individuals of hybrid origin will tend to carry at each locus alleles with larger size differences than will individuals whose parents are from the same subpopulation. Thus, when the product $N\mu$ is >1, d^2 is more closely correlated with fitness than with heterozygosity.

  PII: S0169-5347(02)02467-9

What, then, is $N\mu$ likely to be in natural populations? The mutation rate at microsatellite loci is thought to vary between 10^{-4} and 10^{-3} [3]. Therefore, the size of each of the two subpopulations before admixture occurs needs to be >10 000 individuals for $N\mu$ to be >1. Such subpopulation sizes are likely to be relatively uncommon, especially for large vertebrates. Moreover, populations of such a size are unlikely to fix many deleterious alleles, because those alleles should spread to fixation by genetic drift only in relatively small populations [4]. Therefore, under admixture of large subpopulations (i.e. the restrictive conditions where d^2 might perform well), the fitness differences among individuals whose parents came from the same or different subpopulations should be small, hence restricting the probability of detecting a correlation between fitness and any measure of inbreeding.

There is an additional problem for d^2 in providing a useful method to determine the level of inbreeding, when there is admixture among different populations: differences in the level of inbreeding among individuals are expected to quickly decay once the populations start interbreeding. This is because every individual will rapidly have a mixed ancestry. After five generations of admixture, for instance, less than one individual in a billion will remain of pure parental origin. Therefore, unless the two subpopulations just came into contact, differences in inbreeding among individuals should vanish very swiftly, which will lower the probability of detecting a correlation between fitness and inbreeding.

Is the correlation between d^2 and fitness real?

Overall, it seems that the conditions where fitness should be more closely correlated with d^2 than with heterozygosity are very limited. So why has fitness been found to be more closely correlated with d^2 in so many studies (reviewed in [1,2])? One possible explanation is that important factors other than those considered in Tsitrone *et al.*'s [1] model are involved, although it is unclear what these factors could be. Alternatively, it might be that too much enthusiasm has accompanied the development of d^2 and that a more careful examination of the data will lead to a different conclusion. For example, only one or a few components of fitness were correlated with d^2 in most studies where fitness was better correlated with d^2 than was heterozygosity. Perhaps these few significant results correspond to the 5% false positive expected in statistical testing. Indeed, the significant correlation between d^2 and both birth weight and juvenile survival reported previously in the deer population on the isle of Rhum [5] disappeared when a larger number of loci was used (71 instead of nine; [2]). Conversely, heterozygosity, which was not significantly correlated with birth weight in the original study, became significantly correlated in the new study. Moreover, Slate and Pemberton [2] show that individual heterozygosity correlates, albeit weakly, across loci, whereas d^2 does not, leading them to conclude that heterozygosity is a more robust measure of inbreeding than is d^2.

The work of Tsitrone *et al.* [1] is therefore important for several reasons. First, it indicates that one should not use heterozygosity and d^2 indiscriminately as measures of inbreeding. If inbreeding is expected to have occurred only recently, then heterozygosity should always be preferred to d^2. Second, it shows that the conditions under which d^2 performs better than does heterozygosity are quite restricted. Third, their work provides hope that it will soon be possible to determine whether inbreeding occurred deep in the pedigree or more recently (close inbreeding). This will probably require developing new measures of inbreeding and modelling the effect of long- and short-term inbreeding on these measures, but the prospect that this will be done is good. Indeed, new measures of inbreeding accounting for the level of variability at each marker locus have been developed recently [6,7] and Tsitrone and colleagues are currently generalizing their modelling approach to multiple loci.

References

1 Tsitrone, A. *et al.* (2001) Heterosis, marker mutational processes and population inbreeding history. *Genetics* 159, 1845–1859
2 Slate, J. and Pemberton, J.M. (2002) Comparing molecular measures for detecting inbreeding depression. *J. Evol. Biol.* 15, 20–31
3 Weber, J.L. and Wong, J.C. (1993) Mutation of human short tandem repeats. *Hum. Mol. Genet.* 2, 1123–1128
4 Lynch, M. *et al.* (1995) Mutation accumulation and the extinction of small populations. *Am. Nat.* 146, 489–518
5 Coulson, T.N. *et al.* (1998) Microsatellites measure inbreeding depression and heterosis in red deer. *Proc. R. Soc. London B Biol. Sci.* 265, 489–495
6 Coltman, D.W. *et al.* (1999) Parasite-mediated selection against inbred Soay sheep in a free-living, island population. *Evolution* 53, 1259–1267
7 Amos, W. *et al.* (2001) The influence of parental relatedness on reproductive success. *Proc. R. Soc. London B Biol. Sci.* 268, 2021–2027

Jérôme Goudet*
Laurent Keller
Institute of Ecology, Biology Building, Lausanne University, CH-1015 Lausanne, Switzerland.
*e-mail: jerome.goudet@ie-zea.unil.ch

Meeting Report

Impacts and extent of biotic invasions in terrestrial ecosystems

Carla M. D'Antonio and Salit Kark

The workshop on Impacts and Extent of Biotic Invasions in Terrestrial Ecosystems was held in Barcelona, Spain, from 19 to 22 September 2001.

Published online: 18 February 2002

Invasive species have become a priority concern for conservation biologists and land managers. For basic research scientists, they are a tool with which to examine controls over community structure and ecosystem processes. However, the field of 'invasion biology' has been almost as resistant to generalization as the broader field of ecology. As a step toward greater synthesis, the Impacts and Extent of Biotic Invasions in Terrestrial

  PII: S0169-5347(02)02454-0