Conservation Genetics Inbreeding, Fluctuating Asymmetry, and Captive Breeding Exercise

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OVERVIEW

Conservation genetics has many applications in conservation biology. It is central to successful captive breeding and subsequent re-introduction of threatened species. Many species have been saved from extinction by captive breeding in zoos, botanical gardens and aguaria, which provide a safety net when other protective measures have failed. Yet because of costs and space limitations, bringing a species into captivity often means having to manage small numbers of individuals. Under these conditions inbreeding and genetic drift can become important considerations. Captive management thus involves careful analysis of breeding programs, construction of pedigrees, and monitoring of the effects of inbreeding. One external manifestation of inbreeding is sometimes a change in body symmetry, which can be evaluated from measurement of morphological features of the individuals in captivity. This exercise focuses on many of these issues and seeks to integrate concepts in conservation genetics using captive breeding programs as the context to do so. The objectives of the exercise are four-fold: (1) to become familiar with the concept of inbreeding, (2) to learn how to construct and analyze pedigrees, and (3) to gain experience in morphological analysis for analyzing the effects of inbreeding, and (4) gain experience in making expert recommendations about captive breeding plans.

PEDIGREE ANALYSIS

Recall that the inbreeding coefficient is the probability that an individual has a pair of alleles that are identical by descent from a common ancestor, that is, they are autozygous. By extension the inbreeding coefficient is also the proportion of loci that carry alleles that are identical by descent from a common ancestor. We are interested in estimating levels of inbreeding for particular individuals for several reasons. One is that it can tells us whether individuals are at risk of disease and disorders associated with lack of genetic variation. Another is that levels of inbreeding indicate the extent to which an individual has a unique versus shared component of genetic diversity relative to other individuals in the population. This unique component is something we might want to capture or exclude in the course of designing future breeding programs.

The first step in estimating levels of inbreeding is to draw a pedigree that describes the relationships among all individuals (live and dead) in a population.

Pedigrees systematically summarize the breeding relationships among a group of individuals. In a pedigree, females are indicated as circles, males by squares. Offspring are indicated by lines joining their parents. Here is an example of half first cousins:



Now let's construct a pedigree for an actual population of organisms. Consider the following summary of the breeding history within a hypothetical captive population of 13 triple nose-leaf bats (*Triaenops persicus*, Chiroptera, Hipposideridae). The species occurs along the coastal regions of East Africa and is threatened by disturbance of the caves in which it breeds, particularly by disturbance from tourists to the coral caves the bats inhabit. As you can see from an illustration below (from Blanford 1888) of the 11 living individuals in the colony, the species is aptly named.



The captive colony was founded in 1990 by 5 bats, a male named Bright that died in 1993, a male named Blue that died in 1995, and 2 females still alive – Fluffy and Skinny, and another male, Tiny, that also is still alive. All of their offspring remain alive. Bright and Fluffy have produced a male named Lucky that mated with Skinny to produce a female named Sneaky. Fluffy also mated with

Blue to produce a female Star that has subsequently mated with Tiny to produce a male named Shy. Of the captive born offspring, Lucky and Star have mated to produce a male named Timid, and Sneaky and Shy have mated to produce three daughters: Sleepy, Fuzzy, and Triumphant.

(1) Construct a pedigree that captures relationships among individuals in this breeding population, using the appropriate symbols as well as indicating which individuals are alive versus dead.

CALCULATING LEVELS OF INBREEDING

The next step is to analyze the pedigree to determine the levels of inbreeding of each individual. For any given individual, we must first identify all common ancestors in the pedigree, because an allele can only be autozygous in an individual if it was inherited through both of the individual's parents from a common ancestor. Note that in many cases individuals will not have common ancestors (or at least any known common ancestors) and thus inbreeding will not be an issue for them. But for those that do, the third step is to trace all paths of gametes that lead from one of an individual's parents back to the common ancestor and then back down again to the other parent. Focusing on individual I in the pedigree above, there is only one such path – DBACE, with A being the common ancestor:



The fourth step is to calculate the probability of autozygosity for the individuals with common ancestors. Because at each step the likelihood of passing any particular allele is 1/2, the probability of the same allele being reunited in a particular offspring is $(1/2)^{i}$, where *i* is the number of individuals in the path through the common ancestor. Last, the common ancestor may already be inbred at a level of F_A. If this is the case, we add a correction factor to account

for this inbred common ancestor. Thus, that the probability of autozygosity of an individual, F_1 , is:

$$F_1 = (1/2)^i (1 + F_A)$$

(2) How inbred is each living individual in the captive colony of threenose leaf bats for which you drew the pedigree? Note that for this captive colony, all founders are/were non-inbred, except for Fluffy, that was highly inbred at F = 0.125. Use your pedigree to make estimates for each individual.

(3) Last, if in this species a level of $F_1 > 0.1$ is known to predispose these animals to genetic disease, which individuals are at risk?

EFFECTS OF INBREEDING ON FITNESS

Fluctuating asymmetry (FA) is deviation from perfect bilateral symmetry. It is thought that the more perfectly symmetrical an organism is, the better it has been able to handle stresses during its development because it possess more allele variants that can buffer its development processes. Thus, individuals experiencing high levels of inbreeding often experience high levels of autozygosity and exhibit high FA. Low FA is often associated with higher mating success because FA is frequently assessed by potential mates during courtship (including us humans!). This may occur because an individual's degree of symmetry may be an "honest" signal of one's underlying fitness. FA is even tied to population status; for example, a recently study (Lens et al. 2002) indicated that birds in more disturbed habitats had higher FA than those in less disturbed habitats – this is a good article to consult for more background on application of FA to conservation problems. The point is that levels of fluctuating asymmetry may thus be a useful measure of overall fitness.

FA can be fairly straightforward to measure. For a full treatment of the matter, see (Palmer 1984, also presented at:

http://www2.biology.ualberta.ca/palmer/asym/asymmetry.htm).

The key concept is that we need to measure consistent parameters on the left and right side of an organism relative to the organism's midplane. In the very simplest approach, the absolute value of the left-side measurement (L) minus the right side measurement (R) is an often adequate index of asymmetry. If the organism was perfectly symmetrical, then |L - R| = 0. If either the left or right was greater than the other, then |L-R| would be > 0. Thus, the index starts at zero and increases positively as the individual becomes more asymmetrical.

(4) What is your prediction about the relationship between levels of **FA and F**_i? State your prediction in writing and provide a justification for it.

Let's turn back to the three-nosed left bats.

(5) Calculate an index of FA for each bat.

Note that you first must establish the midplane for each individual. Do this carefully with a ruler, drawing a faint line straight through the center of each individual's face. Make sure each midplane is determined as PRECISELY as possible – all subsequent measurements are relative to it! Next measure on each side of the individual the same corresponding parameter. You will need a ruler with units of millimeters. We suggest measuring the distance from the center of the eye to the center of the nose for starters. But you are free to use another metric. You might measure several metrics and compare results among them – they should yield the same conclusions.

(6) Now compare the *mean* FA for inbred versus noninbred individuals. Are the estimates the same?

RECOMMENDATIONS FOR A FUTURE BREEDING PROGRAM

Now that you have established the pedigree and learned something about levels of inbreeding in this population, let's turn the focus to broader issues.

(7) If you were to make recommendations for the breeding program in the future to capture as much genetic variation as possible that remains in this small population, which animals would you suggest to include in future breeding efforts?

Look over the pedigree carefully. Genes of which individuals are already well represented in the population? Genes of which individuals are relatively poorly represented? Consider even the dead individuals and ask in which living individuals are their genes best represented. *What individuals would you preferentially involve in future breeding programs and which would you exclude? Remember that you want to keep inbreeding levels to a minimum in whatever recommendations you make.*

LITERATURE CITED

- Blanford, W.S. 1888. The Fauna of British India, including Ceylon and Burma Mammalia. Taylor & Francis, London, UK.
- Lens, L. Stefan Van Dongen, and Erik Matthysen. 2002. Fluctuating Asymmetry as an Early Warning System in the Critically Endangered Taita Thrush. Conservation Biology 16:479-487.
- Palmer, A.R. 1994. Fluctuating asymmetry analyses: A primer. Pages 335-364 in T. Markow, editor. Developmental Instability: Its Origins and Evolutionary Implications. Kluwer, Dordrecht. Available online at http://www2.biology.ualberta.ca/palmer/asym/asymmetry.htm.