Demography, Genetics, or Statistics: Comments on a Paper by Heschel and Paige

N. JOOP OUBORG AND JAN M. VAN GROENENDAEL

Department of Ecology, University of Nijmegen, Toernooiveld 1, 6525 ED NIJMEGEN, The Netherlands, email joopo@sci.kun.nl

Is it demography or is it genetics? Seven years after the publication of a provocative paper by Lande (1988), this question is still central to the discussion of what imposes the biggest threat to small, isolated populations. Over this period many have concentrated on measuring genetic variation in small populations, but only a few studies have tried to assess the consequences of small population size or low genetic diversity in terms of fitness components. We are therefore in great need of studies on the relationship between population size and components of fitness. The few studies that provide data on this subject are becoming classics (e.g., Jennersten 1988; Menges 1991), and any new paper providing such data would likely follow the same fate.

Recently, Heschel and Paige (1995) published a study on the effects of variation in population size on fitness components in natural populations of the scarlet gilia (*Ipomopsis aggregata*). They purportedly demonstrated that small, isolated populations of this species have reduced seed size, reduced germination success, and enhanced susceptibility to environmental stress. By performing crossing experiments, moreover, they demonstrated that the reduced fitness in the small populations had genetic causes.

But anomalies can be found in their results. They first show that small populations (≤ 100 individuals) have reduced seed size and present an analysis-of-variance (ANOVA) table with significant F values for the effect of population size on seed size. Their calculated F values are wrong, however. They tested all their mean squares against the residual MS; but because this is a nested analysis, mean squares of one level should be tested against mean squares of the next level. When calculated in the proper way (based on the sum of squares presented in their Table 1) the F values for small versus large are $F_{1991} = 0.87$ and $F_{1992} = 0.70$, both of which are far from significant.

They further present t tests for the differences in average seed size and germination success between small and large populations. Apart from the questions of why seed size is tested in a t test when it was already tested in an ANOVA, and why germination success was not tested in an ANOVA, more oddities appear. Although Heschel and Paige presented degrees of freedom only for the germination test, it is clear that they pooled all the data in both tests. Their own Table 1 gave significant differences among populations, in which case pooling is not allowed. In this case pooling inflated the degrees of freedom, giving rise to very strong significance where in fact there is no effect (as our recalculated F values show). The same mistake was made in the test of the effects of stress, in which again all data in the population size categories were pooled without prior testing for differences within groups.

The next problem is with the pollination treatment. When small populations have reduced fitness for genetic reasons (loss of alleles or high inbreeding level), importing pollen from a distant population should lead to an increase in fitness in the small populations, but not in or not as much as in the large populations. In fact, that is what Fig. 3 shows: increased seed size in the small populations after hand pollination with pollen from a distant population, but not in the large population. But the interpretation that this result implies genetic causes of reduced fitness in the small populations is based solely on the absence of a pollination effect in one large population in one year and on one fitness component. In the few studies that related population size to the effect of between-population crossing on progeny fitness, it was shown that the pollination effect is variable in each group, small and large (Van Treuren et al. 1993; Ouborg & Van Treuren 1994). In particular, there were populations in each group that responded to the pollination treatment with a large increase in fitness, and there were also populations that did not respond. Given this large variation among populations, the evidence presented by Heschel and Paige can hardly be convincing.

The last point of concern regards the germination results presented in Fig. 4. There is a well known relationship between seed size and germinability of seeds (Rees 1993). Finding a better germination for larger seeds restates the previous findings presented in Fig. 3. In contrast to this figure, no further distinction is made between the two small populations, notwithstanding the large differences in seed size across treatments between these two populations. To assure an independent effect on germination of pollen source, seed size has to be factored out in the analysis.

Apart from this (again) technical point, we are left in doubt as to the fate of the ungerminated seeds. Assuming that at least part of the seeds are dormant, an assumption based on the short-lived, monocarpic life history of the species, such dormancy could constitute a benefit to a small population because it buffers the population against decline and allows the species to await better times, as shown by a number of authors (Venable & Brown 1988; Van Groenendael et al. 1994). This casts serious doubts on the claim by the authors that distant pollen confers a fitness increase in these small populations, because it might in fact hasten the decline by untimely germination of seeds.

It is our impression that the paper is statistically flawed and methodologically fuzzy. We think that assessing the relationship between population size, genetic diversity, and fitness lies at the very heart of conservation biology. It is important that we sample as much data as we can and try to interpret them with respect to the

question. Performing experiments with rare species is a difficult and contentious task. The data we collect this way deserve the best analytical tools we have and careful interpretation of the results.

Literature Cited

Heschel, M. S., and K. N. Paige. 1995. Inbreeding depression, environmental stress, and population size variation in Scarlet Gilia (*Ipomopsis aggregata*). Conservation Biology 9:126-133.

Jennersten, O. 1988. Pollination in *Dianthus deltoides* (Caryophyllaceae): Effects of habitat fragmentation on visitation and seed set. Conservation Biology 2:359-366.

Lande, R. 1988. Genetics and demography in biological conservation. Science 267:226-228.

Menges, E. S. 1991. Seed germination percentage increases with population size in a fragmented prairie species. Conservation Biology 5: 158-164.

Ouborg, N. J., and R. Van Treuren. 1994. The significance of genetic erosion in the process of extinction. IV. Inbreeding load and heterosis in relation to population size in the mint *Salvia pratensis*. Evolution 48:996-1008.

Rees, M. 1993. Trade-offs among dispersal strategies British plants. Nature 366:150-152.

Van Groenendael, J., H. de Kroon, S. Kalisz, and S. Tuljapurkar. 1994. Loop analysis: Evaluating life history pathways in population projection matrices. Ecology 75:2410-2415.

Van Treuren, R., R. Bijlsma, N. J. Ouborg, and W. van Delden. 1993. The significance of genetic erosion in the process of extinction. III. Inbreeding depression and heterosis effects caused by selfing and outcrossing in *Scabiosa columbaria*. Evolution 47:1669-1680.

Venable, D. L., and S. J. Brown. 1988. The selective interactions of dispersal, dormancy, and seed size for reducing risks in variable environments. American Naturalist 131:360-384.