

Action on amphibians

The first World Congress of Herpetology (University of Kent, Canterbury, UK, 1989) brought together field biologists who swapped stories about mysterious disappearances of various species of frogs. Sufficient concern was expressed to cause the National Research Council (USA) to convene a special workshop (February, 1990) to explore the matter¹. The organization of a special task force on declining amphibian populations (DAPTF), allied with the Species Survival Commission (IUCN, Gland, Switzerland), was a direct outcome of this workshop. Throughout this decade, substantial research has been conducted on amphibian populations and, although many questions remain and many species and several areas of the world appear unaffected, there have been extensive species declines and extinctions (10–14 recent species extinctions can be documented, and evidence of additional extinctions exists).

At the third World Congress of Herpetology (Prague, Czech Republic, 1997), a symposium highlighted new evidence of declines and hypotheses for their causes. The need for an in-depth analysis of the problem and development of a general research strategy was expressed by many, and in the months since the congress new developments have lent a sense of urgency. For example, Bruce Babbitt (Secretary of the Interior, USA) has shown a personal interest in the matter, requesting several briefings. At the most recent (May, 1998), two other USA presidential cabinet officers, Donna Shalala (Health and Human Services) and Carole Browner (the Environmental Protection Agency) were present, in addition to Neal Lane [the Director of the National Science Foundation (NSF), now the Presidential Science Advisor] and Harold Varmus (the Director of the National Institutes of Health).

Why the sudden attention, after nearly a decade of concern? The explanation is complicated, but a combination of dramatic loss of populations in the great national parks of western USA (Ref. 2), the discovery of numerous instances of seriously deformed frogs (mainly reported in the popular news media), and evidence of new, virulent diseases that affect amphibians in lower Central America, Australia³ and Arizona⁴ are the main factors.

What is to be done?

Because of widespread concern, a workshop sponsored by the NSF entitled 'Amphibian population dynamics – is the threat of extinction increasing for amphibians?' (NSF headquarters, Arlington, VA, USA;

28–29 May 1998) attracted attention from scientists around the world, from USA governmental officials and from media representatives. The workshop, led by James Collins and coorganized by Andrew Storfer and Elizabeth Davidson (Arizona State University, Tempe, AZ, USA), was attended by more than 70 individuals, ranging from field naturalists and population biologists to toxicologists, pathologists, virologists and developmental biologists.

The first day of the workshop was devoted to analytical reports on the status of research on amphibian declines. Collins outlined the two aims: (1) to determine if the threat of extinctions is increasing, and (2) to recommend research and management actions in response to recent evidence of changing amphibian population dynamics. The work of DAPTF was summarized by Tim Halliday (International Director, Open University, Milton Keynes, UK), and geographically based presentations (of South America, Central America, Australia, western USA, eastern USA, Europe and Canada) focused the proceedings. The afternoon featured reports on emerging issues (UV radiation, toxicology, malformations and deformities, disease, global warming and immunology). Mary Clutter (Assistant Director for Biological Sciences, NSF), Neal Lane, and Bruce Babbitt joined the group later, and Bruce Babbitt pledged his support for developing a coordinated approach to the problems.

Agreement was obtained on the fundamental points:

- The phenomenon is real – there has been documentation of species loss and/or decline in Cordilleran America from the Rocky Mountains and Cascades through the Sierra Nevada to Costa Rica and Panama, and the high paramos of Venezuela and Ecuador. Results from Australia are also convincing.
- Not all species in a given region are similarly affected. Some regions, especially upland streams in the tropics of Central America and Australia, are hard hit, whereas other regions report little or no discernible effects.
- There are so many potential causative agents that the focus of investigation has shifted from these to factors leading to widespread, generalized stress.
- Basic issues in population biology are central to an understanding of species loss and decline.
- Lack of information on work in progress and problems of accessing data are increasingly becoming barriers to effective communication among researchers.
- More coordination of activity is required.

The results of the first day galvanized action on the second. A focused discussion of issues and needs set the stage for specific proposals for action. The community of amphibian biologists is small, and there is a mismatch between distribution of resources, both human and material, and that of amphibian species and reported problem areas. For example, there are many amphibian specialists in Canada, where there are no endemic species, whereas Mexico, with enormous amphibian diversity and high endemism, has very few amphibian specialists. Information on declines and disappearances is best for nature reserves in Australia, Costa Rica, Panama and the USA, but little is known about less studied sites, especially in countries with high amphibian diversity, such as Colombia and Indonesia. Issues that field biologists do not normally face mean that investigators representing other approaches will have to be recruited. A sense of urgency exists concerning new fungal and viral diseases, and the increased impact of algal and bacterial infections. These issues were discussed by virologists and pathologists attending the workshop, but researchers in these disciplines are rarely involved in field work. Several participants emphasized the role of generalized stress. Environmental stress factors remain poorly understood. We need to understand natural versus anthropogenic stressors, and the impact of stressors on amphibian immune systems and physiology. In general, more field workers are required, especially in tropical areas.

Proposals for action

The participants developed the following specific proposals.

(1) Development of a new distributed institute: this came to be termed the 'Cordilleran Initiative'. The institute would link agency and independent research units in western North America with workers focusing attention on Costa Rica and Panama, although Mexican, Guatemalan, Honduran and other appropriate groups would also be included. The institute would also sponsor collaborative interactions, such as internships for training professionals, graduate students and post-doctoral fellows, as well as providing opportunities for regular communication via workshops and the Internet. Data management systems and communication systems are also a necessary component of such an institute. The approach would be multidisciplinary and would involve the development of rapid response teams.

(2) A series of specialized workshops:

- Amphibian diseases and immunity – there is a role for organizations such as the Centers for Disease Control in Atlanta, which should be invited to participate

(this workshop was immediately funded by NSF and will take place in San Diego on 26–28 July 1998).

- Population and community ecology and monitoring – focused critical thinking is needed to deal with statistical issues in assessing population dynamics.

- Ecotoxicology – airborne contaminants are implicated in amphibian declines, and existing test protocols might be inappropriate.

- Developmental biology – malformations in amphibians demand investigation because normal developmental processes are being disrupted.

(3) Bioinformatics: the community of concerned individuals requires an effective, web-based communication and information system to which each individual can turn for authoritative, current information. Promising prototypes exist that might serve as effective models. Initiatives integrating the systematic biology and ecological communities should be encouraged. Such a system would be most effective if it were based on systematic authority files for taxa, on specimen-based information for historical documentation, and on georeferenced locality data (using geographical information system technology) for mapping information on normal distributions and declines. Alliances with diverse

agencies should be established to take advantage of information gathered on weather, climate and related phenomena.

The participants in this productive workshop concluded by adopting a resolution affirming that the empirical evidence for amphibian declines and disappearances is strong, that the existence of unaffected species and areas offers opportunities to conduct appropriate investigations of the phenomenon, and urging adoption of the broad-based approach to the study of amphibian population dynamics outlined here, in the expectation that such an approach might serve as a model for the study of the global biodiversity crisis.

Immediate action is being taken to set up an interagency Task Force on Amphibian Declines and Deformities (chaired by William Y. Brown, science advisor to Bruce Babbitt) in the USA, which will be important in the establishment of an interdisciplinary and collaborative research program. Tasks include quantification of the direct and indirect factors affecting amphibian population dynamics, starting with the study of patterns of change through the use of historical records, field-based correlative data, and controlled, multifactorial experiments. Organization of interdisciplinary incident response teams was

also recommended. These teams could be assembled in 'hot spots' of amphibian decline to identify factors to mitigate these declines. Finally, the workshop participants called upon both public and private agencies and institutions for the funding and policy support required for future activities.

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Mother's boy or daddy's girl? Sex determination in Hymenoptera

Sex-determination in hymenopteran insects is superficially simple: fertilized eggs develop as diploid females; unfertilized eggs develop as haploid males. But this apparent simplicity hides a more difficult question. How can two copies of a genome cause an embryo to develop as a female, whereas one copy of the same genome causes an embryo to develop as a male, when no gene that is present in one sex is absent in the other, and when all ratios among sex-determining genes are unchanged? Whiting proposed an answer to this conundrum based on his discovery of diploid males among inbred progenies of the parasitoid *Bracon hebetor*¹. In his model of complementary sex determination (CSD), heterozygotes at a multi-allelic sex-determining locus develop as females, whereas hemizygotes and homozygotes develop as males.

Diploid males have now been reported in more than 30 hymenopteran species – including sawflies, bees, ants, braconid and

ichneumonid wasps – and CSD is believed to be the mechanism of sex determination in these species^{2,3}. However, CSD cannot explain the difference between the sexes in species that do not produce diploid males, even after prolonged inbreeding. One such species is the chalcidoid wasp *Nasonia vitripennis*. A recent paper now provides evidence that femaleness in *N. vitripennis* is determined by the presence of a paternal genome, and maleness by its absence⁴.

Dobson and Tanouye⁴ tested alternative theories of sex determination in *N. vitripennis* by mating triploid females to haploid males, some of which carried the paternal sex-ratio chromosome (PSR). Triploid females came from a polyploid strain of *N. vitripennis* that had been maintained by repeated matings of diploid males to triploid females (diploid males produce diploid sperm; triploid females lay abundant eggs, most of which are aneuploid, but a few have haploid or diploid chromosome complements and produce viable offspring).

In crosses using this strain, unfertilized eggs (whether haploid or diploid) produce males, whereas fertilized eggs produce diploid, triploid or tetraploid females⁵. The PSR chromosome is a B chromosome, transmitted via sperm, that causes the elimination of all paternal chromosomes (except itself) from early embryos^{6,7}.

The results of Dobson and Tanouye's crosses (Box 1) were incompatible with most published hypotheses about the mechanism of haplodiploid sex determination. Single-locus and multilocus CSD models could be rejected because unfertilized, heterozygous diploid eggs developed as males. The act of fertilization itself could not be female-determining (as proposed by Whiting⁵) because eggs fertilized by PSR-bearing sperm developed as males. Finally, 'maternal-effect' and 'genic-balance' models could be rejected because these models predict a one-to-one correspondence between sex and ploidy. The only model that was not contradicted by the data was Beukeboom's hypothesis that one or more loci are differentially modified (imprinted) in male and female germ lines, such that a paternal genome is necessary for female development^{8,9}. Imprints must be reversible in this model because a paternal allele in one generation is a maternal allele in the next.