

GENETICS AND DEMOGRAPHY IN THE CONSERVATION OF BIODIVERSITY

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ABSTRACT

Half of Thailand's remaining species of plants, animals and microorganisms are at risk of extinction in the next few decades. The role of genetic and demographic considerations in determining the future ability of individual populations to survive and evolve are described. Population viability analysis (PVA) points to the need to maintain as much innate genetic variability as possible. The concepts of a minimum viable population (MVP) and of the genetically effective population size (N_e) are described. The serious consequences of increased inbreeding and the loss of genetic variation following range fragmentation are discussed. Genetics is also very important in defining evolutionarily significant units (especially species) for management purpose. These considerations underscore the urgent need to complete the inventory of Thailand's biodiversity and to gather the types of genetic and demographic data required to ensure its persistence. This will necessitate more scientifically based intervention than has been required in the past. In addition, further shifts in national natural resource policy towards sustainability and the enhancement of educational, training and research opportunities are foreseen.

INTRODUCTION

The biodiversity crisis is truly international in its occurrence and consequences.^{1, 2} The situation in Thailand is similar to that prevailing in many other tropical countries. Much of what remains of Thailand's remarkable flora and fauna is threatened with extinction in the next few decades. Forest and wetland destruction, proceeding at a rate of more than 2% per year, will eliminate or degrade most wildlife habitats. The surviving populations and communities will be small and fragmented and their conservation will require far more management than was necessary in the past. There is thus an urgent need to develop appropriate management guidelines based on sound scientific principles.

Unfortunately, the Thai biota is still rather poorly known, making it difficult to set conservation priorities. Similarly, the development of procedures and technologies for population viability assessment is still in its infancy. We therefore face the double challenge of completing the national biodiversity inventory matrix and, at the same time, solving some most difficult scientific problems relevant to biodiversity management. As the societal costs of failing to

conserve biodiversity are increasingly clear, it is appropriate that a larger fraction of the national scientific enterprise be devoted to this area. In this paper, I will review the genetic and ecological underpinnings of biodiversity conservation. For personal reasons, I shall focus more on the application of genetics to the problems of maintaining population viability and to the identification of evolutionarily significant conspecific units for management.

Population Viability Analysis

The equilibrium theory of island biogeography clearly predicts the magnitude of the biodiversity crisis.³ The positive linear relationship between the logarithms of species number and habitat area suggests that if only 2% of the earth is protected for wildlife, then only 10% of the biota will survive. In Thailand, where human activities have already destroyed or degraded 75% of the habitat, present trends lead to the prediction that 90% of the original plant and animal species have already disappeared or are at high risk of extinction in the next 100 years.⁴

The causes of extinction (complete disappearance) or extirpation (local disappearance) of species are well known.⁵ The four major causes, the "evil quartet," are habitat loss and fragmentation, demographic overkill, introduced species, and secondary effects.⁶ The last are well illustrated by the cascade of extinctions that may be precipitated by the removal of keystone species from a community.⁷ In addition, we must also consider a fifth potential cause of extinction, namely, the loss of a population's ability to adapt to environmental change. This will become increasingly significant as the ongoing anthropogenic global climatic changes proceed.

Although the equilibrium theory of species diversity leads to predictions about the magnitude of species losses, it tells us very little about the extinction probabilities of individual species. This important issue is addressed by population viability analysis (PVA), the goal of which is to identify and quantify all the factors that can lead a population to extinction.⁸ PVA is thus multivariate and involves both reductionist and synthetic techniques. It goes well beyond the single factor (census count) minimum viable population (MVP) approach that was in vogue during the early 1980s.^{9, 10} PVA also goes beyond the traditional wildlife ecology approach to species viability by considering both genetics and the dynamic extinction and colonization interactions over a fragmented habitat.¹¹ Regarding a species as comprising a metapopulation—a system of local units, each of which has its own stochastic dynamics—rather than as a single large population, has very significant consequences for managers. The persistence of populations on individual habitat patches and the between-patch colonization rates will, in part, determine the persistence of the metapopulation as a whole. Within each patch, population persistence is affected by both environmentally dependent factors and environment-independent factors such as genetic drift, inbreeding, and the loss of genetic variability. PVA thus attempts to quantify overall species persistence from a consideration of patch-turnover dynamics. As this approach is relatively new, it should not surprise us that there are still very few examples, although the Concho water snake, the black-footed ferret and the northern spotted owl have received preliminary attention.¹²⁻¹⁴ I use the word preliminary, as at the outset, it is assumed

that PVA must be interactive, i.e. initial analyses and the recommendations that emerge from them must be modified as management policies are implemented and as environmental factors change. PVA therefore does not produce the type of simple answers generated by the MVP approach. MVP sizes, it may be recalled, were defined in terms of the smallest effective population size, N_e , with an x% probability of persisting years.¹⁵ Early MVP analyses led to the conclusion that short- and long-term viability required an N_e of 50 and 500, respectively.¹⁶ As discussed below, it is now recognized that there are no such "magic numbers" and that, instead, we should be seeking to define more general thresholds for population survival and evolution.

Genetic Variation

The loss of genetic variability caused by reduced population size and by range fragmentation has important consequences for biodiversity conservation. As a generalization, genetic variation is a highly desirable characteristic.¹⁷⁻¹⁸ There are numerous cases where individual variation is positively correlated with evolutionary fitness. Developmental stability, growth rate, adult size, metabolic efficiency, fertility, survival and disease resistance are among those fitness traits shown to be positively related to genetic variability.^{19, 20} Although the genetic mechanisms underlying these relationships are still not well understood, the consequences of losing genetic variation are so harmful that we should make every effort to maintain innate or existing levels of variation in natural and managed populations.

Genetic variation can be monitored directly and indirectly in a number of ways. Studies of allozymic variation have been the most commonly employed approach during the last 20 years. In this technique, the allelic variants of soluble enzymes and other proteins that can be visualized biochemically on a gel after electrophoresis are counted directly. Studies of a few thousand plants and animals show that the majority of sexually reproducing organisms are moderately to highly variable.²¹ The proportion of loci in a population that are polymorphic, P , is typically $P = 0.30$. The mean individual heterozygosity, H , (the proportion of loci in an individual that is heterozygous) is typically $H = 0.07$. Electrophoretic surveys have made major contributions to our understanding of natural levels of genetic variation, population structure, and geographic differentiation.

More recently, we have seen the introduction of several molecular genetic approaches to monitoring genetic variation, including mitochondrial DNA (mtDNA) restriction fragment length polymorphism (RFLP) analyses, whole genomic DNA-fingerprinting, and direct sequencing of mtDNA and nuclear DNA loci. Such techniques facilitate very fine detail analysis of microevolutionary processes, but for economic reasons, allozyme electrophoresis remains the preferred technique for most biodiversity conservation problems.

Alternate and indirect approaches to assessing the genetic variability include the study of quantitative variation of traits and the asymmetry of bilaterally paired features. Recent advances in quantitative population genetic theory may lead to the development of new methods of monitoring variability inexpensively in some species.

Genetic Consequences or Range Fragmentation

The fragmentation of the range of a species leads directly to two harmful genetic consequences. Although wildlife managers have until now been able to ignore these genetic effects, in the future, as the size of individual populations and habitat patches are further reduced, they will become dominant concerns. The two genetic consequences, due to inbreeding and genetic drift, may now be considered in turn.

Inbreeding is the mating of close relatives. Father-daughter and full-sibling matings, common in mismanaged zoo populations, have the same potential for failure as do such matings in the human population. Although there are numerous groups including some molluscs and plants which regularly practice self-fertilization (the ultimate inbreeding), the majority of species of concern to biodiversity managers are outcrossing, i.e. they avoid inbreeding. It is well documented that outcrossing populations that suddenly decline in numbers (N) usually experience reduced viability and fecundity known as inbreeding depression.²² Inbreeding produces increased homozygosity of recessive deleterious mutants and, by chance, in small populations these alleles become fixed.

Gradual inbreeding or slow reduction in N allows natural selection to purge deleterious recessive alleles as they become homozygous. Such normally outbreeding populations and those of species that normally reproduce by self-fertilization generally suffer little inbreeding depression. However, the heterosis or hybrid vigor seen when inbred lines are crossed shows that they still carry many slightly deleterious alleles.²²

Inbreeding is a demonstrable genetic problem in many captive populations. In captive mammals, juvenile mortality averages 33% higher in offspring of parent-offspring or full-sibling matings than in the offspring of unrelated parents. With hundreds of years of experience, animal breeders have learned to try to keep the rate of inbreeding at $< 2\%$.²³ Increasingly, the management of wildlife in small fragmented populations must involve the mitigation of harmful effects of inbreeding.

The second negative genetic consequence of range fragmentation is due to genetic drift. Genetic drift involves the random or chance loss of uncommon alleles in small populations. In small populations, such random fluctuations in allele frequencies (genetic drift) reduce genetic variation with time. This stochastic process leads to increased homozygosity and, as a consequence, a reduction in adaptability. If the maintenance of a population's ability to evolve in response to environmental changes is important, then biodiversity managers must pay increasing attention to genetic drift.

The long-term hazards of the loss of genetic variability are well-illustrated by the observed decimation of mammal populations with very low levels of variability when they are exposed to novel virus pathogens. Recent examples include cheetahs and feline infectious peritonitis, black-footed ferrets and canine distemper, and koalas and a still-to-be described retrovirus.²⁴

In general, to maintain population fitness and evolutionary potential, the largest possible populations should be maintained. Population genetic theory has advanced to the point that we can begin to answer the question: how large is large enough? Sewall Wright's

genetic concept of effective population size provides an entrée to this problem.²⁵ The genetically effective size, N_e of a population is typically far less than the actual census count, N . Numerous factors representing departures from the theoretical behavior of a genetically ideal population reduce N_e below N . These include the presence of non-reproducing individuals, unequal numbers of males and females, increased variance in family size, temporal fluctuations in N , and fluctuations in the degree of inbreeding or outcrossing. The cumulative effects of these real world factors are multiplicative and the result is that N_e will often be much less than N . The genetically effective size, N_e , of a population is therefore an estimate of the size of an ideal or theoretical population that would experience genetic drift at the same rate as the actual real world population.

The concept of N_e is very important, as it allows us to estimate the genetic effects of range fragmentation due to genetic drift.²² In the absence of factors promoting genetic variation (mutation, gene flow or immigration), the expected rate of loss of allelic heterozygosity is $1/(2N_e)$ per generation in sexually reproducing species. The predicted rate of loss of genetic variance in quantitative characters is the same. In theory, little variation is lost in any one generation, but small N sustained for several generations can severely deplete variability. Most of the original genetic variability in a population is lost within $2N_e$ generations. Thus, if a wildlife manager is working with a species with a population size of a few hundred individuals and a genetically effective population size of a few tens of individuals maintaining the viability of that species for 100 years will be difficult.

About ten years ago, two influential papers were published relating N_e to population viability.^{15, 26} A consideration of three quantitative (polygenic) character mutation rate estimates suggested that a population with $N_e = 500$ could maintain innate amounts of genetic variation for hundreds of generations. Similar data were used to argue that a population's short-term viability required a minimal $N_e = 50$. As noted above, despite their rapid adoption by managers around the world, we now recognize that there can be no simple "magic numbers" for the MVP of all species.

There are two serious difficulties with the 50/500 approach to the MVP problem. The first involves some weaknesses in the data base from which they were derived. The second is that in addition to quantitative variation, there are other types of genetic variation relevant to estimating MVP size. These include the recessive lethal component of inbreeding depression (mentioned above), selectively neutral polymorphisms, and single genes of large effect. As an example of the latter, consider the human hemoglobin-S allele where heterozygosity is critical in resisting a major disease (malaria). The point is that definitions of MVP based on the original magic numbers and genetic uniformitarianism (all genes are equal in their significance) are unsound.

The applications of theoretic population genetics to conservation biology is in its infancy.^{22, 27} The complexity of the scientific issues involved has made progress in this area slow and often, as in the case of the magic numbers, subject to correction. Nevertheless, I strongly advocate the incorporation of genetic considerations into the management of the Thai biota. Unless more attention is paid to inbreeding and genetic drift, our reliance on protected nature

reserves to automatically or naturally sustain viable populations is doomed to failure. Our goal should be to preserve the innate or remaining genetic variability of carefully selected keystone and umbrella species whose survival will ensure the conservation of numerous other species. Steps must be taken to maximize the N_e of these selected species and management plans should be developed to monitor and maintain genetic variability. In cases where N_e is already very low, it should be increased by the judicious movement of individuals between patches of the metapopulation.

Demographic Consequences of Range Fragmentation

Despite my argument for greater attention to genetic aspects of conservation, it must be emphasized that extinction is fundamentally a demographic process. For many very small and endangered natural populations, demography is frankly more important than genetics. I would therefore argue that managers should, in most cases, pay more attention initially to increasing N even at the expense of genetic niceties involving N_e .

Small populations, especially those produced by range fragmentation, have a much greater chance of extinction because of random demographic accidents and local environmental variation. The Allee effect (diminished viability and reproduction) can be observed in cases where numbers or density are too low. Edge effects, both abiotic and biotic, further diminish patch size and may lead to emigration. Mathematical modeling currently focuses on critical patch size estimation and on the probabilities of between-patch dispersal. One interesting result is that as the amount of suitable habitat decreases, so too does the proportion of the suitable habitat that is occupied. Extinction occurs if the habitat area falls below some value related to an organism's life history and dispersal behavior. Thus, although the emphasis in this review is on genetic aspects of biodiversity conservation, PVA requires the consideration of both genetics and demography.

Defining Evolutionarily Significant Units for Biodiversity Management

The effective conservation of biodiversity presupposes the availability of a species inventory matrix. I use the term matrix because it is not sufficient to have traditional species lists of birds, plants, butterflies, etc. Conservation planning requires that we also know how these disparate species interact with one another. For example, a plant may require a specific species of butterfly as a pollinator and a specific species of bird as a seed disperser. Managers cannot conserve species out of their ecological contexts. We therefore face problems in Thailand as species inventories are still incomplete and more emphasis has traditionally been placed on taxonomically constrained research rather than on establishing the interdependencies of species in natural communities.

The most important units for biodiversity managers are species. Species are natural evolutionarily significant units that can be defined in terms of various morphological, behavioral, ecological and genetic criteria. Several evolutionary species concepts currently compete for the specialist's attention. Most readers will be familiar with species definitions based on reproductive isolating mechanisms. The "isolation" species concept may be contrasted with the antithetical

“recognition” species concept. Recent criticism of these concepts has led to the proposal of the “cohesion” species concept—the most inclusive group of organisms having the potential for genetic and/or demographic exchangeability.²⁸ Genetic exchangeability involves the factors (fertilization system, developmental system, and isolating mechanisms) that define the limits of spread of new genetic variants by gene flow. Demographic exchangeability involves the factors that define the fundamental niche and the limits of spread of new genetic variants through genetic drift and natural selection. Both replaceability and displaceability must be taken into account.

This brief theoretical aside on the complexity of the biological species concept underscores the challenge facing modern taxonomists and systematists. Far from being merely descriptive work (akin to stamp collecting, according to some ill-informed critics), taxonomy is increasingly recognized as requiring first-rate science and very complex problem solving. In Thailand, where perhaps one-third of the freshwater fish and flowering plants still need formal taxonomic attention, conservation planners are handicapped. Until managers know the limits of taxa of interest, cases of inadvertent mismanagement are inevitable. Without a reliable species-level taxonomy and a clear understanding of the evolutionary significance (if any) of recognized subspecies, races, etc., managers can make otherwise avoidable mistakes. All this is, of course, not limited to Thailand—the problem of defining evolutionarily significant units for biodiversity managers is a global one. Recent cases that underscore the importance of defining species properly have involved such allegedly well-known mammals as ibex, tigers, black rhinoceros, white rhinoceros, spider monkeys, and orangutans.^{29, 30} In the case of the black rhinoceros, managers urgently need to know which of the surviving 75 (mostly subviable) populations, now referred to several subspecies, can be pooled. In the case of the orangutans it is now clear that populations from Borneo are well-differentiated genetically from those on Sumatra; their management requires that they be kept separate. Questions of this type are just beginning to receive consideration in Thailand. In the case of the endangered brow-antlered deer *Cervus eldii*, should survivors of Burmese origin be reintroduced in areas of the northeast formerly occupied by a subspecies now restricted to Cambodia? If captive white-handed gibbons, *Hylobates lar*, are to be rehabilitated and reintroduced to sanctuaries in, for example, western Thailand, does it matter if those individuals originated in the far north, far south, or east of the country? Current data are inadequate to answer these questions but it is a good sign that such issues are now receiving attention.

Let me illustrate the importance of a sound taxonomy from two cases I have studied. In the Mekong River and Mun River there is a small snail, *Tricula aperta*, which transmits the human blood fluke, *Schistosoma mekongi* in Laos. In the Mekong, the snails are separated into two sympatric morphologically defined races: alpha and gamma. In the Mun river, all individuals are referred to a third, beta, race. Our studies of genetic variation showed that this taxonomy is flawed.³¹ This snail (now called *Neotricula aperta*) is actually four separate species—two in the Mekong (both of which have α and γ ecophenotypes) and two in the Mun. Which of these species are epidemiologically important in schistosome transmission will have to be reestablished. The second case involves the freshwater clams of the genus *Corbicula*.

Traditionally 28 species have been recognized in Thailand and a comparative study of their variation has just been completed. Genetically, it was discovered that at least 23 of these species are identical and should be synonymized.³² As this species is a human food source in some areas, and is a serious pest species elsewhere in the world, the importance of correct taxonomy cannot be overemphasized.

Species Management Practices

As noted above, simply providing a population with a suitable habitat may not ensure its survival. Conservation often requires active intervention. Among the important practices required for the management of small populations are:

- Maximize the genetically effective population size, N_e , for reasons explained above.
- Minimize the variance in population growth rate, r , as recent theoretical analyses suggest it is negatively related to persistence time under a fluctuating environment.
- Attain viable population size as soon as possible so as not to lose genetic variability.
- Monitor and maintain inherent qualitative and quantitative genetic variation for evolutionary fitness and disease resistance.
- Avoid outbreeding depression caused by the inadvertent mixing of well-differentiated populations or species.
- Facilitate natural behaviors including social and reproductive behavior, dispersal, and migration.

These and other management practices are discussed in greater details elsewhere.^{29, 33-34}

These practices are, of course, very difficult for wildlife managers to apply. In most cases, we lack the necessary information about genetic variation, geographic differentiation, and natural levels of gene flow. Gene flow is related to such variables as gamete, embryo (or seed), juvenile and adult dispersal. Furthermore, genetic changes often occur far faster than managers can respond. For example, range fragmentation can reduce a population's effective size, N_e , by 1-2 orders of magnitude in a single generation. Introduced species can extirpate a local population before it is missed.

Another type of problem involves the question of choosing species for management attention. On a world-wide basis, we have often selected species for their direct utility; they provide us with food, clothing, medicine, companionship or esthetic satisfaction. We have also devoted a disproportionate amount of attention to the charismatic megavertebrates like kouprey, elephant and giant panda. Far less attention has been given to equally spectacular endangered plants. In countries where endangered species have legal standing, we are beginning to see conservationists searching for species whose presence will block exploitative development. In the U.S., such inconspicuous endangered species as the snail darter (a small freshwater fish) and the spotted owl (a bird dependent on old-growth Douglas fir forests) have been used to focus public attention on the mismanagement of whole ecosystems.³⁵

In choosing species for attention in Thailand, it is important to remember that is very much easier and cheaper to manage species in the wild than it is in fragmented habitats and

small reserves. Zoos and botanical gardens are really the last resort, as the costs of maintaining a handful of species in captivity are the same as are required to manage large tracts of nature with thousands of species.³⁶ So the best approach appears to be to identify and conserve keystone species in nature. Keystone species play a disproportionately significant role in the communities in which they occur: in stabilizing biotic interactions, in biogeochemical cycles, in providing resources for other species. When keystone species are removed, we often observe a cascade of secondary extinctions among interdependent species. When detailed ecological studies required to identify keystone species have not been conducted, an alternative is to preserve umbrella species whose conservation will effectively protect many others. One example, mentioned above, is the spotted owl; protecting forest for the owl effectively conserves habitat for dozens of other species restricted to the old-growth forest community. Another involves the 12 species of plants (out of a flora of 2000) that sustain nearly the entire frugivore community, including 160 birds, for three months of the year in Manu National Park, Peru.³⁷ Focusing management efforts on a few well-chosen species can effectively conserve whole communities.

This discussion leads to another important principle of conservation: it is difficult to manage single species in isolation. One cannot hope to manage species in the face of climatic change and range fragmentation unless one understands their ecological interactions with other species. This includes such interactions as predation, competition, parasitism, pollination, seed and gamete dispersal. Such considerations often lead managers to the recognition that what goes on outside a park or reserve may be more important than what goes on inside. For example, in Costa Rica's western dry forest, many plants depend on 40 species of large sphingid moths for pollination.³⁸ These insects migrate 15-50 km annually to spend the "dry" season in rainforest habitats on the east side of the country. Conserving one community necessitates conserving the other; neither can be sustained in isolation. Such considerations complicate the manager's task.

This brief review of species management practices points to a number of general research needs in Thailand. First, we still need to complete the national biodiversity inventory. This is an urgent requirement, as habitats and species are rapidly disappearing. Achieving this goal will require the creation of greater opportunities for scientists to pursue careers in taxonomy and systematics, including strengthening the institutions which facilitate such research. Second, we need to pay more attention to defining conservation management units (populations, subspecies, species) carefully. This involves encouraging the genetic and demographic study of carefully selected species which serve as models of guilds of related species or as keystone species for whole communities. Population viability analysis, the analytical technique upon which future conservation management practices should be based, presupposes the availability of such information. The necessary research requires intensive and sustained field work coupled with sophisticated laboratory and computer work. It can rarely be undertaken by one person, but this problem can be overcome by fostering collaborative projects involving scientists from different institutions. Finally, we need to know more about the way species interact in various natural communities in Thailand. Many reserves are simply too small to permit proper functioning of ecosystem-level processes, and as a consequence, the conservation of some species

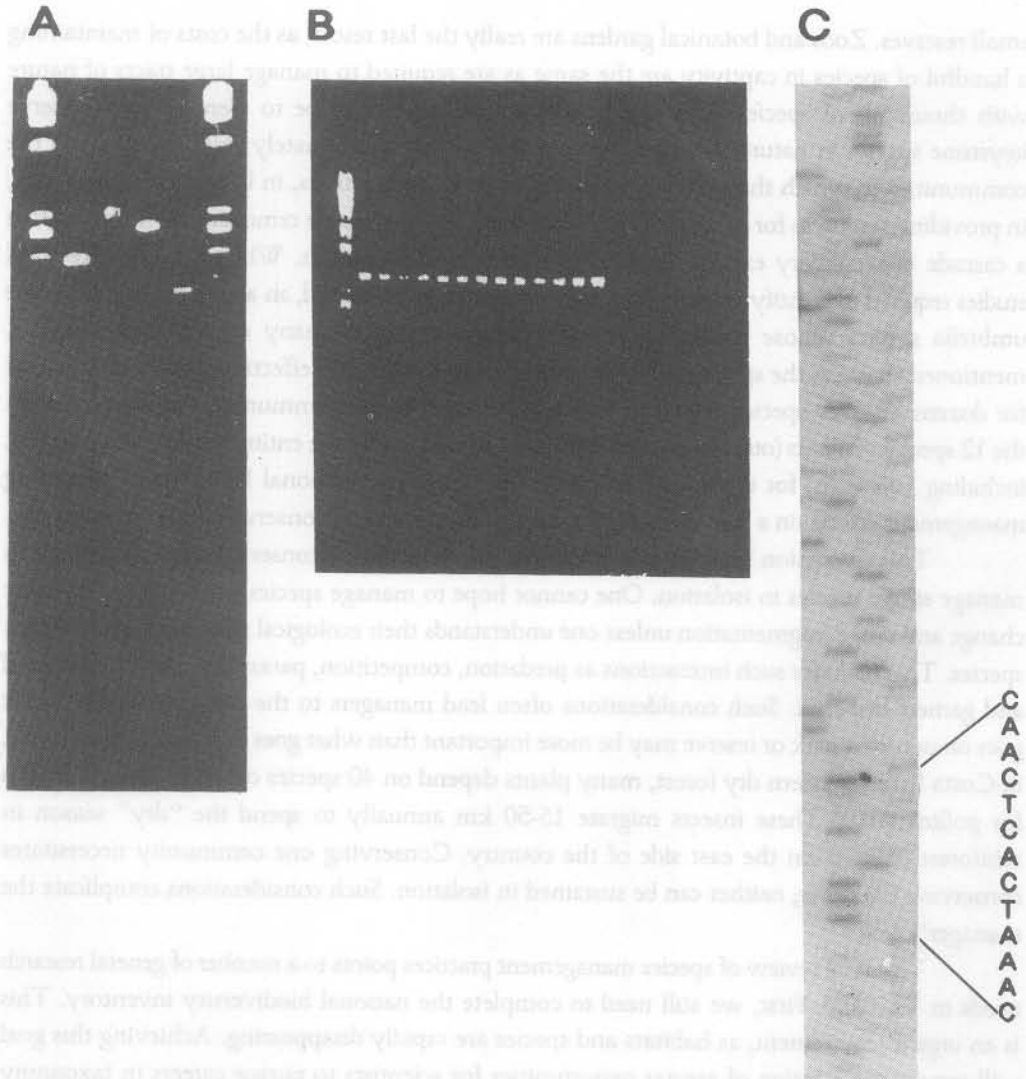


Fig. 1. Non-invasive method of studying genetic variation in Thai vertebrates. DNA is extracted from hair or feathers and selected genes are amplified by the polymerase chain reaction and sequenced directly.

A. Photograph of agarose gel stained with ethidium bromide. Fluorescing bands in lanes 2-5 are a nuclear gene (Mfd 23) isolated and amplified from hair of gibbons, *Hylobates lar*. Differences in band mobility between these four individuals indicate size polymorphism at this hypervariable microsatellite locus may be useful in pedigree analyses. Lanes 1 and 6 contain size standard $\phi\times 174/\text{HaeIII}$.

B. Photograph of agarose gel stained with ethidium bromide. Fluorescing bands in lanes 2-14 are a mitochondrial gene (cytochrome b) isolated and amplified from feathers of four species of hornbills. In the absence of size polymorphism at this locus (all bands have the same mobility) the gene must be sequenced to determine its usefulness for pedigree or phylogenetic studies.

C. Photograph of ^{32}P -labeled autoradiograph with the direct sequence of the single-stranded product of cytochrome b gene obtained from lane 10 of the gel figured in B. This provides the first sequence for the great hornbill, *Buceros bicornis*. The nucleotide sequence for a small portion of this gene is shown (A adenine, C cytosine, T thymine).

will require interventive management. The long-term survival of species represented by a number of isolated and subviable populations will depend, for example, on the artificial movement of genes between isolated populations and the rapid reintroduction of individuals following local extirpation. Research on this aspect of conservation science, and on the even more difficult task of community restoration,³⁹ is still in its infancy. Here, Thai scientists could benefit greatly from increased opportunities to interact with their colleagues elsewhere in the tropics and at the leading research centers elsewhere.

Genetics in the Conservation of Biodiversity

I conclude this review with a brief discussion of some of my own research. For the last decade, I have been primarily concerned with studies of mollusc species and their evolution.⁴⁰⁻⁴⁴ During this period, an increasing number of my study populations went extinct as a result of human activities, including habitat destruction and over-collecting. I have consequently become more interested in the application of genetics to the study of future evolution of species-to the conservation of small and threatened populations. Rather than continuing with molluscs, I have shifted my attention to mammals and birds.^{45, 46}

Studying DNA sequence-level variation in large samples of representative populations was impossible until recently. Furthermore, collecting tissue samples from free-ranging animals was difficult or impossible. Two technical advances have changed this situation. First, the polymerase chain reaction (PCR) technique now enables investigators to amplify minute quantities of DNA, thus eliminating the need for the acquisition of large blood or tissue samples which are difficult to handle in the field.^{47, 58} Second, new protocols for direct sequencing of PCR products eliminate the need to clone DNA samples and permit the study of large numbers of individuals without tedious blotting and hybridization.⁴⁹ These advances have enabled me to embark on several studies of evolution and conservation involving Thai mammals and birds.

First, in collaboration with Mr. Chira Meckvichai and Dr. Alongkorn Mahannop (Thai Zoological Organizations) and Dr. Warren Brockelman (Mahidol University), I have initiated a study of genetic variation in *Hylobates lar*, the white-handed gibbon. Phillip Morin and Carlos Garza, in my laboratory, have established that we can obtain adequate quantities of DNA from the base of hair plucked from captive animals (Fig. 1A). This approach will enable us to establish relationships among caged individuals and identify animals from different geographic areas. Such data will be useful in selecting appropriate animals for rehabilitation and reintroduction into protected forests and for improved management of captive animals.

Second, in collaboration with Miss Pilai Poonswad (Department of Microbiology, Faculty of Science, Mahidol University),⁵⁰⁻⁵¹ we have begun a study of genetic variation in hornbills. Phillip Morin has been successful in amplifying selected gene sequences from DNA obtained from the shaft of feathers (Fig. 1B). One such sequence is shown in Fig 1C. Our initial object was to establish relationships among social groups, but if we can obtain sufficient data on geographic variation, we will also be able to estimate current and former levels of gene flow. Such information, on natural levels of dispersal, are useful in the management of increasingly fragmented populations.

Third, we are planning an empirical study of the genetic and demographic changes in mammal populations that become isolated on fragmented rainforest patches. We are interested in monitoring changes in population viability in very small habitat patches, as a better understanding of these processes will lead to the improved management of fragmented populations of a type that are becoming so common in Thailand. For theoretical reasons involving the relationship between N_e and the rate of loss of genetic variation, we are especially interested in changes in population viability during the first 20 generations following isolation. We have identified a situation in Surat Thani Province where forest patches created by the filling of the Chiew Larn Reservoir in 1986-87 provide an excellent experimental setting. Following on the work of our collaborators, Mr. Seub Nakhasathien and Mr. Sawai Wanhongsa (Royal Forest Department),⁵²⁻⁵⁴ we will monitor genetic and demographic changes in several species of small mammals (rats, tree shrews and ground squirrels) by regular live trapping surveys. Mr. Antony Lynam, a graduate student committed to this project, has already established that we can obtain informative DNA sequences from a few hairs pulled from animals when they are released from the traps.

These projects are unusual in that they are among the first studies of genetic sequences based on non-invasive DNA sampling. If successful, they will provide results of considerable generality for the conservation of biodiversity in the tropics. I hope that in due course, it may be possible to report the results of these studies in this journal.

In conclusion, I have argued that genetic considerations will play an increasing role in conserving the world's biodiversity. Although the need for the application of genetic criteria to the management of cultivated plants and animals and the need to conserve the genetic resources in the wild relatives of cultivars is widely appreciated, in the future, many "wild" populations will require the same attention.⁵⁵⁻⁵⁷ As long as habitat alteration and destruction fragment natural populations, genetic processes will automatically reduce the viability of surviving populations. Managers will have to actively counter these undesirable effects or resign themselves to the ultimate consequences: extirpation and extinction. Paraphrasing Dr. M. S. Swaminathan, President of the International Union for Conservation of Nature and Natural Resources, the genes contained in plants and animals provide the foundation for sustaining life on our planet.⁵⁸ In our well-intentioned efforts to save individual species and communities, we must not lose sight of biodiversity's genetic basis. Gene conservation must be an essential component in any program to sustain living organisms.

Postscript. Since the October 1989 Biodiversity Seminar three notable developments have occurred. First, the proceedings of an international meeting on the relevance of metapopulation dynamics to conservation biology have been published.⁵⁹ Second, other reports on the effects of habitat fragmentation on species extirpation and community change have appeared.⁶⁰⁻⁶⁴ Third, colleagues have, appropriately, asked me to clarify the alarming opening sentence of this paper's Abstract; to specify just which species are going to disappear. Towards this end I have begun to assemble data on risk assessment for selected vertebrate species. Initial reviews of the various lists of officially threatened species show how true species-level endangerment has been underestimated.⁶⁵⁻⁶⁶

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REFERENCES

1. Wilson, E. O. and Peter, F. M., eds. (1988). *Biodiversity*. National Academy Press, Washington, DC. 521 pp.
2. Woodruff, D. S. (1990). Review of *Biodiversity*. (Wilson, E. O. and Peter, F. M., eds. 1988. National Academy Press, Washington, DC.) *Nat. Hist. Bull. Siam Soc.* 37, 259-261.
3. Wilson, E. O. (1989). Threats to biodiversity. *Scientific American* 261, 108-116.
4. McNeely, J. A., Miller, K. R., Reid, W. V., Mittermeier, R. A. and Werner, T. B. (1990). *Conserving the World's Biological Diversity*. IUCN, Gland, Switzerland, WRI, CI, WWF-US and the World Bank, Washington, D. C. 193 pp.
5. Myers, N. (1989). A major extinction spasm: predictable and inevitable? In: *Conservation for the Twenty-First Century* (Western, D. and Pearl, M. C., eds.) Oxford University Press, New York. pp. 42-49.
6. Diamond, J. (1989). Overview of recent extinctions. In: *Conservation for the Twenty-First Century* (Western, D. and Pearl, M. C., eds.) Oxford University Press, New York. pp. 37-41.
7. Reid, W. V. and Miller, K. R. (1989) *Keeping Options Alive: The Scientific Basis for Conserving Biodiversity*. World Resources Institute, Washington, D. C.
8. Soulé, M. E., ed. (1987). *Viable Populations for Conservation*. Cambridge University Press, New York. 189 pp.
9. Gilpin, M. E. and Soule, M. E. (1986). Minimum viable populations: processes of species extinction. In: *Conservation Biology. The Science of Scarcity and Diversity* (Soulé, M. E., ed.) Sinauer Assoc., Sunderland, Massachusetts. pp. 19-34.
10. Woodruff, D. S. (1987). Review of *Conservation Biology. The Science of Scarcity and Diversity* (Soulé, M. E., ed., 1986) Sinauer Assoc., Sunderland, Massachusetts. *Nat. Hist. Bull. Siam Soc.* 35, 63-68.
11. Gilpin, M. E. (1989). Population viability analysis. *Endangered Species UPDATE* 6, 15-18.
12. Soulé, M. E. (1989). Risk analysis for the Concho water snake. *Endangered Species UPDATE* 6, 22-25.
13. Brussard, P. F. and Gilpin M. E. (1989). Demographic and genetic problems of small populations. In: *Conservation Biology and the Black-footed Ferret*. (Seal, U. S., Thorne, E. T., Bogan, M. A. and Anderson, S. H., eds.) Yale University Press, New Haven. pp. 37-48.
14. Doak, D. (1989). Spotted owls and old growth logging in the Pacific Northwest. *Conservation Biology* 3, 389-396.
15. Shaffer, M. L. (1981). Minimum population sizes for species conservation. *BioScience* 31, 131-134.
16. Frankel, O. H. and Soulé, M. E. (1981). *Conservation and Evolution*. Cambridge University Press, New York. 327 pp.
17. Oldfield, M. (1989). *The Value of Conserving Genetic Resources*. Sinauer Assoc., Sunderland, Massachusetts. 360 pp.
18. Orians, G. H., ed. (1990). *Conservation of Genetic Resources*. University of Washington, Seattle.
19. Allendorf, F. W. and Leary, R. F. (1986). Heterozygosity and fitness in natural populations of animals. In: *Conservation Biology. The Science of Scarcity and Diversity* (M. E. Soulé, ed.) Sinauer Assoc., Sunderland, Massachusetts, pp. 57-76.

20. Ledig, F. T. (1986). Heterozygosity, heterosis, and fitness in outbreeding plants. In: *Conservation Biology. The Science of Scarcity and Diversity* (M. E. Soulé, ed.) Sinauer Assoc., Sunderland, Massachusetts, pp. 77-104.
21. Nevo, E., Beiles, A. and Ben-Shlomo, R. (1984). The evolutionary significance of genetic diversity: ecological, demographic and life history correlates. In: *Evolutionary Dynamics of Genetic Diversity*. (Mani, G. S., ed.) Springer Verlag, Berlin. pp. 13-213.
22. Lande, R. (1988). Genetics and demography in biological conservation. *Science* **241**, 1455-1460.
23. Ralls, K., Harvey, P. H. and Lyles, A. M. (1986). Inbreeding in natural populations of birds and mammals. In: *Conservation Biology. The Science of Scarcity and Diversity* (M. E. Soulé, ed.) Sinauer Assoc., Sunderland, Massachusetts. pp. 35-56.
24. O'Brien, S. J., and Evermann, J. F. (1988). Interactive influence of infectious disease and genetic diversity in natural populations. *Trends Ecol. Evol.* **3**, 254-259.
25. Wright, S. (1969). *Evolution and the Genetics of Populations*. University of Chicago Press, Chicago. vol. 2.
26. Franklin, I. R. (1980). Evolutionary change in small populations. In: *Conservation Biology: An Evolutionary. Ecological Perspective*. (Soulé, M. E. and Wilcox, B., eds.) Sinauer Assoc., Sunderland, Massachusetts.
27. Simberloff, D. (1988). The contribution of population and community biology to conservation science. *Ann. Rev. Ecol. Syst.* **19**, 473-511.
28. Templeton, A. R. (1988). The meaning of species and speciation. In: *Speciation and its Consequences*. (Otte, D. and Endler, J. A., eds.) Sinauer Assoc., Sunderland Massachusetts. pp. 3-27.
29. Woodruff, D. S. (1989). The problems of conserving genes and species. In: *Conservation for the Twenty-First Century* (D. Western and Pearl, M. C., eds.) Oxford University Press, New York. pp. 76-88.
30. Woodruff, D. S. (1990). Genetically based measures of uniqueness: Commentary. In: *The Preservation and Valuation of Biological Resources*. Orians, G. H., Brown, G. M., Kunin, W. E. and J. E. Swierzbinski, eds. University of Washington, Seattle. pp. 119-132.
31. Staub, K. C., Woodruff, D. S., Upatham, E. S., and Viyanant, V. (1990). Genetic variations in *Tricula aperta*, the intermediate snail host of *Schistosoma mekongi*: allozyme differences reveal a group of sibling species. *Amer. Malacol. Bull.* **7**, 93-103.
32. Kijviriya, V. Upatham, E. S., Viyanant, V. and Woodruff, D. S. (1991). Genetic studies of Asiatic clams, *Corbicula* in Thailand: allozymes of 21 nominal species are identical. *Amer. Malacol. Bull.* **8**, 97-106.
33. Schonewald-Cox, C., Chambers, S. M. MacBryde, B. and Thomas, L., eds. (1983). *Genetics and Conservation. A Reference for Managing Wild Animal and Plant Populations*. Benjamin/Cummings, Menlo Park, California. 722 pp.
34. Fiedler, P. L. and Jain, S. K., eds. (1990). *Conservation Biology. The Theory and Practice of Nature Conservation, Preservation and Management*. Chapman and Hall, New York.
35. Ehrlich, P. R. and Ehrlich, A. H. (1981). *Extinction: The Causes and Consequences of the Disappearance of Species*. Random House, New York. 305 pp.
36. Conway, W. G. (1989). The prospects for sustaining species and their evolution. In: *Conservation for the Twenty-First Century* (Western, D. and Pearl, M. C., eds.) Oxford University Press, New York, pp. 199-209.
37. Terborgh, J. (1986). Keystone plant resources in the tropical forest. In: *Conservation Biology* Sinauer Assoc., Sunderland, Massachusetts. pp. 330-344.
38. Janzen, D. H. (1986). The eternal external threat. In: *Conservation Biology* (Soulé, M. E., ed.) Sinauer Assoc., Sunderland, Massachusetts. pp. 286-303.
39. Jordan, W. R., Gilpin, M. E. and Aber, J. D., eds. (1987). *Restoration Ecology*. Cambridge University Press, Cambridge.
40. Woodruff, D. S., McMeekin, L. L., Mulvey, M. and Carpenter, M. P. (1986). Population genetics of *Crepidula onyx*: Variation in a Californian slipper snail recently established in China. *The Veliger* **29**, 53-63.

41. Woodruff, D. S., Carpenter, M. P., Saunders, W. B. and Ward, P. D. (1987). Genetic variation and species differentiation. In: *Nautilus: The Biology and Paleobiology of a Living Fossil*. (Saunders, W. B. and Landman, N. H., eds.) Plenum, New York. pp. 65-83.
42. Woodruff, D. S., Staub, K. C., Upatham, E. Suchart, Viyanant, V. and Yuan, H. C. (1988). Genetic variation in *Oncomelania hupensis*: *Schistosoma japonicum* transmitting snails in China and the Philippines are distinct species. *Malacologia* 29, 347-361.
43. Woodruff, D. S. (1989). Genetic anomalies associated with *Cerion* hybrid zones: the origin and maintenance of new electromorphic variants called hybridzymes. *Biol. J. Linn. Soc.* 36, 281-294.
44. Woodruff, D. S. and Solem, A. (1990). Genetic variation in *Cristilabrum primum*: a prolegomenon. *The Veliger* 33, 129-139.
45. Woodruff, D. S. and Ryder, O. A. (1986). Genetic characterization and conservation of endangered species: Arabian oryx and Pere David's deer. *Isozyme Bull.* 19, 33.
46. Merenlender, A. M., Woodruff, D. S., Ryder, O. A., Kock, R. and Vahala, J. (1989). Allozyme variation and differentiation in African and Indian rhinoceroses. *J. Hered.* 80, 377-382.
47. Innis, M. A., Gelfand, D. H., Sninsky, J. J. and White, T. J., eds. (1990). *PCR Protocols*. Academic Press, San Diego. 482 pp.
48. Mullis, K. B. (1990). The unusual origin of the polymerase chain reaction. *Scientific American* 262, 56-65.
49. Higuchi, R., von Beroldingen, C. H., Sensabaugh, G. F. and Ehrlich, H. A. (1988). DNA typing from single hairs. *Nature* 332, 543-546.
50. Poonswad, P., Tsuji, A. and Ngarmpongsai, C. (1983). A study of the breeding biology of hornbills (Bucerotidae) in Thailand. *Proc. Delacour/IFCB Symp.* pp. 239-265.
51. Poonswad, P., Tsuji, A. and Ngarmpongsai, C. (1989). A comparative ecological study of four sympatric hornbills (Family Bucerotidae) in Thailand. *Acta 19th Internat. Congr. Ornithol.* 2, 2783-2791.
52. Nakhasathien, Sueb (1988). The first wildlife rescue operation in Thailand. *Thai J. For.* 7, 250-265.
53. Nakhasathien, Sueb (1989). Chiew Larn Dam wildlife rescue operation. *Oryx* 23, 146-154.
54. Wanghongsa, Sawai (1989). Behavioral comparison of dusky leaf monkey (*Presbytis obscura*) on island habitat and undisturbed habitat of Khlong Saeng Wildlife Sanctuary, Suratthani Province. *Nat. Hist. Bull. Siam Soc.* 37, 1-24.
55. U. S. Congress, Office of Technology Assessment. (1987). *Technologies to Maintain Biological Diversity*. OTA-F-330. U. S. Government Printing Office, Washington, DC.
56. Woodruff, D. S. (1991). Genetics and conservation. In: *Integrating Conservation Biology and Agricultural Production*. (Gall, G. A. E., ed.) Elsevier Science, New York.
57. Western, D., Pearl, M. C., Pimm, S. L., Walker, B., Atkinson, I. and Woodruff, D. S. (1989). An agenda for conservation action In: *Conservation for the Twenty-First Century*. (Western, D. and Pearl, M. C., eds.) Oxford University Press, New York.
58. Swaminathan, M. S. (1990). Foreword. In: *Conserving the World's Biological Diversity*. (McNeely, J. A., Miller, K. R., Reid, W. V., Mittermeier and Werner, T. B. eds.) IUCN, Gland, Switzerland pp. 9-10.
59. Gilpin, M. and Hanski, I., eds. (1991). Metapopulation Dynamics. *Biol. J. Linn. Soc.* 42, 1-323.
60. Saunders, D. A., Hobbs, R. J. and Margules, C. R. (1991). Biological consequences of ecosystem fragmentation: a review. *Conservation Biol.* 5, 18-32.
61. Blake, J. G. (1991). Nested subsets and the distribution of birds on isolated woodlots. *Conservation Biol.* 5, 58-66.
62. Newmark, W. D. (1991). Tropical forest fragmentation and the local extinction of understory birds in the Eastern Usambara Mountains, Tanzania. *Conservation Biol.* 5, 67-78.
63. Laurance, W. F. (1991). Ecological correlates of extinction proneness in Australian tropical rain forest mammals. *Conservation Biol.* 5, 79-89.
64. Woodruff, D. S. (1991). Genetics and the conservation of animals in fragmented habitats. *Proc. Intl. Confr. Tropical Biodiversity*, June 1990, Kuala Lumpur, in press.

65. Woodruff, D. S. (1991). Threatened vertebrate species of Thailand. *Nat. Hist. Bull. Siam. Soc.* 38, 163-177.
66. Treesucon, Uthai and Round, P. D. (1990). Report on threatened birds in Thailand. *Tiger Paper* 17, 1-9.

บทคัดย่อ

พรรณพืช สัตว์ และจุลินทรีย์ ที่มีอยู่ในประเทศไทย กว่าครึ่งหนึ่งอยู่ในภาวะที่เสี่ยงต่อการสูญพันธุ์ในช่วง 2-3 ทศวรรษหน้า ลักษณะทางพันธุกรรมและการกระจายตัวของประชากรสิ่งมีชีวิต มีส่วนในการตัดสินใจอนาคตของสิ่งมีชีวิตนั้น ๆ ที่จะอยู่รอดและมีวิวัฒนาการต่อไป การวิเคราะห์การมีชีวิตรอดของประชากรชี้ให้เห็นความจำเป็นที่จะต้องพยายามรักษาความแปรผันของหน่วยพันธุกรรมดั้งเดิมของสิ่งมีชีวิตนั้น ๆ ไว้ให้มากที่สุด แนวคิดที่เกี่ยวกับประชากรที่ยังมีชีวิตรอดอยู่น้อยที่สุด และขนาดของกลุ่มประชากรในแง่ของลักษณะทางพันธุกรรม รวมทั้งผลกระทบที่เกิดจากการผสมพันธุ์ภายในสายพันธุ์เดียวกัน และการสูญเสียความแปรผันทางพันธุกรรม ได้ถูกกล่าวไว้ในบทความนี้ เนื่องจากพันธุกรรมมีความสำคัญในการอธิบายที่มาของสิ่งมีชีวิตแต่ละสปีชีส์ ดังนั้นจึงมีความจำเป็นรีบด่วนในการรวบรวมข้อมูลอย่างมีระบบด้านความหลากหลายทางชีวภาพ ลักษณะทางพันธุกรรม และการกระจายตัวของประชากรสิ่งมีชีวิตในประเทศไทยเพื่อการรักษาพันธุ์พืชหรือสัตว์เหล่านี้ไว้ นอกจากนั้นนโยบายที่เกี่ยวกับทรัพยากรธรรมชาติ จะต้องให้ความสำคัญในด้านการสงวนทรัพยากร การให้ความรู้ ฝึกอบรม และการทำงานวิจัยด้านนี้

65. Woodruff, D. S. (1991). Threatened vertebrate species of Thailand. *Nat. Hist. Bull. Siam. Soc.* 38, 163-177.
66. Treesucon, Uthai and Round, P. D. (1990). Report on threatened birds in Thailand. *Tiger Paper* 17, 1-9.

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