

GUEST EDITORIAL: DAVID LAMBERT



*David Lambert was born in Toowoomba, Australia and his initial degrees were at the University of Queensland. He later studied at the University of the Witwatersrand in Johannesburg for his PhD. He was appointed at the University of Auckland in 1980 and was, until recently, Leader of the Ecology and Evolution Research Group in the School of Biological Sciences and Director of Auckland University's Centre for Conservation Biology. David Lambert has now taken up the post of Professor and Head of the Department of Ecology at Massey University, Palmerston North. His early research was in the area of evolutionary cytogenetics of *Drosophila* (Australia) and *Anopheles* mosquitoes (Africa). Since then his interests have included evolutionary theory generally and the use of DNA tools to answer a broad range of questions in conservation and ecology. In particular Prof. Lambert's research group has studied a range of New Zealand animal species including insect groups, the kiore, Antarctic fish and a range of birds. A recent focus has been the use of minisatellite DNA technology to study parentage in species such as pukeko and skuas and to estimate levels of genetic variation in tuatara and the endangered Chatham Island black robin.*

THE NEW SCIENCE OF MOLECULAR ECOLOGY

The origins of Molecular Ecology

Most biologists, like scientists generally, have heroes - those who through their influence have changed the way we think about the world. These people typically open up new ideas about what is possible. One of my heroes is Paul Weiss who originated the term "Molecular Ecology". Weiss was an enigmatic character whose interests were extraordinarily diverse. His thought style was distinctly novel and iconoclastic. He made major contributions to the study of morphogenesis, systems theory and evolutionary biology. Weiss used the term 'Molecular Ecology' to mean the entire continuum of biological interactions between the molecular, cellular, organismal levels to the environment (Fig. 1). For example, he thought of the typical eucaryote cell comprising an array of molecular 'species' whose densities, distributions, arrangements and groupings are determined by their mutual dependencies and interactions, as well as the physical conditions of the space they occupy. He was philosophical about his new concept remarking: "Whether it [Molecular Ecology] will prove adequate, only the future can tell. Its main merit for the moment is that it presents us with a workable model by which the problems can be

stated with greater precision; it helps us 'focus our target'."

What is Molecular Ecology and is it really new?

Contemporary Molecular Ecology has come to focus much more on the outer shell that Weiss envisaged; the interactions of the organism with 'the environment' in general and, of course, such interactions are the stuff of ecology itself. Exciting recent developments in Molecular Ecology now provide scientists with a wide array of DNA tools by which to map and explore these interactions. Although collectively these techniques can assist in the resolution of a number of contemporary ecological problems, each of them has particular strengths and weaknesses and is applicable usually to a subset of problems (see Lambert and Millar, 1995, for a detailed discussion). Some of these problems have been, until recently, essentially intractable using more conventional approaches. Molecular Ecology has come to represent the use of DNA nucleotide sequence variation, nuclear genotypes and organelle haplotypes to gather information about natural populations. With the expansion of DNA tools there has been a dramatic increase in the application of ecological problems.

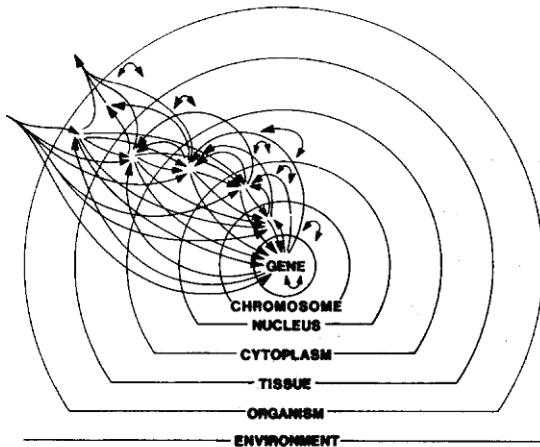


Figure 1: Molecular Ecology as depicted by Paul Weiss (Weiss, 1950); modified with kind permission.

While it can be reasoned that there really is nothing substantially new in Molecular Ecology - that it is simply the population genetics of the 1970s and 80s - to my mind, contemporary Molecular Ecology does represent a substantial advance. Developments in the technology of genetic analysis have brought into focus questions that were not specifically asked in the past. Consider, for example the common argument for the evolution of dominance hierarchies in animals. EO Wilson (1975) remarked:

"In the language of Sociobiology, to dominate is to possess priority of access to the necessities of life and reproduction. This is not a circular definition; it is a statement of a strong correlation observed in nature. With rare exceptions, the aggressively superior animal displaces the subordinate for food, for mates, and for nesting sites. It only remains to be established that this power actually raises the genetic fitness of the animals possessing it. On this point the evidence is completely clear."

What is clear is that at the time Wilson wrote, there was a general lack of data on precise estimates of reproductive success in many species. The relationship between dominance and reproductive success was simply assumed. With the developments of a range of new genetic technologies it is apparent that the situation is far from clear. DNA "fingerprinting" (an increasingly generic term for an array of both DNA probe and PCR-based methods) has revealed extraordinary diversity in the genetic consequences of mating systems, particularly in animal populations. For example, our own work using both single and multilocus minisatellite DNA probes has shown that there is no relationship between dominance and reproductive success in New Zealand pukeko (Lambert *et al.*, 1994). The important point

here is that the question - do dominant males achieve high reproductive success? - was, prior to the advent of Molecular Ecology, not commonly even asked in an empirical sense, owing to a lack of techniques available to answer the question.

Perhaps of even more importance is the point that Molecular Ecology has provided more powerful tools to refute existing ideas in ecology and evolution. For example, in the case discussed above, an easy and seductive idea is that behavioural dominance is an obvious biological 'mechanism'. But it turns out to be dangerous to assume that the universal 'function' (Lambert, 1995) of dominance is to enable genetically superior individuals to differentially reproduce, precisely because in our study of pukeko, we have shown that there is no relationship between reproductive success and dominance. Hence, Molecular Ecology can help us to refute existing ideas. Although this approach is a very powerful one it does not directly lead to novel concepts in ecology and evolution. Its role is, via refutation, to free people to think in new ways and thereby indirectly to encourage the introduction of new concepts.

Genes as markers

Molecular Ecology uses genes essentially as markers to estimate ecologically important variables. In fact, there are two broad approaches that biologists take to the study of genetics. Typically, their focus is either to investigate genes in relation to the role they play in the development of organismic form (*sensu*, behaviour, morphology etc.). From this perspective, genes are typically conceived of as representing some sort of 'Central Directing Agency' in relation to ontogeny. In contrast, Mendel himself used phenotypes of organisms (e.g., the shape of pea seeds) as markers to investigate the action of genes themselves. By examining the phenotypic patterns in different generations, Mendel was able to infer the action of genes at meiosis. Over the subsequent history of population genetics, biologists have used genes themselves as markers to investigate population level phenomena. Throughout the history of population biology these genetic markers have become progressively more precise and specific. We have moved from chromosome markers to isozymes and latterly, with the proliferation of DNA methods, to the direct examination of single locus, multilocus and nucleotide sequence variation.

An important use of genetic markers has been the detection of cryptic species - those that were indistinguishable on morphological grounds. There are many examples of the use of genetic markers in this way. My own studies on the African mosquito *Anopheles marshallii* revealed the existence of a complex of species using polytene chromosomes as genetic markers (Lambert, 1982). In recent years the use of more genetically variable markers such as multilocus minisatellite DNA approaches has enabled

the determination of parentage in natural populations, a problem typically unresolvable using chromosome or even isozyme markers. In fact, Moore (1993; quoted in Burke, 1994) has recently reported that the most cited papers in ecology and environmental biology from the period 1987-92 were those that first reported this approach. In New Zealand minisatellite DNA studies have been used to develop methods for sexing brown skua on the Chatham Islands (Millar *et al.*, 1992) and to examine population structure and inbreeding in blue duck (Triggs *et al.*, 1992). Parentage has been precisely determined in two communally breeding New Zealand avian species, the pukeko (Lambert *et al.*, 1994) and brown skua (Millar *et al.*, 1994). Christen Williams and coworkers (Williams *et al.*, 1994) have also recently employed a polymerase chain reaction (PCR) test to trace the origin of the New Zealand populations of Argentine stem weevil. Using randomly amplified polymorphic DNA (RAPDs) to examine nine populations from South America, five from New Zealand and one from Australia they presented evidence that the New Zealand populations originated from the east coast of South America. Mitochondrial DNA sequence variation now allows the investigation of the evolution of such ecologically important molecules as the antifreeze proteins of Antarctic fish (Bargelloni *et al.*, 1994).

The potentials of Molecular Ecology

A wide array of contemporary molecular tools now allow ecologists to answer an ever increasing range of questions. Craig Millar and I have recently discussed which of these tools is appropriate to which ecological questions, particularly in relation to the levels of genetic variation that each detects and their modes of inheritance (Lambert and Millar, 1995). In addition to the more commonly addressed issues, Molecular Ecology can now be used to determine the diet of animals from an analysis of their faecal pellets (Höss *et al.*, 1992), to the development of new vaccines for avian diseases (Purhase, 1989) and the identification of black rhinoceros horn (Arnheim *et al.*, 1990) (see Lambert and Millar, 1995 for a full discussion). Molecular ecology is an essential tool in ensuring the proper assessment of the risks of the release of genetically modified organisms (GMOs) (Williamson, 1992).

Our knowledge of the molecular processes that underlie evolutionary change has been, until recently, based on comparisons of the genes of living species. Unlike the remains of animals and plants themselves, DNA does not leave impressions in the rocks. However, ancient DNA, although degraded, can survive the ravages of time. To date, DNA from a number of extinct animals and plants has been amplified using PCR and the DNA sequence successfully determined. The oldest and most important instance is the woolly mammoth, a frozen

carcass found in the permafrost of Siberia. This species is thought to have lived 40 000 years ago and the amplification and the subsequent analysis of its DNA represents a major advance for ancient DNA studies. The Antarctic environment is less disturbed, more isolated, and colder and drier than that of Siberia. Hence tissues recoverable from this environment are likely to represent a major reservoir of ancient DNA. In fact, the Antarctic is potentially the most productive source of ancient DNA owing to these very unusual conditions which are ideal for the long-term preservation of DNA. Subfossil skin and bones of several species, including Adélie penguins, found in Antarctica are prime resources for ancient DNA studies.

Microsatellite DNA are ideal markers to investigate the problems associated with the biology of Adélie penguins colonies in the Antarctic and, in addition, are potentially able to provide a source of genetic data for the study of subfossil populations. New studies in Molecular Ecology will enable the direct examination of changes in gene frequencies, not only across space, but over considerable periods of geological time - an achievement never before possible!

Molecular Ecology of plants and microbes

Owing to my own personal bias, the above comments have focused on animal species. There are, in addition, a similar range of opportunities involving plant species in New Zealand. An array of genetic techniques involving minisatellite and microsatellite DNA, restriction fragment length polymorphisms (RFLPs) and other single locus genetic analyses are also available for plants (see the recent review by Kochert, 1994 for details). DNA 'fingerprinting' has been used to investigate aspects of population structure in blackberries and raspberries (Nybom *et al.*, 1989) and recently to identify clones of rubber tree (Besse *et al.*, 1993). In addition, the review by Steffan and Atlas (1991) examines an array of PCR-based tests to detect the presence of bacteria and viruses in both water and soils. Deverex, Kurtz and Mundfrom (1993) have used DNA probes and PCR techniques to detect variation in 16S ribosomal RNA genes, consequently revealing previously unsuspected diversity among sulphate-reducing bacteria. In addition, these methods provide information about the changes in the composition of microbial communities, which might be indicative of environmental stress. It is clear that Molecular Ecology will be equally influential in the study of plants and microbes.

The future for New Zealand's Molecular Ecology

That the tools of molecular genetics will continue to play a major role in ecological studies is beyond dispute. I suspect that, as such tools become even more powerful and diverse in their ecological

applications, the new science of Molecular Ecology will become increasingly more diffuse. In fact, a mature science of Molecular Ecology will inevitably be subsumed by 'ecology' proper. This is exactly as it should be. Since ecology is fundamentally a problem oriented science (which is not to say that it has no theoretical constructs), it will need to engulf any and all relevant approaches and disciplines in order to be better able to answer contemporary questions. And through the reciprocal interactions between the technologies of Molecular Ecology and the theories of ecology, new questions will be formulated and old ones tested.

Novel DNA technologies, useful in the study of natural populations, appear almost daily. Realistically New Zealand can play only a modest role in these worldwide developments, but many of these new technologies are being marketed commercially and are becoming readily available to a wide range of ecologists. There are consequently, exciting opportunities for New Zealand ecologists to use these developments to study our unique fauna and flora. Thought of in this way, we have a tremendous advantage in this country. By bringing together new genetic methods and our remarkable plants, animals and microbes, there are exciting ecological opportunities ahead.

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