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# 20838 Consultant: Kalph KIREY Bedistop Of Ms. Rhinel IRD/INF Genetic Diversity in South Africa - Conservation & Sustainable Utilisation.

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### Introduction

Africa and southern Africa as part of the continent have a series of pressing and important problems with respect to the conservation of genetic diversity. South Africa has an additional one because it is the country with the world's third most abundant biodiversity covering only 0.8% of the world's land surface, but South Africa has greater than 8% of the world's higher plant species, 8% of the world's bird species, 5.8% of the world's mammal species and 4.6% of the world's reptile species. The biodiversity of lower animal, lower plant, fungi and bacteria in South Africa's many diverse enviroments is unknown. The problems revolve round the need to support an ever rising population with increased expectations of improved living standards by exploitation of the natural resources of the continent. Physical natural resources have a finite lifespan when exploited and their value depends on the benification of those resources within the place of origin as to their overall value to the population. Biological natural resources, by contrast, are renewable and represent a permanent natural asset which should be exploitable in the long term by the country containing the asset.

However, mankind is part of the biosphere and therefore interacts directly with biological natural resources and the very pressures that require the exploitation of Africa's natural resources also directly put pressure on the biological resources by competition. Like physical resources, biological resources can be split into two groups: firstly, those resources which are known and accessible; and secondly, those resources which are unknown and require discovery. In the case of physical resources, the two groups are obvious: the first group is the visible and already exploitable resources, for example, gold in South Africa, diamonds in Namibia and oil in Nigeria; the second group consists of minerals etc which have yet to found. Many companies spend millions of US dollars on exploration for the second group of resources with vast returns to the company and the country that finds of such a resource. Biological resources of the former type consist of known biological species with a variety of known uses. However, outside of academic circles, with a few notable exceptions, very little is done to identify unknown biological entities such as new species and sub-species. In this article, I wish to explore the approaches available to African scientists to study, enumerate and monitor the biological resources of Africa both on the land, in its inland waters and in its coastal area with particular emphasis on the approaches being used in South Africa to analyse these problems. Also under consideration will be the methodologies presently used in South Africa can be transferred simply and cheaply to other African countries. It is patently obvious that African scientists and preferably scientists from within the country under study should be responsible for the evaluation of the biodiversity within Africa rather than scientists from the western world. The biological resources of an African country belong to that country and even though part of that resources may be exploitable by a multinational company, only if the initial discovery, research and evaluation is done by an African scientist does the research funds, scientific reputation and any monetary value return to the scientist, his institution and his country. In many cases, especially with ongoing evaluations of biodiversity, a scientist from outside the country is at a disadvantage and repeated travel to and from an outside country increases significantly the cost of such research.

The International Union of Biological Societies adopted the *Diversitus* programme at its 24th General Assemby in 1991 to study species diversity at all levels including animals,

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plants, miroorganisms, cells, species, communities, ecosystems and landscapes. The specific scientific themes include:

1) Ecosystem function in biodiversity.

2) Origins, maintenance and loss of biodiversity.

3) Biodiversity inventorying and monitoring.

4) Conservation of the wild relatives of cultivated plants and domesticated animals.

5) Marine diversitry.

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6) Microorganism diversity.

7) The human dimension of biodiversity.

Although European, North American, West Pacific Asian and Ibero-American regional programmes have been set up, Africa lags behind in this programme and other UNESCO based programmes. Funding is mostly country based which means than in poorer nations, biodiversity must have a lower priority unless profit can become a driving force.

### Genetic Diversity and Sustainable Development: The Problems.

### The Known Species.

The problems which fall into this group tend to be associated with macro-organisms for the most part. These organisms are the ones which have the highest profile in the human mind and have been studied and analysed for many years. The need to conserve these organisms has been in the forefront of governments minds for many years and South Africa has done its part in the preservation of these ecosystems with two examples coming to mind, the creation of the Kruger National Park and the Addo Elephant National Park which have been extremely important in the preservation of two distinct populations of elephant in South Africa. However, the impact of man on the environment continues to fragment wild populations creating a number of problems which must be addressed. This is particularily important as the number of private game reserves in Africa increases. In the 1970's in the Northern Transvaal of South Africa, Mala Mala, Londolozi and Sabi Sabi removed the cattle from their large ranches and began bush clearing to restore open grasslands previously encroached by bush due to overgrazing. The result was game lodges commanding tariffs up to US\$500 per night which have become significant employers. As this type of enterprize has spread through all of southern Africa, amny new jobs based on ecotourism have been created. However, the long term viability of this job creation depends on sustainable ecosystems which become more difficult to maintain as the size of the game racnch becomes smaller. On line management of this type of ecosystem is required and some of the problems are decribed below.

### Inbreeding.

As populations have become fragmented by urbanisation and the building of fences, the natural migrations of individuals and groups has been disrupted. This is true for plants as well as animals where vast areas of cultivation have isolated individual plants and plant populations from nearby relatives. This phenomena is exacerbated by the trend to private game parks where animal populations are isolated by fences for economic reasons. Small populations give rise to two genetic problems inbreeding and lose of heterozygosity. Both can lead to depressed levels of reproduction and the appearance of aberrant individuals as genes which are normally recessive within the population are expressed. It is important that these phenomena are monitored and populations are maintained in as viable condition as possible

### Translocation.

This leads on from the previous point. If one is maintaining a large number of relatively small populations, it is essential that individuals be translocated from population to populations to maintain gene exchange. It is also important when establishing new

populations, for example in a private game park, that when the animals and chosen for this park, the greatest genetic diversity possible is obtained. Thus, in any case where translocation takes place, the maximising of the genetic diversity of the populations undergoing translocation is carried out. In many cases this can be done empirically but bring in animals or plants from populations which are not known to be related to the population receiving them. However, in some cases where the populations small and interrelated, a means of establishing genetic diversity should be used.

#### Local Adaption.

Translocation can give rise to another problems. Particular populations, especially those that are highly isolated by physical barriers can become genetically adapted to a particular environment, this being a step in the evolution to new species. If there is an introduction into such a population or such a population is translocated to a new environment where it interbreeds with another population, this can potential give rise to the loss of the genes particular to the local environment. Thus, it is necessary to establish if natural populations are highly genetically distinct and therefore their local gene pools should be preserved perhaps by captive breeding.

### The Unknown Species

Separate from the above problems but linked to increasing urbanisation and destruction of habitats are the problems arriving from unidentified levels of genetic diversity. These can be divided into two groups.

### **Cryptic Species**

These occur where isolation and other evolutionary genetic effects have created what amounts to new species or sub-species which have not been detected by classical morphological means. Any type of environmental impact on such small unique populations could result in an unwitting extinction. In many respects, the smaller that the organism is, the more difficult it is to detect such cryptic species; however, in general, most such species are eukaryotes.

### Novel Organisms

The microbiological world probably contains millions of yet undiscovered organisms which may be capable of producing novel useful compounds and processes. Modern medicine is dependent on antibiotics isolated from soil microorganims and molecular genetics depends for many processes on enzymes isolated from organisms from extreme enviroments. Detection of such organisms from environmental samples is exploitation of the resources of the country from which the sample originated.

### Molecular Genetic Methods and their Applications

The above problems require studies on individuals and populations which are rapid, relatively cheap, functional in an african environment and allow analysis of the individual genetic structure of members of the population and comparison between individuals. The molecular genetic techniques described below make this possible and their suitability is commented on.

### Allezyme Analysis.

This method of analysis involves the isolation of active enzymes by non-denaturing gel electrophoresis using usually starch gels but sometimes using acylamide gels. The proteins are separated on both charge, shape and molecular weight such that these parameters cannot be estimated from the mobility. Once separated, the proteins are detected using specific chemical stains which produce a colour reaction at the position of the band due to the specific activity of the enzyme. A very wide range of such stains have been developed for enzymes such as dehydrogenases, amylases, nucleases, etc. A number of different bands can be identified from one gel representing either different mutant versions of the same gene in a polypoid organism or different genes making the same type of enzyme at different genetic locations in the genome of the organism. In complex higher organisms, the enzymes present are tissue specific and only samples from the same pure tissue can be directly compared with each other. Because enzyme activity must be retained, the samples must be treated very carefully and repeated freezing and thawing or storage at high temperatures must be avoided. Analysis is usually carried out manually due to the complex nature of the interpretation needed. Some problems are associated with this method, particularly in the context of southern Africa: firstly, sampling of specific tissues tends to require dead specimens and for rarer species this can be a major problem; secondly, the collection and transportation of delicate samples over long distances can be a problem in Africa where a cold chain can be difficult to maintain; thirdly, separate gels must be run for each enzyme although methods of reducing the workload due to this have been established; fourthly, in animals and plants which a large number of isoenzymes and which are of two or higher ploidy, interpretation of gels can be difficult even for an experienced scientist and finally, the availability, transportation and cost of the specialist chemicals required for each enzyme detection system can be problematic. The method has two major advantages. Firstly, that it allows a direct measurement of the heterozygosity at specific loci and an overall estimation of the average heterozygosity for the loci studied. Inbreeding and small populations are characterised by low levels of heterozygosity. Secondly, that specific alleles of a particular gene can be traced from population to population which can allow conclusions to be drain how populations are or were connected by interbreeding.

### SDS Acrylamide Gel Electrophoresis of Proteins.

This method also uses protein samples and requires them to be taken from specific tissues in higher animals and plants. However, the proteins are separated by electrophoresis

on acrylamide gels in the presence of sodium deodycle sulphate (SDS) which is a charged detergent. SDS denatures the proteins randomising their shape and binds to them equalising their charge. Thus separation on the gel is accomplished on molecular weight alone. Staining is carried out using generalised protein stains such as commassie blue or silver stain which means that specific functions can not be linked to particular protein bands, unlike above. Different proteins of the same molecular weight will run to the same point in the gel meaning that they can superimpose. However, samples are not as vulnerable to damage from changes in their environment although a good cold chain is still needed.

Changes in molecular weight of specific proteins occurs more slowly in genetic terms than changes in charge and therefore changes in the proteins in a population measured by SDS acrylamide gel electrophoresis must take a longer time to occur. Thus although they can be used to intra-population and inter-population genetic variability, they become more useful at the discrimination level of sub-species and species. Analysis can be carried out either manually or electronically and are usually calculated as a percentage of band shared between two individuals. This methods major advantages are firstly: its simplicity, requiring relatively few tools and chemicals; secondly that it is fast and can be used cost effectively for preliminary studies.

### **RFLP Analysis of Complete Organelle DNA.**

Mitochondria and chloroplasts are subcellular organelles that contain their own genetic material in the form of organelle specific circular DNA molecules. The size of these DNA molecules vary and can be as small as 18,000 base pairs in many vertebrate mitochiondia but can be as large as 200,000 base pairs in the chloroplasts of higher plants. The organelles and DNA are, in general, maternally inherited and thus can be used to trace maternal lineages. Changes to the DNA sequence in the organelles occurs from time to time by mutation and these changes are perpetuated from generation to generation if they do not directly affect the

function of the organelle, which many do not. Such changes can be detected if the whole circular DNA molecule is isolated and subjected to a battery of type II DNA restriction endonucleases which cut the DNA a specific sequences in the DNA ranging from four base pairs to eight base pairs. Thus a change within a specific site could either eliminate a so called restriction site already present or create a new site. The pieces of DNA produced by the restriction endonuclease are separated on agarose or acylamide gels and the fragments detected either using the interchelating agent ethidium bromide or silver staining. The bands produced can either be used to deduce a complete two dimensional map of the circular organelle (possible easily with 18,000 base pairs of the vertebrate mitochondrion) or allowing the calculation of percentage band shared between individuals. The changes are called restriction fragment length polymorphisms or RFLPs. In either case, it is possible to estimate the sequence divergence between individuals and the average sequence divergence within and between populations. It is also possible to trace mitochondrial types from one population to another. The rate of mutation of organelle DNA is about ten times higher than for nuclear DNA and it thus changes quite rapidly with time. Large diverse populations contain many different RFLP types within their organelle populations. Inbred, bottlenecked, small or recently evolved populations contain few organelle types.

The advantages of this approach rest on fact that good methods of isolation of organelle DNA and particularly vertebrate mitochondrial DNA have been developed and that this type of analysis can rapidly give an indication of populations which are under threat. The disadvantages are tissue sampling and storage can be important in getting good yield of analysable DNA, the analysis is relatively time consuming if done to completion and that it covers a limited part of the genome of the complete organism which is specialised and maternally inherited.

### Analysis of Polymerase Chain Amplified Fragments of Organelle DNA.

The complete and partial DNA sequences of a significant number of mitochondrial and chloroplast DNAs are known and detailed comparisons of such DNAs have been carried out such that it is possible to identify conserved regions of such DNA and other regions which are subject to greater variability. Polymerase chain reaction allows the amplification of as little as a single molecule of DNA to ugm quantities of identical DNA if two specific priming sites can be identified which flank the area which is to be amplified. This is possible for a number of regions in organelle DNA but, in particular, the D-loop region, the rRNA region and the cytochrome oxidase II region of mitochondrial are commonly used for this purpose. Once such a sequence has been amplified, it can be analysed either using the RFLP technique described above which is relatively easy and rapid or by DNA sequencing of the complete fragment which is more costly, time consuming and difficult. Either way, the results can be analysed to provide sequence divergence between individuals, within populations and between populations for these maternally inherited sequences.

RFLP analysis on such small pieces of DNA (about 1000 base pairs in size) is not highly efficient but can give good results. The more complex approach to DNA sequencing is more difficult to set up and more costly to run in an African context although it gives better results. An example of this is the identification of meat from protected whales in shops in Japan by PCR amplification using a portable thermocycler. This emphasises one major advantage of this approach which is that it can be carried out in the field using a generator and minimum equipment. The amplified sample can then be returned to the laboratory for analysis avoiding the need to transport samples. However, this method is probably the least vulnerable to sample deteriation and preservation using freezing, drying or alcohol have been successfully used. It can also be used on museum specimens, mummified samples and samples preserved in other ways, for example, in amber. This allow studies of genetic variation in both the physical and temporal dimensions for the same criteria as complete organelles. It major disadvantage is the same as for complete organelle analysis which is that it studies an even more limited range of maternally inherited genes.

### Randomly Amplified Polymorphic DNA Sequence (RAPDS) Fingerprinting.

This is another polymerase chain reaction based technique in which short (10 base pair long) single primers are used to amplify random fragments from the complete genome of an organism. The genome size of organisms vary from  $2x10^6$  base parts for the smallest bacteria up to 10<sup>9</sup> base pairs upwards for higher plants and animals. Thus, under the conditions of amplification applied, the primers allow random DNA sequences, usually less than 1000 base pairs in size to be amplified. The resulting DNA fragments are separated by either agarose or acrylamide gel electrophoresis, detected and their size measured. Each band represents a particular pair of 10 base pair sites with a unique sized piece of DNA between the two primer sites. Thus, each band samples the organisms genome for variation in that region. Percentage band sharing between organisms and within/between populations can be easily estimated and even sequence divergence can be calculated with a few assumptions. Heterozygosity cannot easily be estimated because DNA bands are dominant and paired alleles from the diploid chromosome pairs are hard but not impossible to find. RAPDS fingerprinting allow identification of unique populations and the measurement of genetic diversity within them. It can be used to relation different populations to each other and to do limited phylogenetic analysis at the sub-species and closely related species level. It cannot be used to study distant phylogenetic relationships which is possible with organelle DNA.

The advantages of RAPDS fingerprinting is that it is quick and cheap allowing large numbers of samples to be processed; that it gives excellent information on the structure and relationship within and between populations and that it covers a significant area of the total organisms genome. As there are thousands of ten base pair primers available, the coverage is only limited by the number of primers used in the study. A minimum of four and usually between six and ten are adequate. Depending on the questions being asked, then one or two primers giving significant genetic variation can then be targeted for the complete study. The techniques disadvantages are that it is laboratory and instrument dependent; that sample preservation can be a problem but usually is not and that analysis of the data objectively for a large number of samples requires an electronic data capture and analysis system.

### Microsatellite DNA Fingerprinting.

This is another polymerase chain reaction based method which examines the variation in specific very short repeats found in the eukaryotic genome. A cloned chromosomal DNA library of the organism under study is made in a high copy number bacterial plasmid vector with a insert target size of about 500bp. This library is probed using a radioactively labelled short oligonucleotide which has been synthesised to contain the repeat motif which has been targeted. Clones which give a strong signal with the probe using autoradiography are identified and the plasmid DNA extracted from them. The insert DNA within these plasmids is then sequenced and the repeat motif found within the cloned sequence. This motif will be flanked by unique DNA sequences which are then used to design two primers for polymerase chain reaction amplification of the repeat motif. Repeat motifs within the eukaryotic DNA are subject to slippage on replication and the number of repeat units within the motif can increase or decrease over a number of generations. Thus when a number of individuals from a population are analysed using the pairs of primers and polymerase chain reaction on an acylamide gel system variation in the overall number of repeat units in the specific locus of the organism can be detected. Overall genetic variability can be estimated and heterozygosity can be measured as the two alleles in the diploid can be identified unlike with RAPD fingerprinting. Furthermore, a limited amount of multiplexing can be carried out allowing up to three or four different loci to be analysed in one polymerase chain reaction.

This process can give the most detailed results for a specific species and population including the heterozygosity within populations, the variation in heterozygosity between populations, genetic variation within populations and genetic variation between populations, all at the DNA level. However, in the context of Africa, the method has a number of disadvantages. Firstly, the microsatellite repeats which are studied are not characteristic of the whole of the genome of the organism and may not reflect, in some cases, variation in the unique genes which are genes most important to the organism. Secondly, this system works for higher eukaryotes and not for prokaryotes and some lower eukaryotes. Thirdly, the primers for each locus must be identified and sequenced for each species studied; although some primers may be used in a number of closely related species, results are much better if the primers are targeted directly at the species from which they came. Finally, the technology required to carry out a microsatellite project is relatively high, not easily transportable into a low technology environment and expensive from the point of view of the work required to obtain only one set of primer and between ten and twenty are required for a good study. However, if suitable target primers have been identified elsewhere, the microsatellite analysis of a population using polymerase chain reaction is a good option.

### Analysis of Targeted Polymerase Chain Reaction Amplified Genes.

This method gets round one of the disadvantages of microsatellites in that unique genes are targeted for amplification by specific pairs of primers using polymerase chain reaction. Again heterozygosity and genetic diversity within and between populations can be estimated for each targeted unique gene. However, this process requires that the sequence of the gene in question is available. This requires either the cloning and sequencing of the gene in question or the sequence of the gene to be available from other workers in the DNA sequence databanks. This approach is viable for sequences from well studied organisms of all types such as the great apes and the ability to identify polymorphism can be increased by

using such techniques as Thermal Gradient Gel Electrophoresis. However, for the majority of organisms in Africa and any novel species, this method is not really practical.

### South African Molecular Population Genetic Research Groups

Below is a list of the major research groups in South Africa in molecular population genetics with their areas of technical expertise and their major research interests. The list is not complete and Departments of Genetics, Zoology or Botany exist at a number of other Universities which have specific interests in certain problems.

## Department of Biochemistry and Microbiology, Rhodes University, Grahamstown and JLB Smith Institute of Ichthyology, Grahamstown.

### Technical Expertise:

SDS acrylamide gel protein profiling.

Polymerase Chain Reaction (PCR) amplification of DNA .

DNA sequence analysis.

Molecular analysis of mitochondrial DNA.

Randomly Amplified Polymorphic DNA (RAPD) fingerprinting.

Thermal Gradient Gel Electrophoresis (TGGE) analysis of genes.

### Research Interests:

Population genetics of threatened species of marine and freshwater fish.

Population genetics of dolphins.

Population genetics of ostriches.

Population genetics of endangered fynbos plant species.

Population genetics of African bees.

Population genetics of marine gastropods.

Identification of novel bacterial fungal species from the southern African environment.

Population genetics of medically important bacteria in southern Africa.

### Department of Chemical Pathology, Medical School, University of Cape Town and Sir Percy Fitzpatrick Research Institute for Ornithology, University of Cape Town.

### Technical Expertise:

Polymerase Chain Reaction (PCR) amplification of DNA .

DNA sequence analysis.

Molecular analysis of mitochondrial DNA.

### Research Interests:

Population genetics of birds.

Population genetics of rhino.

### Mammal Research Institute, University of Pretoria.

### Technical Expertise:

Polymerase Chain Reaction (PCR) amplification of DNA.

DNA sequence analysis.

Molecular analysis of mitochondrial DNA.

### Research Interests:

Population genetics of mammals in general and threatened species in particular.

### Department of Genetics, University of the Witwatersrand.

### Technical Expertise:

Allozyme Analysis

Theoretical Population Genetic Modelling

### **Research Interests:**

Speciation

### Types of Research Projects.

Below are examples of the type of projects carried out by our group at Rhodes University which is made up of the Department of Biochemistry and Microbiology and JLB Smith Institute of Ichthyology with strong collaboration with the Department of Zoology and Entomology, Rhodes University, the Department of Botany, Rhodes University, Cape Nature Conservation, Transvaal Nature Conservation, the Port Elizabeth Museum and a number of departments from other Universities and other interested bodies and companies. The overall theme of the research is measurement of genetic diversity in important species and the application of this to sustainable utilisation of the genetic resources of southern Africa

### Genetic Characterisation of Small Isolated Freshwater Fish Populations.

Southern Africa contains a number of habitats which because of geographic geological and climatological factors contain small isolate.' bodies of fresh water. The limited number of large fresh water systems consist of a few major river systems such as the Vaal, the Orange and the Tugela and number of man made lakes which are the result of the construction of dams. The majority of natural water bodies at the coast consist of a large number of small river systems, while inland small lakes, springs and sinkholes predominate in many areas. The relative scarcity of water supplies through the southern African region creates large scale pressure on these resources. Many of the water bodies are unique, isolated and have not been studied in detail.

One such system is the dolomitic springs and sinkholes of the western Transvaal. The diverse and competing aspects of utilisation and exploitation of these springs and sinkholes pose a major threat to the unique biota inhabiting these water bodies. A multi-disciplinary

project aimed at the identification and characterisation of the ichthyofauna of six selected study sites in the western transvaal was initiated by the Department of Nature and Enviromental Conservation, Transvaal and set up in collaboration with the JLB Smith Institute of Ichthyology, Grahamstown. Genetic characterisation of the unique members of the ichthyofauna of these sites, Molopo Oog, Wondergat, Malmani, Marico, Schoonspruit and Klerkskraal was carried out and specific recommendations on the conservation of these species was made and on the sustainable utilisation of these unique water resouces for farming and recreational purposes.

### Genetic Characterisation of Known Threatened and Rare Fish Populations.

*Barbus andrewi* is a fish species found in a limited number of rivers in the Western Cape of South Africa. It is particularly threatened for two reasons: firstly that attempts to eradicate the species to improve fishing occurred in the past; and secondly from competition with introduced northern hemisphere fish species. The species is rare enough to make sampling difficult and fin clips were used to obtain tissue samples to avoid destructive sampling of the populations. Support from the Department of Nature and Environmental Conservation, Cape Province, allowed the study of two natural populations in Berg River and the Buffelsjagsdam/Breede River. A captive population bred at the Amalinda Fish Hatchery was also studied with the aim of developing a reintroduction programme. We have used RAPDS fingerprinting to make recommendations on the best approach to the conservation of this species using captive breeding and re-introduction.

### Genetic Variation within and between rare and common taxa of Cape Proteaceae.

The Fynbos biome is unique and contains a complete floral kingdom part of which is made up of many taxa of Proteaceae which are not found any where else in the world. These range from populations of plants in the 100,000 to millions to ones which contain less than 50 individuals. These species, not withstanding their unique place in the worlds flower population represent a economic resource for the Cape.

Harvesting of these flowers from the wild provides employment for a significant number of people and the eco-tourist potential of the floral kingdom is only just beginning to be exploited. An initial screening of a number of threatened species by molecular genetics is underway to measure both their genetic diversity and gene pool of these species and to elucidate some of the taxonomic problems associated with very rare plants. The very small gene pool that some of these species may have poses a particular threat especially as rationalisation of Nature Conservation resources may need to de-designation of nature reserves. Preservation of the plants by propagation outside of the wild would require knowledge of the genetic makeup of the plants taken from the wild to maximise the gene pool and could be used to encourage the move to commercial protea farming rather than exploitation of wild plant populations. We are using protein profiling and RAPDS fingerprinting to confirm species and sub-species status for rare proteas and to identify viable populations for preservation and further study. Recent results have shown us that RAPDS fingerprints can be obtained from preserved herbanium material which opens up the possibility that changes in the genetic diversity of these rare species can now be studied over time.

### Population Genetics of Two Dolphin Species from the East Coast of Southern Africa.

Two species of dolphin are found commonly of the shores of southern Africa, the bottlenose dolphin and the humpback dolphin. Both are under threat from a variety of human agencies including pollution, overfishing and the using of shark nets to protect swimmers. Although not confined to the southern African coast, they are an important of the coastal ecosystem as well as being an ecotourist attraction. We are trying to answer a two questions concerning these species. Firstly, what is the genetic diversity of the populations along the southern African coast and are these populations unique compared to the rest of the world.

Secondly, as the degree of threat to the dolphin populations varies along the coast from being high close to Durban in Natal/Kwazulu to relatively low off the coast further south, do the dolphins form one continuous interbreeding population or is there only relatively few individuals much move from one distinct population to another. Finally, can we discover something about the breeding and social structure of dolphins. We are using mitochondrial DNA studies, RAPDS fingerprinting and possibly, in the future, microsatellite analysis to answer these questions and make recommendations on how best to stabilise the populations of these two dolphin population and retain their ability to attract ecotourists.

### Identification of Microbial Population Diversity in Enviromental Samples.

Since the discovery of antibiotics during the 1940's, pharmaceutical companies have collected samples from a massive number of different environments in almost every country of the world and screened these samples for the presence of organisms that make new antibiotics. Almost all the groups of antibiotics in use today were discovered this way and in almost every case the country of origin of the organism is lost in obscurity unless preserved in the species name such as *Streptomyces natalensis*. More important, the country of origin which in most cases was not the country of the company which exploited the discovery, received not monetary reward for the exploitation of its natural resource. The United Nations Treaty on the Environment has changed this to some degree but it is up to each country to protect its own resources. We have developed a RAPDS tingerprinting based method for studying the genetic diversity within the actinomycetes found in soil samples. Thus unique organisms can be identified against a database for further study if necessary by commercial companies and the organisms can be protected from unwanted exploitation. by those not permitted so to do.

### Conclusions

Conservation and sustainable development require that genetic diversity be identified, measured, analysed over time, exploited where possible and protected from threats where possible. Modern methods of studying genetic diversity at the organism are becoming increasingly complex but lack two important features. Firstly, this approach does not usually find a vaste range of new resouces it is an extention of the classical approach to species identification although it is very good as an overall measurement of totally resources available. Secondly, it tells the observer almost nothing about the genetic structure of the population and does not have a direct predictive ability on how well the population will survive. Only molecular genetic tools are now able, to some extent, to make such predictions, identify easily new cryptic species, analyse what happens to a population over time, and protect any novel organisms identified from unauthorised exploitation.

Of the range of molecular genetic tools available to study population genetics in an African context, three have decided advantages of the rest in ease of application in a difficult enviroment, cost and large scale sample analysis. These are SDS acrylamide gel electrophoresis of proteins for rapid analysis of population diversity, RAPDS fingerprinting for detailed analysis of diversity between and within population, sub-species and species, and microsatellite analysis if detailed analysis and heterozygosity measurements are required of specific species in a long term study. Mitochondrial analysis using polymerase chain reaction also has its place in studies of the systematics and taxonomy of species especially as it can be easily applied to museum specimens.

South Africa is not unique in Africa in having the expertise to use molecular population genetics but it is a resource centre which is already being exploited in Namibia, Botswana, Malawi and Zimbabwe. The research and training resources in South Africa should be used to the advantage of the whole of Africa.