### **Attachment 3**

# Strzelecki/ South Gippsland Koala

An investigation carried out by Dr Bronwyn Houlden, School of Biological Science, University of New South Wales, 20th March 1997 and 6th April 1998 confirmed that the genetic pool of South Gippsland koalas had not been compromised. Dr Houlden indicated that on a national basis koalas generally are not considered to be threatened. She advised that this assessment has unfortunately led to an extremely simplistic view of conservation of biodiversity in the species.

Her report was entitled "Low genetic variability of the koala Phascolarctos cinereus in south-eastern Australia following a severe population bottleneck" - Published in Molecular Ecology 1996, 5 269-281. Through extensive analysis by herself and her collaborators Houlden revealed that the species is composed of highly differentiated populations with low levels of gene flow between populations throughout their range. The Strzelecki Koala population constitutes a separate management unit and is significant in terms of management of biodiversity on a regional and state basis. Dr Houlden found that the Strzelecki Ranges had the highest level of genetic variation, of any Victorian population she analysed. This is important, given the low levels of genetic variability found in many populations in Victoria, which have been involved in the translocation program.

"The Strzelecki koala population has high levels of genetic variability which have been detected by rare and unique genetic markers. These animals are statistically significantly differentiated from other Australian populations and therefore constitute a separate management unit. Because biodiversity in the species as a whole is dependent of conservation of populations throughout the species range, the Strzelecki Ranges population, together with the South Gippsland population is nationally significant as well. The lack of genetic diversity amongst Australian koalas could be critical to the survival of the species as a whole."

Attachment 3
Extracts from
Inbreeding and testicular abnormalities in a bottlenecked population of koalas (*Phascolarctos cinereus*) Critescu et al 2009

**Abstract.** Habitat destruction and fragmentation, interactions with introduced species or the relocation of animals to form new populations for conservation purposes may result in a multiplication of population bottlenecks. Examples are the translocations of koalas to French Island and its derivative Kangaroo Island population, with both populations established asinsurance policies against koala extinction. In terms of population size, these conservation programs were success stories.

However, the genetic story could be different. We conducted a genetic investigation of French and Kangaroo Island koalas by using 15 microsatellite markers, 11 of which are described here for the first time. The results confirm very low genetic diversity. French Island koalas have 3.8 alleles per locus and Kangaroo Island koalas 2.4. The present study found a 19% incidence of testicular abnormality in Kangaroo Island animals. Internal relatedness, and individual inbreeding coefcient, was not signicantly different in koalas with testicular abnormalities from that in other males, suggesting the condition is not related to recent inbreeding. It could instead result from an unfortunate selection of founder individuals carrying alleles for testicular abnormalities, followed by a subsequent increase in these alleles' frequencies through genetic drift and small population-related inefciency of selection. Given the low diversity and possible high prevalence of deleterious alleles, the genetic viability of the population remains uncertain, despite its exponential growth so far. This stands as a warning to other introductions for conservation reasons.

#### Introduction

Past and present threats to wildlife, from habitat destruction and fragmentation to intensive exploitation and competition or predation from introduced species, produce a considerable number of populations that have or will endure dramatic decline. A small population size is a risky situation because environmental and demographic stochasticity threaten population viability. But the genetic consequences of a reduced size may be as important, and continue to affect a population even after it recovers in size (Sherwin and Murray 1989; Bijlsma et al. 2000). Since European settlement in Australia, the koala's (*Phascolarctos cinereus*) distribution has been dramatically reduced. By 1920, the cumulative effect of the destruction of Eucalyptus forest, intensive fur trade, diseases and fires made the survival of the species uncertain (Strahan and Martin 1982). Koalas became extinct in SouthAustralia, and the Victorian population declined to less than a thousand individuals (Lee et al. 1988). The deliberate introduction of koalas to some southern islands is thought to be the first conservation program involving marsupials. The French Island (Victoria) population is said to have come from as few as two or three animals at the end of the 19th century (Taylor et al. 1997; Martin and Handasyde 1999). The favourable environment and lack of predators ensured the growth of the population. French Island (FI) became a source for a translocation program to repopulate the Victorian mainland as well as to colonise several islands. In all, 18 koalas (and an unknown number of dependent young or embryos) were taken from FI and introduced to Kangaroo Island (KI, South Australia) in 1923–25 (Martin and Handasyde 1999). Koalas are now considered pests on KI, with population estimates as high as 27 000 in Masters et al. (2004). In terms of population size, the success of these conservation programs is obvious. However, at a genetic level they are questionable. Indeed, a small founding population size is often accompanied by a loss of genetic variability and an increase in the frequency of deleterious genes, a process usually referred to as a bottleneck effect (Frankham 1998). The production of offspring derived from only a few animals can also cause inbreeding, and thus an increase of homozygosity (Falconer 1960)

#### **CSIRO** PUBLISHING

www.publish.csiro.au/journals/wr *Wildlife Research*, 2009, **36**, 299–308 CSIRO 2009 10.1071/WR08010 1035-3712/09/040299

#### Attachment 3

High effective inbreeding coefficients correlate with morphological abnormalities in populations of South Australian Koalas (*Phascolarctos cinerus*). Seymour, Montgomery, Costello, Ihle, Johnson, St. John Taggart, Houlden.

Animal Conservation (2001) 4, 211-219 2001 The Zoological Society of London accepted 5 Feb. 2001

#### **Abstract**

Koalas have undergone a series of sequential founding events on islands in SE Australia in recent times. Populations in South Australia at the Eyre Peninsula and Mt. Lofty Ranges were founded in the 1960's from a colony on Kangaroo Island. The Kangaroo Island colony was derived from animals introduced to French Island from mainland Victoria over a century ago.

In this study, we first use microsatellite markers to quantify levels of genetic variation within South Australian koala populations and the relatively undisturbed Strzelecki Ranges population from mainland Victoria. This analysis revealed low levels of allelic diversity and heterozygosity in the 3 South Australian koala populations relative to the Strzelecki Ranges population which has the highest levels of allelic diversity and heterozygosity in Victoria. Second, we measured the incidence of testicular aplasia, a unilateral or bilateral failure in testicular development in the Eyre Peninsula and Kangaroo Island populations and in the ultimate founding population at French Island. Testicular aplasia was present at a frequency of ... in the Eyre Peninsular and Kangaroo Island populations and in the Eyre Peninsular, but was undetectable in the non-bottlenecked Pilliga State Forest population in NSW......

Therefore we quantified differentiation between the 3 South Australian populations and the Strzelecki Ranges and French Island population.....

### Discussion

We revealed that levels of allelic diversity are significantly lower in the Kangaroo Island and Eyre Peninsular population relative to the Strzelecki population in Victoria, as was expected from their population history

In Victoria and South Australia Koalas are considered to be secure, based on

...... In Victoria and South Australia Koalas are considered to be secure, based on population numbers.

### **Attachment 3**

# Strzelecki/Sth. Gippsland DNA

# **Extracts from Emmins PHD Thesis 1996**

According to John Emmins in his 1996 (PhD) Thesis entitled; "The Victorian Koala (manuscript): Genetic Heterogeneity, Immune Responsiveness & Epizootiology of Charydiosis;

"It is immediately obvious in South Gippsland DNA profiles that there is great genetic variability in this population... The table of D values ... and the statistical analysis of the D values derived from comparisons within that population show a mean D value of 0.48 which indicates that the South Gippsland koala population is more outbred than any other study population. This was an exciting finding and indicates the importance of this population as a rich gene pool for Victorian koalas." Emmins, John Jeffrey (The Victorian Koala (manuscript): Genetic Heterogeneity, Immune Responsiveness & Epizootiology of Chlarydiosis 1996 Thesis (PhD)-Monash University.

p285"... The most important conclusions to be drawn from the genetic study are:

- 1 French Island koalas are an inbred population, and their lack of genetic diversity is consistent with them having gone through a severe genetic bottleneck implicit with the colony having been established from only a few animals approximately 100 years ago.
- 2 The colonies (located throughout South Eastern Australia) which have been set up largely from French Island descendants during the 70 years of translocations also have limited genetic diversity. Their gene pool may be somewhat larger however due to being additional translocations of koalas from Phillip Island which presumably did not go through as severe a bottleneck as French Island animals, (or at least were derived from different individuals). The exception to this are populations set up from French Island animals without additions from Phillip Island or elsewhere. Kangaroo Island (S.A.) demonstrates very clearly with the resident koala population appearing as inbred as laboratory rats with the only detectible genetic differences in the population being attributable to sex differences. (Shimmin et al, 1995 showed the mean D-value of laboratory rat strains to be very close to 1.0)
- 3 The Victorian koala is genetically quite different from New South Wales and Queensland koalas.
- 4 The Gold Coast koalas are as inbred as French Island koalas and therefore may also be derived from a limited number of individuals.
- 5 The South Gippsland koala population is the most outbred and genetically diverse of all the populations studied".
- p286 "These findings have very significant repercussions in the management of the Victorian koala. They also show that the relic South Gippsland koala population is of great genetic importance and that this population (and its habitat) should be conserved at all costs to maintain a large gene pool. The genetic make-up of the remainder of Victoria's koalas is lacking in this diversity and needs to be addressed". Emmins, John Jeffrey (The Victorian Koala (manuscript): Genetic Heterogeneity, Immune Responsiveness & Epizootiology of Chlarydiosis 1996 Thesis (PhD)-Monash University

