GENETIC LINKAGE MAPS AND POPULATION GENETICS OF MACROPODS

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GLOSSARY

Ascertainment bias – The hypothesis that a microsatellite selected in a focal species will differ systematically from its orthologues in related species due to the criteria used to isolate it in the focal species.

Cross species amplification – Primer binding sites sufficiently conserved in a related species allows amplification using primers designed in a different species.

Effective population size – The number of individuals that actually reproduce and contribute genes to the next generations.

Genic analysis – allele and / or haplotype frequencies are population statistics that can be changed by genetic drift, founder effect, gene flow and selection. They are estimated most accurately from multiple, separate, nuclear loci and mtDNA.

Genotypic analysis – consist of composite genotypes of multiple loci. Individual genotypes are labile – a single round of sexual recombination usually destroys a genotype. They are quantified most rigorously using multiple single-locus nuclear markers (typically microsatellites).

Gene synteny – Where loci are located on the same chromosome.

Homoplasy – Similarities in character states for reasons other than inheritance from a common ancestor. At a microsatellite locus, homoplasy occurs when two allelic lineages converge to the same size but have different histories of mutations. Thus, identity by state does not always entail identity by descent.

Likelihood ratio – The ratio of probabilities of obtaining the observed data under different hypotheses concerning the assumed model used to generate the expected values.

Linkage disequilibrium – Departure from the predicted frequencies of multiple locus gamete types assuming alleles of different loci are randomly associated.

Meiotic drive – Aberrant segregation ratios among the gametes of heterozygotes.

Monophyletic group – Set of species containing common ancestor and all its descendants.

Nucleolar organiser region – A region on a chromosome that contains the ribosomal RNA genes and associated spacers.

Null allele – An allele that fails to be expressed under the conditions analysed.

Orthologous loci – Loci in two or more species where sequences are similar because of their common derivation from a common ancestor.

Paraphyletic group – Set of species containing an ancestral species together with some, but not all, of its descendants. The species included in the group are those that have continued to resemble the ancestor; the excluded species have evolved rapidly and no longer resemble their ancestor.

Parsimony – Principle of phylogenetic reconstruction in which the phylogeny of a group of species is inferred to be the branching pattern requiring the smallest number of evolutionary changes.

Polyphyletic group – Set of species descended from more than one common ancestor. The ultimate common ancestor of all the species in the group is not a member of the polyphyletic group.

$\Delta T_{mH}$ – The difference between the median melting temperature of homoduplex DNA and heteroduplex DNA formed in a DNA-DNA hybridisation reaction.

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<td>androgen receptor</td>
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<tr>
<td>CASA</td>
<td>alpha - casein</td>
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<td>cDNA</td>
<td>complementary DNA</td>
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<td>cM</td>
<td>centimorgan</td>
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<td>DBB</td>
<td>MHC class II B-chain</td>
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<td>ESUs</td>
<td>evolutionary significant units</td>
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<td>ETL</td>
<td>economic trait locus</td>
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<td>fluorescence in situ hybridisation</td>
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<td>glucose-6-phosphate dehydrogenase</td>
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<td>haemoglobin beta chain</td>
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<td>$H_E$</td>
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<td>LLP</td>
<td>late lactation protein</td>
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<td>lipoprotein lipase</td>
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<td>MFL</td>
<td>mean fragment length</td>
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<td>MHC</td>
<td>multiple histocompatibility complex</td>
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<td>MP</td>
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<td>MUs</td>
<td>management units</td>
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<td>Mya</td>
<td>million years ago</td>
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<td>$N_e$</td>
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<td>TF</td>
<td>transferrin</td>
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<tr>
<td>tRNA</td>
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<td>VIC</td>
<td>Victoria</td>
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DECLARATION

I declare that this submission is my own work and that, to the best of my knowledge it contains no material written by another persons nor material which has been submitted for a higher degree to this or any other institution, except where due acknowledgement has been made in the text.

Kyall R. Zenger

November 2001
The analysis of DNA using molecular techniques is an important tool for studies of evolutionary relationships, population genetics and genome organisation. The use of molecular markers within marsupials is primarily limited by their availability and success of amplification. Within this study, 77 macropodid type II microsatellite loci and two type I genetic markers were characterised within *M. eugenii* to evaluate polymorphic levels and cross-species amplification artifacts. Results indicated that 65 microsatellite loci amplified a single locus in *M. eugenii* with 44 exhibiting high levels of variability. The success of cross-species amplification of microsatellite loci was inversely proportional to the evolutionary distance between the macropod species. It is revealed that the majority of species within the Macropodidae are capable of using many of the available heterologous microsatellites. When comparing the degree of variability between source-species and *M. eugenii*, most were significantly higher within source species (*P < 0.05*). These differences were most likely caused by ascertainment bias in microsatellite selection for both length and purity.

The production of a marsupial genetic linkage map is perhaps one of the most important objectives in marsupial research. This study used a total of 353 informative meioses and 64 genetic markers to construct a framework genetic linkage map for *M. eugenii*. Nearly all markers (93.7%) formed a significant linkage (LOD > 3.0) with at least one other marker. More than 70% (828 cM) of the genome had been mapped when compared with chiasmata data. Nine linkage groups were identified, with all but one (LG7; X-linked) allocated to the autosomes. These groups ranged in size from 15.7 cM to 176.5 cM, and have an average distance of 16.2 cM between adjacent markers. Of the autosomal linkage groups, LG2 and LG3 were assigned to chromosome 1 and LG4 localised to chromosome 3 based on physical localisation of genes. Significant sex-specific distortions towards reduced female recombination rates were revealed in 22% of comparisons. Positive interference was observed within all the linkage groups analysed. When comparing the X-chromosome data to closely related species it is apparent that it is conserved both in synteny and gene order.

The investigation of population dynamics of eastern grey kangaroos has been limited to a few ecological studies. The present investigation provides analysis of mtDNA and microsatellite data to infer both historical and contemporary patterns of population structuring and dispersal. The average level of genetic variation across sample locations was exceedingly
high \( h = 0.95, H_E = 0.82 \), and is one of the highest observed for marsupials. Contrary to ecological studies, both genic and genotypic analyses reveal weak genetic structure of populations where high levels of dispersal may be inferred up to 230 km. The movement of individuals was predominantly male-biased (average \( N_{m} = 22.61 \), average \( N_{m} = 2.73 \)). However, neither sex showed significant isolation by distance. On a continental scale, there was strong genetic differentiation and phylogeographic distinction between southern (TAS, VIC and NSW) and northern (QLD) Australian populations, indicating a current and/or historical restriction of geneflow. In addition, it is evident that northern populations are historically more recent, and were derived from a small number of southern eastern grey kangaroo founders. Phylogenetic comparisons between \( M. g. giganteus \) and \( M. g. tasmaniensis \), indicated that the current taxonomic status of these subspecies should be revised as there was a lack of genetic differentiation between the populations sampled.