

The Koala and its Retroviruses: Implications for Sustainability and Survival

edited by

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The Koala Genome Consortium

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ABSTRACT. The koala (*Phascolarctos cinereus*) is an iconic Australian animal. Koalas are both biologically unique and evolutionarily distinct as the only living representative of the marsupial family Phascolarctidae. Their unique and highly specific diet of eucalyptus leaves, combined with the increasing threats of predation and habitat loss through urbanisation, mean that koalas are particularly vulnerable to the deleterious effects of fragmented habitat and population bottlenecks. They are further threatened by disease such as *Chlamydia* and there is increasing interest in the varying strains of the Koala Retrovirus. We present preliminary transcriptome and genome data for the koala and introduce the Koala Genome Consortium (KGC), a group working towards the production of a high quality draft assembly of the koala genome. The KGC is currently comprised of several Australian research institutes and Universities although our intention is to recruit researchers from around the world to contribute to the genome assembly and annotation process and ultimately make use of the assembled genome. Once available as an annotated draft, we anticipate the genome sequence will add significant value to the extensive body of existing research for koalas.

JOHNSON, REBECCA N., MATTHEW HOBBS, MARK D. B. ELDRIDGE, ANDREW G. KING, DONALD J. COLGAN, MARC R. WILKINS, ZHILIANG CHEN, PETER J. PRENTIS, ANA PAVASOVIC, ADAM POLKINGHORNE AND PETER TIMMS. 2014. The Koala Genome Consortium. In *The Koala and its Retroviruses: Implications for Sustainability and Survival*, ed. Geoffrey W. Pye, Rebecca N. Johnson and Alex D. Greenwood. *Technical Reports of the Australian Museum, Online* 24: 91–92.

Overview of the Koala Genome Consortium

The purpose of the Koala Genome Consortium is to generate a high quality draft assembly of the koala genome, which will be useful to all koala researchers and have real and measurable conservation outcomes for koalas. Prior to the sequencing of the koala genome, we also undertook to determine the transcriptome from several tissues from two

separate koalas. The Koala Genome Consortium is a project co-led by the Australian Museum and Queensland University of Technology but since this is a large undertaking, demanding the expertise of a wide group of researchers in the fields of koala biology, bioinformatics and marsupial genomics, we are currently seeking expressions of interest from potential new Consortium members and will continue to do so over the course of the project.

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Koala genome

We obtained tissue samples from a female koala named Pacific Chocolate from Port Macquarie Koala Hospital in New South Wales, and high molecular weight gDNA extractions from liver tissue were used to prepare libraries suitable for massively parallel sequencing using the Illumina platform. Our initial genomic sequencing datasets represent approximately 100-fold coverage of the genome with the large task ahead to assemble and annotate the genome. It is our intention that the formation of the Koala Genome Consortium (www.koalagenome.org) will facilitate new collaborations between all groups with a scientific interest in the koala genome data, thereby producing a comprehensive and well annotated assembly which will benefit the already substantial efforts dedicated to koala research globally.

Koala transcriptome

We have generated koala transcriptome data from eight different tissues from two separate koalas, (a) Pacific Chocolate, from the Port Macquarie region of NSW, and (b) Birke, from the South East region of Queensland. We chose multiple tissues to allow preliminary comparison between gene expression in different koala tissue types (i.e. brain, heart, lung, liver etc). Our transcripts are now fully assembled and are undergoing annotation. We have sequenced our transcripts to a very high level of coverage, estimated to be $\times 100$ fold, which we anticipate will enable us to detect genes with low expression levels.

Why is the Australian Museum so interested in a genome sequencing project?

Our own research focus is largely around population and evolutionary genetics, as well as utilizing our historic natural history collections. The Australian Museum is not only the oldest museum in Australia but is one of the oldest in the world, and has extensive natural history collections of Australia's most iconic animals dating from present day back to the mid 1850's. Museum collections have already been used to give insight into historic KoRV infections and give insight into historic levels of mitochondrial diversity (Ávila-Arcos *et al.*, 2012; Tsangaras *et al.*, 2012). Further, the Australian Museum is periodically approached for population management advice based on population genetic data, including for NSW koalas for which we have developed additional microsatellite markers using next generation sequencing and the KGC data. One of our priority research outcomes is to develop and implement a suite of highly variable SNP markers that can be used for direct management outcomes in addition to the microsatellite data.

The Australian Museum team is especially interested in using the genome and transcriptome data to understand the koala's unique physiological adaptations and in particular, it may be possible to predict response to environmental change to develop strategies to mitigate damage to koala populations in the longer term.

The koala genome and koala retrovirus (KoRV)

With a high quality, deep coverage whole genome and transcriptome assembly from two animals we can ascertain how and where the different strains of KoRV have been endogenized in the koala. By comparing multiple tissue types and two animals initially we will be able to determine if two animals share insertion sites, or if they are more likely to be individually variable. This represents a significant advance in what is currently known about KoRV and will be of benefit to the wider research community.

Get involved in the project via Koalagenome.org

We have established www.koalagenome.org as a focal communication, data-sharing and data-storage point for the Koala Genome Consortium. Please contact us at this site or via our email address: koalagenome@austmus.gov.au if you have an interest in being involved with the project.

ACKNOWLEDGMENTS. We wish to acknowledge the funding of Bioplatforms Australia and the Australian Museum Foundation, as well as the ARC Linkage Scheme and the Queensland Government NIRAP Scheme. MRW and ZC acknowledge support from the Australian Government EIF Super Science Scheme and the NSW State Government Science Leveraging Fund.

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