1. Lead CI Name: Jaime Gongora

2. Project Title: Genomic Insights into Adaptive Diversity: A Comprehensive Analysis of Platypus, Arabian Oryx, and Crocodilian Species

3. Project Description:

This project will leverage the genomic resources generated in our previous successful Adapter project (2022Q4; project code te48). This project will apply the protocols to identify loci under selection developed in our previous Adapter project. These protocols will be used for whole genome sequencing datasets of various crocodile species, Arabian oryx and platypus.

We will extend our protocols by including new analyses to validate whether loci identified as being putatively under natural selection using a two-criteria approach (i.e. confirmation with at least two measures of genetic differentiation such as GST, Jost's D, mutual information and Adjusted AFD; Sherwin, 2022).

We will analyse whole genome sequencing data from four different datasets. The first dataset consists of 26 platypus samples that were collected in the unregulated Tenterfield Creek (n=11) and below the dam (n=8), and above the dam (n=7) in the regulated Severn River (J. Mijangos et al., 2022). The second dataset consists of 57 platypus samples from the whole species range in eastern mainland Australia and Tasmania (Martin et al., 2018). The third dataset focuses on the fourth dataset consists of 71 Arabian oryx samples collected in Oman. The final dataset consists of SNP data from 34 saltwater crocodiles in the Northern Territory.

We will use novel methods to estimate relatedness that corrects for various bias issues from previous methods, particularly in small populations (Wang, 2022).

In the Arabian oryx, we will investigate genes that might contribute to adaptation to desert life (e.g., heat loss, water conservation, and metabolism) and the genomic cost of a near-extinction event.

The data collected via this study will provide valuable insights into the adaptive capacity of these populations and contribute to the development of conservation strategies. The processes devised within this study provide the potential for applicability to alternative populations, with a particular emphasis on small species of conservation species. The genetic resources established in this study will be valuable in guiding reintroduction and captive breeding initiatives.

4. Investigators:

Dr Jaime Gongora is the Associate Dean for Indigenous Strategy in the Faculty of Science at The University of Sydney. Additionally, he serves as an Associate Professor, specialising in conservation biology and animal genetics. Dr Gongora has devoted his professional endeavours to investigating genetic diversity in non-domesticated fauna, alongside examining their population genetics, phylogenetics, and phylogeography.

Relative to opportunity, Dr Mijangos has shown exceptional performance by publishing high-calibre research papers and developing a significant software tool, dartR (Mijangos et al., 2022), which enhances the capabilities of genomics-based research. With a profound understanding of genetic

principles, Dr Mijangos has a distinctive skill set that combines sophisticated bioinformatics and programming capabilities. This unique mix of expertise makes him an exceptional candidate to use genomic wildlife preservation methodologies effectively. Dr Mijangos has extensive knowledge in using computer clusters, including GADI and Artemis, at the University of Sydney.

5. Technical Plan:

We will use the analysis protocols developed for the platypus in our previous Adapter project to explore further the genome adaptive (and neutral) loci diversity in various crocodile whole genome sequencing datasets.

We will use Pretzel (Keeble-Gagnère et al., 2019), an innovative method to visualise several genomes jointly. This program will be instrumental in analysing our dataset's whole genomes containing 18 crocodile species.

Using whole genome sequence data, we will use Inferring relatedness and pedigree of platypuses.

6. Data Plan:

We will utilise the Research Data Store (RDS) available through the University of Sydney, regularly backing up generated data and transferring files no longer used off Gadi. On completion of the quarter, we will transfer all remaining files to RDS.

7. Relationship to other projects:

Preliminary analyses performed in our previous Adapter project identified potential genomic regions under natural selection in genes with metabolic and development functions, including KCNIP4, ADAM10 and OXR1. Our results could indicate that adaption to new environments may have occurred in as short as seven generations.

8. References:

Keeble-Gagnère, G., Isdale, D., Suchecki, R.-ł., Kruger, A., Lomas, K., Carroll, D., Li, S., Whan, A., Hayden, M., & Tibbits, J. (2019). Integrating past, present and future wheat research with Pretzel. *BioRxiv*, 517953.

Martin, H. C., Batty, E. M., Hussin, J., Westall, P., Daish, T., Kolomyjec, S., Piazza, P., Bowden, R., Hawkins, M., & Grant, T. (2018). Insights into platypus population structure and history from whole-genome sequencing. *Molecular Biology and Evolution*, *35*(5), 1238-1252.

Mijangos, J., Bino, G., Hawke, T., Kolomyjec, S., Kingsford, R., Sidhu, H., Grant, T., Day, J., Dias, K., & Gongora, J. (2022). Fragmentation by major dams and implications for the future viability of platypus populations.

Mijangos, J. L., Gruber, B., Berry, O., Pacioni, C., & Georges, A. (2022). dartR v2: an accessible genetic analysis platform for conservation, ecology, and agriculture. *Methods in Ecology and Evolution*.

Sherwin, W. B. (2022). Bray-Curtis (AFD) differentiation in molecular ecology: Forecasting, an adjustment (AA), and comparative performance in selection detection. *Ecology and Evolution*, *12*(9), e9176.

Wang, J. (2022). A joint likelihood estimator of relatedness and allele frequencies from a small sample of individuals. *Methods in Ecology and Evolution*, *13*(11), 2443-2462.