

## SPECIES AND HAPLOTYPE IDENTIFICATION RESULTS REPORT

### Samples

Two beetle specimens were sent to the Ecological Genetics Laboratory at Manaaki Whenua – Landcare Research in Auckland, New Zealand for species and haplotype identification. The samples were received at the laboratory on Thursday 23<sup>rd</sup> May 2019. The following details were included with each specimen:

Island: Efate, Vanuatu. Collection Date: 20/05/19. Collector: L. Howard.

### Project Summary

DNA was extracted from leg tissue of each specimen using readymade reagents from Qiagen® following the manufacturer's instructions. Species and haplotype identification was undertaken by amplification of a highly conserved region of the cytochrome *c* oxidase subunit I (COI) gene using primers LCO1490 and HCO2198 (Folmer *et al.* 1994).

Edited DNA sequences were then compared against sequences from GenBank, administered by The National Center for Biotechnology Information (NCBI). The BLAST (Basic Local Alignment Search Tool) algorithm was used to search for the most closely matching sequences within the NCBI database.

### Results

The resulting DNA sequences both most closely matched the PNG (Papua New Guinea) haplotype (as defined by Marshall *et al.* 2017) of the *Oryctes rhinoceros* (coconut rhinoceros beetle) with a maximum identity of 100%, when compared against nucleotide sequences from GenBank.

### References

Folmer, O., Black, M., Hoeh, W., Lutz, R. & Vrijenhoek, R. (1994). DNA primers for amplification of mitochondrial cytochrome *c* oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, 3(5), 294-299.

Marshall, S. D., Moore, A., Vaqalo, M., Noble, A. & Jackson, T. A. (2017). A new haplotype of the coconut rhinoceros beetle, *Oryctes rhinoceros*, has escaped biological control by *Oryctes rhinoceros* nudivirus and is invading Pacific Islands. *Journal of Invertebrate Pathology*, 149, 127-134.