ISFG Short Term Fellowship 2023

Applicant:
Jessica Watson
PhD Student, University of Technology Sydney (UTS)
15 Broadway, Ultimo, NSW, Australia
Australian Federal Police National DNA Program for Unidentified and Missing Persons
Australian Federal Police Forensic Laboratory, Majura, ACT, Australia

Research collaboration visit to the Armed Forces DNA Identification Laboratory
5th March 2023 to 20th March 2023

The purpose of this visit was to foster a collaboration between the Armed Forces DNA Identification Laboratory (AFDIL) and the Australian Federal Police National DNA Program for Unidentified and Missing Persons (AFP Program). Both organisations are applying emerging forensic genomics techniques using medium- to high-density single nucleotide polymorphism (SNP) panels and whole mitochondrial DNA (mtDNA) sequencing using massive parallel sequencing (MPS) technology. This visit also contributed to the development and advancement of my PhD research.

During this visit, I was able to observe and receive hands-on training in AFDIL's bone sampling, DNA extraction, optimised hybridisation capture and sequencing protocols using the NextSeq 550 System for challenging skeletal samples. Several of these protocols are of interest to the AFP Program due to reported success with low quantity and quality samples encountered in AFDIL casework, including the modified Dabney DNA extraction protocol[1] and hybridisation capture panels including the FORensic Capture Enrichment panel (FORCE)[2], whole mtDNA genome capture[3] and 95K SNP panel[4].

Outside of the laboratory, I was shown their established bioinformatics pipeline using the QIAGEN CLC Genomics Workbench with the AFDIL-QIAGEN mtDNA Expert (AQME)[5] and Parabon® Fx™ Forensic Analysis Platform. The databases in use included the EDNAP mtDNA Population Database (EMPOP) for the determination of Y and mtDNA haplogroups and BLAST® for species identification of suspected non-human samples. The reporting criteria for short-range kinship testing, Y DNA, mtDNA and species identification were also discussed. In total, I was able to learn all the requirements for incorporating these forensic genomics techniques into the AFP Program.

This collaboration also gave me an opportunity to compare and discuss the protocols established by the AFP Program for sample collection, DNA extraction and sequencing using targeted amplicon sequencing kits with those at AFDIL. During the visit, I was able to share the findings of the AFP Program's internal validation of the ForenSeq® Kintelligence Kit for kinship analysis, of which AFDIL is interested in testing to compare to the FORCE panel. I also gave a presentation to AFDIL on the establishment, purpose and achievements of the AFP Program and the validation and operationalisation of both the ForenSeq® Kintelligence Kit and ForenSeq® Whole mtDNA Genome Kit at the AFP.

The visit was invaluable for the development of AFDIL and the AFP Program's relationship for the development and validation of SNP sequencing and analysis pipelines for human remains identification as well as future collaborative research projects.

[1] Dabney, J. & Meyer, M (2019). Extraction of highly degraded DNA from ancient bones and teeth. *Ancient DNA: Methods and Protocols*, 25-29.

[2] Tillmar, A., Sturk-Andreaggi, K., Daniels-Higginbotham, J., Thomas, J.T. & Marshall, C. (2021). The FORCE Panel: An all-in-one SNP marker set for confirming investigative genetic genealogy leads and for general forensic applications. *Genes*, 12(12), 1968.

[3] Ring, J. D., Sturk-Andreaggi, K., Peck, M. A. & Marshall, C. (2017). A performance evaluation of Nextera XT and KAPA HyperPlus for rapid Illumina library preparation of long-range mitogenome amplicons. *Forensic Science International: Genetics*, 29, 174-180.

[4] Gorden, E. M., Greytak, E. M., Sturk-Andreaggi, K., Cady, J., McMahon, T. P., Armentrout, S. & Marshall, C. (2022). Extended kinship analysis of historical remains using SNP capture. *Forensic Science International: Genetics*, 57, 102636.

[5] Sturk-Andreaggi, K., Peck, M. A., Boysen, C., Dekker, P., McMahon, T. P. & Marshall, C. K. (2017). AQME: A forensic mitochondrial DNA analysis tool for next-generation sequencing data. *Forensic Science International: Genetics*, 31, 189-197.