

## THE APPLICATION OF DNA AGE PREDICTION TO THE PRACTICE OF FORENSIC SCIENCE

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The ability to accurately predict the age of individuals or samples obtained in the field may be critical to law enforcement. Treating a suspect as either a legal minor or an adult can directly impact criminal proceedings and the proper application of the law, as occurs during investigations of individuals where no reliable birth records exist. Similarly, being able to accurately assess the age of samples taken from a crime scene may be able to help construct a coherent timeline of events during an investigation. However, many of the current tools used to estimate age are limited by relatively high starting sample requirements, lack sufficient accuracy or precision, or require expensive equipment to carry out the analysis.

Prediction based on DNA methylation epigenetic biomarker analysis is currently the most accurate measurement tool to quantify aging. We recently developed a robust, flexible, and low cost NGS-based targeted bisulfite sequencing panel to analyze DNA methylation profiles at genes highly informative of aging. We demonstrate the use of our method to predict the age of samples in two different law enforcement investigations. In the first, we found that the epigenetic age of a refugee and asylum seeker in Germany, who arrived without any official identification or documentation, predicted the individual to be a legal adult despite his self-reported age as being that of a minor (Mean age,  $\bar{x}$  = 27.7 years; Standard Deviation,  $s$  = 1.2 years; Confidence Interval, CI 95% =  $\pm 1.3$  years;  $p$ -value < 0.0005). In a second case, we applied epigenetic age prediction to samples obtained from a mock crime scene where blood extracted from volunteers (ages between 22-80 years old) was deposited onto 8 different locations of an automobile. We were able to isolate a minimum of 50 ng of DNA from 9 samples and move forward with data generation. DNA methylation biomarker determination demonstrated strong positive correlation for the known age versus the predicted age estimated from the samples ( $r$  = 0.8). Furthermore, replicate measurements showed a relatively small technical error, or difference in predicted age vs. actual age, of just  $\pm 1.2$  years. Our experience indicates that the use of DNA methylation analysis will become a valuable tool to aid law enforcement in cases where the provenance of a sample's or suspect's age is in question or otherwise critical to the outcome of an investigation.