Abstract 1

DETERMINING THE PHYSICAL CHARACTERISTICS OF AN INDIVIDUAL FROM BLOODSTAINS: BIOLOGICAL AGE DETERMINATION

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It is now routine for the forensic scientist to obtain the genetic profile of an individual from DNA recovered from a biological stain deposited at a crime scene. Potential contributors of the stain must either be known to investigators (i.e. a developed suspect) or the questioned profile must be searched against a database of DNA profiles. However, in those instances where there is no developed suspect as yet or there is no match with any database sample, the DNA profile *per se* presently provides no meaningful information to investigators, with the notable exception of gender determination. To aid in these investigations a useful biometric that could provide important probative information is the biological age of an individual. The ability to provide investigators with information as to whether a DNA donor is a newborn baby, an adolescent teenager or an elderly individual could be useful in certain cases, particularly those involving young children such as kidnapping. Currently no reliable validated molecular tests are available for age determination. Two approaches have been evaluated for their ability to determine biomarkers associated with biological age; messenger RNA profiling and telomere length analysis.

The developmental process of ageing is based on the theory that each stage of the human lifecycle (birth through death) is characterized by molecular based biological changes, each requiring genes to be expressed or silenced indicative of that particular stage of life. Candidate genes for biological age determination were obtained by literature searches, as well as, differential messenger RNA expression analysis using the Affymetrix GeneChip. Using an RT-PCR based platform, 319 potential candidate genes were tested, in order to determine if their expression patterns were restricted to a particular developmental stage. This initial screen revealed 23 candidate genes which showed a differential expression pattern between age groups. These candidates namely, AGGF1, ASL, CDC2, YOUNGER1A2, FLJ20344a, NEWBORN1, HBG1n1, HBG2n3, OLD3, LOC151194, LOH11CR2A, MAD1L1, PDCD6, POLM, POLQ, PPARD, PPOX, PRL, SPTRX-1, SPTRX-2, SRC, TBC1, and TEKT2 were next transferred to a quantitative real-time PCR platform and combined with the ribosomal housekeeping gene, S15, into optimized duplex reactions. Results obtained from the YOUNGER1A2, NEWBORN1, HBG1n1, HBG2n3 duplex reactions illustrated they are expressed at elevated levels in newborn individuals (<4-months in biological age), while results from the OLD3 duplex reaction indicated that its expression begins to be up-regulated around puberty and is abundantly expressed in elderly (>65-years) individuals.

The degenerative process of ageing is based on theories which identify increases and decreases in specific biomarkers with increasing age, specifically the shortening of telomeric regions on the ends of chromosomes. Two methods were utilized to determine if telomere length correlated with increasing age, real-time PCR and STELA analysis. For real-time PCR, a delta cycle threshold (dCt) metric was evaluated, in addition to determining the telomere

amplification rate in differently aged individuals, whereas STELA analysis determines individual chromosome telomere lengths. Our results indicate that in blood, there is no correlation between telomere lengths and the biological age of an individual.