## INCREASED POWER OF DISCRIMINATION FROM GENETICALLY VARIANT PEPTIDES IN HUMAN HAIR: REACHING THE "ONE-IN-A-MILLION" THRESHOLD.

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If DNA is not present in biological samples collected as evidence, the options for the forensic investigator are limited. Protein is considerably more stable and abundant than DNA, by many orders of magnitude. Genetic variation found in protein encoding genes, in the form of non-synonymous single nucleotide polymorphisms (nsSNPs), result in changes in protein amino acid sequence. Identifying and detecting these single amino acid polymorphisms in proteins therefore allows genetic content of a subject's DNA to be inferred; allowing peptides to be a surrogate for absent or unusable DNA.

Genetically variant peptides (GVPs) were identified and characterized from 43 genetic nsSNP loci that are expressed in human hair shafts. When the product rule is applied these GVP profiles result in powers of discrimination up to 1 in 5.4 million in the European population. When using allelic frequencies from the African population, the GVP profiles were considerably less common, by a factor of up to 29,000. The increased power of discrimination is a direct result of using the current generation of proteomic mass spectrometer, a Thermo Q-Exactive plus hybrid orbitrap / linear ion trap mass spectrometer. Use of this instrument results in an increase of discrimination by 2.7  $\pm$  1.7 orders of magnitude, as compared to previous data generated on an Agilent 6530 Q-ToF instrument. These numbers have been obtained by applying the equivalent of less than 2 mm of hair shaft.

Expansion of these methodologies beyond hair proteins to include alternative tissue types such as teeth, bone, and skin cells is critical to expanding the scope and application space for this novel approach to human forensics.

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