



B15 Protein-Based Human Identification Using Hair Shafts From Different Body Sites

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After attending this presentation, attendees will better understand a new protein-based approach for analyzing human hair evidence that allows inference of Single Nucleotide Polymorphism (SNP) variation in DNA and is both statistically and scientifically rigorous.

This presentation will impact the forensic science community by informing attendees of a method of analyzing human hair proteins to infer non-synonymous SNP (nsSNP) alleles using hair shaft protein. This expands what can forensically and scientifically be achieved with hair trace evidence.

Nuclear DNA typing is not always an option due to degradation or low quantity of starting material. For example, sufficient nuclear DNA is difficult to obtain from hair shafts due to low copy number. Instead, mitochondrial DNA (mtDNA) is commonly used for analysis as it is present in higher copy numbers in the hair shaft; however, mtDNA cannot be used for individualization because it is maternally inherited and there is no recombination. Consequently, all maternally related individuals have the same mtDNA haplotype. In such cases, it would be helpful for the forensic science community to have an alternative, quantitative method for analyzing hair for identification purposes.

With advances in mass spectrometry technology, it has become possible to use proteomic approaches since proteins are more chemically stable than DNA and are found in greater abundance. Recent studies have demonstrated that it is possible to use protein expression profiling to distinguish hair shafts from different body sites and to differentiate between unrelated individuals.^{1,2} Additionally, proteins contain genetic variation in the form of Single Amino acid Polymorphisms (SAPs) that are the result of nsSNPs in the genome. These SAPs can be detected as Genetically Variant Peptides (GVPs) in proteomic datasets. From these GVPs, it is possible to impute the nsSNPs and calculate frequency estimates of profiles. This approach has been demonstrated in recent studies in which GVPs from scalp hair shafts were used to differentiate between unrelated individuals; however, whether this approach can be used for hair shafts from other body sites has not yet been determined.^{2,3}

This study tests the hypothesis that individuals are distinguishable by GVP analysis of hair shafts from any of four body sites: scalp, beard, axillary, or pubic. It is further hypothesized that the GVP profile will not significantly vary between body sites and that hair between different body sites of an individual will have more in common than hair from another individual. To test these hypotheses, hair samples from the four body sites were collected from five volunteer subjects. The hairs were processed using a sodium dodecanoate-based method, then digested with trypsin. The digested peptides were analyzed by Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS), and the resulting proteomic datasets were analyzed using the Global Proteome Machine (<http://www.thegpm.org/>). The datasets were then screened for previously characterized and validated GVPs. Preliminary GVP analysis of type I keratins from these proteomic datasets exhibits evidence of clustering such that hair shafts derived from any of the four body sites of a single individual are more correlated to each other than to those of another unrelated individual. Future work will focus on analyzing additional keratin, keratin-associated, and other proteins, as well as exploiting new sample processing methods to increase detection of characterized GVPs.

Reference(s):

1. Laatsch, C.N. et al. Human hair shaft proteomic profiling: individual differences, site specificity and cuticle analysis. *PeerJ*. 2014. 2: p. e506.
2. Wu, P.W. et al., Proteomic analysis of hair shafts from monozygotic twins: Expression profiles and genetically variant peptides. *Proteomics*. 2017. 17 (13-14): p. 1600462-n/a.
3. Parker, G.J. et al., Demonstration of Protein-Based Human Identification Using the Hair Shaft Proteome. *PLoS One*. 2016. 11 (9): p. e0160653.

Proteomics, Hair, Genetically Variant Peptides