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EDITORIAL

DNA sequencing and metabolomics: new approaches to the forensic assessment of herbal therapeutic agents

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The role and frequency that herbal therapeutic agents play in unexpected deaths remains unclear. Herbal agents may be directly toxic in their own right, or they may act synergistically with other herbal materials, or with prescribed pharmaceutical agents. Herbal therapies are also known to sometimes contain toxic agents such as herbicides or heavy metals, or to have been adulterated with standard drugs to enhance their efficacy [1]. In the medicolegal arena the consumption of drugs that may be contraindicated in certain conditions may not be apparent at the time of autopsy, such as when steroids or non-steroidal anti-inflammatory agents have been unknowingly taken by an individual with active peptic ulcer disease. Substitution of one plant for another, whether because of financial considerations or from a genuine mistake in plant identification, compounds the complexities associated with attempting to determine whether a herbal preparation has contributed to, or caused, illness or death.

Thus, when compared to the assessment of prescribed medications in a standard forensic autopsy, the evaluation of the role of herbal preparations in a death is problematic [2, 3]. Death scene examiners may not document herbal preparations, as they may be considered to be "natural" products and therefore not likely to cause any health problems. Even if herbal medications are recorded at a death scene, there is no guarantee that the ingredients and dosages listed on the container (if this information is available) are a true reflection of the composition of the contained preparation. As standard preparations come as powders, pill or liquids, standard taxonomic classification of herbs by morphology is not possible. Finally, due to the complex organic molecules present and the blending of a number of plants, searching for active organic ingredients in forensic toxicology laboratories may resemble the time honored search for the "needle in haystack" [4].

Fortunately technology is now available which may assist in focusing the search for these elusive active organic agents. Two recent papers [5, 6] show how DNA sequencing can be used to identify materials in herbal medicines that would otherwise be potentially difficult to analyze. In particular, Coghlan et al.'s [5] use of next generation DNA sequencing (NGS) demonstrates how sophisticated analyses of samples, including complex mixtures unable to be evaluated effectively by previous techniques, can be rapidly screened. NGS analysis of herbal medications confiscated at a death scene may provide valuable data that can then be compared to either in-house databases or to larger collections such as the NIH genetic sequence database, GenBank (http://www.ncbi.nlm.nih.gov/genbank/). Although a potential difficulty with external databases involves the accuracy with which entries have been checked prior to uploading, they may, along with smaller databases, be extremely useful in matching the pattern of a DNA sequence to that of known plants. Thus, modern DNA sequencing can rapidly provide a genetic audit of large numbers of cases that can be matched against established standards to focus the search for ingredients [5] such as toxic plants and/or potential allergens.

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Metabolomics, which initially referred to the "comprehensive and quantitative analysis of all metabolites," has been expanded to include not only endogenous small molecules, but also those derived from environmental exposure and medications [7]. Metabolomic studies looking at metabolic derivatives of cellular processes can therefore also be adapted to assist in focusing traditional forensic toxicological evaluations [8, 9]. Specifically, by using high-resolution mass-spectrometry based techniques, metabolomic laboratories are able to screen for, identify, and measure a vast range of substances/molecules, thus producing a "finger print" to characterize the composition of a particular preparation. This includes both primary and secondary metabolites and xenobiotics, the latter representing introduced chemicals such as drugs or pollutants [8]. An essential component of effective metabolomic fingerprinting is having the statistical and database capabilities to analyze high-information content data sets that are routinely generated [7].

By matching patterns of DNA sequences between the preparation under investigation and recognized reference material, the probability that a certain herb belongs to a particular family, genus or species might ultimately be derived. Screening of preparations in metabolomics laboratories can also provide complementary information on other plant and pharmaceutical agents. Combining information from both of these sources may then enable targeted forensic toxicological evaluation of materials that has not previously been possible. For example finding DNA that suggests a certain plant species and identifying a potentially toxic substance produced by that plant greatly assists with plant identification. As well as investigating the medicines themselves, these technologies have the potential to analyze tissues or fluids taken at scenes or autopsies for the specific intrinsic, or added, pharmaceutically active substances. This extensive screening process may provide important data that gives toxicologists the necessary information to concentrate on one group of potential agents. Practically, this process may effectively reduce the "haystack" to a manageable size. The end result is that by initially screening for particular plants and drugs within herbal medicines, focused forensic toxicological analyses can then be undertaken to clarify the potential role of active ingredients in the terminal episode. This type of integrated approach may form a foundation for the further development of the field of forensic herbal toxicology.

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