



### **E19 Analysis of Blood Traces by Attenuated Total Reflection (ATR) Fourier Transform Infrared (FTIR) Spectroscopy for Species Identification**

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The goals of this presentation are to demonstrate: (1) the advantages and disadvantages of current methods for bloodstain analysis during forensic investigation; (2) the importance of species identification from blood traces found at a crime scene; (3) the significance of a non-destructive method for examination of trace evidence at a crime scene; (4) the advantages of FTIR spectroscopy in forensic investigation; (5) the FTIR spectroscopy for bloodstain examination; and, (6) the use of chemometrics for distinguishing between human and animal blood and species identification.

This presentation will impact the forensic science community by demonstrating that a non-destructive, quick, and confirmatory method for species identification based on blood traces found at a crime scene would be of great help to law enforcement. This study reveals great potential of FTIR spectroscopy combined with statistical data analysis for differentiating between species based on bloodstains.

In a forensic investigation, biological evidence can be very helpful for identifying a victim or suspect, as well as for solving a criminal case. Blood identification is typically based on the following steps: visual examination, a presumptive assay, a confirmatory assay, and lastly, the stain can be subjected for species identification before DNA profiling is performed. Presumptive and confirmatory assays are utilized for confirming a stain to be blood. The disadvantage of presumptive tests is the amount of potential false positive reactions with some environmental contaminants. Both presumptive and confirmatory assays require reactants to initiate a chemical reaction and therefore damage the sample. Once a trace found at a crime scene is identified as a bloodstain, it can be subjected for further analysis. Determining the origin of blood is critical in forensic casework since it can streamline the investigation process by including or excluding non-human stains. Immunochromatographic assays are an example of tests for confirming human origin of a stain. The ultimate goal for analyzing blood is DNA profiling, which requires DNA extraction from the sample. Therefore, current standard methods employed for the analysis of blood samples are destructive and time consuming. Another inconvenience for all current types of blood examination, besides DNA analysis, is the limited sample size found at a crime scene. Therefore, it would be highly advantageous to implement a method which is quick, non-destructive, and requires only a small sample with little to no preparation for the identification of a species' blood at a crime scene.

In this study, Attenuated Total Reflection (ATR) FTIR spectroscopy was used as a confirmatory, non-destructive, and rapid method to distinguish blood from different species.<sup>1</sup> Bearing in mind forensic purposes, differentiation of human and non-human blood samples was targeted, and the Partial Least Squares Discriminant Analysis (PLSDA) model that was made demonstrated complete separation between human and animal donors. In addition, the models revealed complete distinction between blood spectra from three species, namely human, cat, and dog. The method was subjected to external validation using samples that were not a part of the training dataset. Classification predictions of unknown blood donors performed by the model resulted in 100% accuracy. This study demonstrated ATR FTIR spectroscopy's great potential for bloodstain analysis and species discrimination. Furthermore, the commercial availability of portable ATR FTIR instruments affirms the potential for the implementation of such bloodstain analyses, at a crime scene as well as in the lab.

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#### **Reference(s):**

- <sup>1</sup> Mistek E., Lednev I.K. Identification of species' blood by attenuated total reflection (ATR) Fourier transform infrared (FT-IR) spectroscopy. *Anal Bioanal Chem.* 2015;407(24):7435-42.

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#### **ATR FTIR Spectroscopy, Blood, Human Origin**