ISSN: 2582-4686 SJIF 2021-3.261,SJIF 2022-2.889, 2024-6.875 ResearchBib IF: 10.25 / 2025

THE MULTIDISCIPLINARY JOURNAL OF SCIENCE AND TECHNOLOGY

VOLUME-6, ISSUE-3

THE APPLICATION OF THE RADIAL IMMUNODIFFUSION METHOD IN AN ANTIBODY-GEL MATRIX FOR DETERMINING BLOOD ORIGIN IN FORENSIC MEDICAL EXAMINATIONS

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ABSTRACT

Currently, the scientific assessment and analysis of biological evidence during criminal investigations is one of the most critical areas of forensic practice. In particular, the identification of blood traces, confirmation of their presence, and differential diagnosis of their origin (human or animal) are of great importance in forensic analysis. Such examinations play a significant role not only in the just evaluation of crimes against persons but also in cases such as poaching, illegal animal slaughter, and livestock-related offenses.

Keywords: forensic medical examination, biological evidence, blood origin, species identification, forensic serology.

Practice shows that existing serological and biochemical methods do not always possess high sensitivity and accuracy. As a result, in 11-16% of cases, it remains impossible to determine the species specificity of the blood. This can directly impact the process of solving a crime and proving or refuting guilt. Furthermore, the age of bloodstains, the influence of environmental factors, contamination, or an insufficient sample size can negatively affect the results. Consequently, there is a growing need in forensic biological examinations for innovative laboratory methods that are highly sensitive, work with minimal sample quantities, and produce reliable and reproducible results. The radial immunodiffusion method in an antibody-gel matrix is one such promising, scientifically-grounded approach that meets these requirements and criteria, enabling the high-precision determination of whether blood is of human or animal origin. The introduction of this method into forensic practice is expected to significantly expand the diagnostic capabilities of forensic laboratories.

The purpose of this study is to evaluate the scientific and practical effectiveness of the radial immunodiffusion method in an antibody-gel matrix for determining the origin of blood traces during forensic biological examinations, and to identify its advantages over traditional serological methods. The study will analyze this method's capability for the differential identification of species-specific proteins in blood, as well as its sensitivity, specificity, reproducibility, and ability to work with minimal samples. Additionally, the diagnostic value of the method in forensic situations - that is, when working with old, contaminated, or absorbed bloodstains on various carriers - will also be investigated.

To conduct a comprehensive study of the applicability of the radial immunodiffusion method, specifically by assessing the theoretical and practical effectiveness of this technique in the differential identification of human and animal blood in forensic biological examinations;

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To determine the sensitivity, specificity, and reproducibility of the method, and to evaluate its capacity to obtain a reproducible result with a minimal sample size and strong signal when identifying species-specific proteins;

To study the influence of the bloodstain substrate, storage duration, and degree of contamination on the analysis results, and to assess the stability of the method when working with blood traces preserved on various types of surface materials, under different environmental conditions, and with varying degrees of contamination.

As research materials, 107 human blood serum samples and 238 blood serum samples belonging to various animal species were used. Additionally, to simulate a forensic scenario, experimental bloodstains prepared on various substrates - such as filter paper, fabric, wood, and other surfaces - were utilized. The bloodstains were stored under various environmental conditions for periods ranging from 2 days to 6 months, bringing their state closer to reflecting real-world examination conditions.

In the study, specially prepared precipitating sera were used for the immunological analysis. The titer of the sera was no less than 1:10,000, and their specificity was previously confirmed by a precipitation reaction in a liquid medium. The primary diagnostic method employed was the radial immunodiffusion technique on an antibody-gel matrix. This method is based on the principle that the antigen and antibody diffuse towards each other, forming precipitation rings within the gel medium.

For the analysis, it was sufficient to extract a minimal amount of biomaterial from the bloodstains - only 2 µl of extract. The process of identifying the species-specific proteins in the samples was conducted under standard laboratory conditions, and the results were evaluated based on the reproducibility of the experiments.

The method demonstrated high sensitivity and accurately differentiated between human and animal blood. The small required sample size was noted as a significant advantage of the analysis. The stability of the results was maintained when working with bloodstains on various substrates. Although sensitivity decreased slightly with increased storage duration of the stains, the analyses remained effective.

Contaminated samples were also analyzed to a satisfactory degree.

In conclusion, the radial immunodiffusion method on an antibody-gel matrix is recommended for determining the species-specificity of blood in forensic biological examinations, as it is a method with high efficiency, minimal sample requirements, and high sensitivity. This method is proposed for application in real-world forensic expert examinations.

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