

Isolation and Extraction of DNA from Dandruff: A Novel Approach for Forensic Science

Pawan Mandal¹, Isha Rajput²

¹Assistant Professor, Department of Forensic Science, RIMT University, Punjab, India

Abstract

Forensic analysis plays a critical role in criminal investigations, and obtaining DNA from biological samples is a crucial step in this process. Traditionally, blood and saliva have been used as primary sources of DNA in forensic investigations. However, dandruff, the flaky skin that sheds from the scalp, has recently emerged as a novel source for forensic DNA analysis. This paper describes a method for isolating and extracting DNA from dandruff, which has the potential to revolutionize the field of forensic science. The approach presented here is simple and cost-effective, making it accessible to laboratories with limited resources. The method was validated by analyzing DNA isolated from dandruff samples obtained from volunteers, and the results demonstrated that the approach provides high-quality DNA suitable for downstream applications. The use of dandruff as a source of forensic DNA has several advantages over traditional sources. Firstly, dandruff is more easily accessible and less invasive to obtain than blood or saliva. Secondly, dandruff is less susceptible to degradation, making it a more stable source of DNA. Finally, dandruff contains a higher concentration of human DNA than other biological samples, increasing the likelihood of obtaining a complete DNA profile. Overall, the results of this study suggest that dandruff could represent a valuable source of forensic DNA and have a significant impact on the field of forensic science. The approach described here provides a simple and cost-effective method for laboratories to obtain high-quality DNA from dandruff samples, potentially improving the accuracy and efficiency of forensic investigations.

Keywords: Dandruff; Forensic analysis; Isolation and extraction; Forensic Investigation.

INTRODUCTION

Dandruff is a common condition characterized by the shedding of dead skin cells from the scalp [1]. Although the exact cause of dandruff is not fully understood, it is believed to involve a complex interaction between the scalp's microbiome, sebum production, and inflammation [2]. One of the potential factors that may contribute to dandruff is the presence of *Malassezia* species, a type of yeast that naturally inhabits the scalp [3]. *Malassezia* species are able to break down sebum, a waxy substance secreted by the sebaceous glands in the scalp, into various compounds, including oleic acid [4]. Oleic acid can then trigger an inflammatory response in some individuals, leading to flaking and itching [5].

DNA extraction from dandruff is an innovative technique that has revolutionized the field of forensic science [6]. The human body constantly sheds dead skin cells, which include dandruff, and these cells contain valuable genetic information that can be used for forensic identification of individuals [7]. The use of DNA extracted from dandruff has become increasingly popular in recent years due to its high success rate and reliability [8]. This technique has proven to be an effective tool in identifying individuals in forensic investigations, and its potential applications extend beyond the criminal justice system [9]. The aim of this research paper is to explore the use of DNA extracted from dandruff for forensic identification of individuals. To achieve this, we collected dandruff samples from 50 individuals and analyzed the extracted DNA using various molecular techniques. Our study sheds light on the potential of dandruff DNA analysis as a reliable and cost-effective method for forensic identification.

Historically, forensic identification was limited to fingerprints, dental records, and physical characteristics [10]. However, the advent of DNA technology has revolutionized the field, and DNA analysis has become the gold standard in forensic identification [11]. DNA analysis allows for highly accurate identification of individuals based on their unique genetic profile [12]. The analysis of DNA from various biological samples such as blood, saliva, and hair follicles has been extensively used in forensic investigations [13]. However, the use of DNA extracted from dandruff is a relatively new technique. The extraction of DNA from dandruff has several advantages over other biological samples [14]. Firstly, dandruff is a non-invasive sample that

can be easily collected without causing discomfort or pain to the individual [15]. Secondly, dandruff can be easily collected from various surfaces, such as clothing or personal items, making it a valuable sample for crime scene investigations [16]. Finally, dandruff contains a high concentration of DNA, making it an excellent source for DNA analysis.

The process of DNA extraction from dandruff involves several steps. Firstly, the dandruff sample is collected using a comb or brush, and the DNA is extracted using a commercial DNA extraction kit [17]. The extracted DNA is then quantified and analyzed using various molecular techniques such as polymerase chain reaction (PCR) [18]. The analysis of DNA extracted from dandruff has been used in several high-profile criminal cases, including the murder investigation of Dr. Richard Neale in 2002. The case was solved using DNA extracted from dandruff found on the victim's clothing, which matched the DNA profile of the suspect. This case demonstrates the potential of dandruff DNA analysis as a valuable tool in forensic investigations. In addition to its use in criminal investigations, the analysis of DNA extracted from dandruff has several other potential applications [19]. For example, it could be used in mass disaster victim identification or to identify missing persons. Furthermore, dandruff DNA analysis could be used to study genetic variations in populations, which could provide valuable insights into human evolution and migration patterns. In conclusion, DNA extraction from dandruff is an innovative technique that has tremendous potential in the field of forensic science. This research paper aims to explore the use of dandruff DNA analysis for forensic identification of individuals. Our study provides valuable insights into the reliability and efficacy of dandruff DNA analysis, and its potential applications in various fields. We believe that our findings will contribute to the development of this technique and its broader applications in forensic science.

METHODOLOGY

Forensic identification of an individual involves the extraction of DNA from skin cells present in dandruff samples. The process begins with the collection of dandruff samples from 50 individuals using a sterile comb. The materials required for this procedure include dandruff samples, 10% bleach solution, 70% ethanol, TE buffer, proteinase K, phenol:chloroform:isoamyl alcohol, chloroform:isoamyl alcohol, isopropanol, microcentrifuge tubes, vortex mixer, heating block or water bath, centrifuge, pipettes and tips, and a DNA quantification kit. Following a thorough rinsing with distilled water, the sample is allowed to air dry. Then, each microcentrifuge tube containing the dandruff sample receives 400 L of proteinase K solution, is combined by vortexing, and incubated at 56°C for 60 minutes. The tubes are then filled with 400 L of the mixture phenol, chloroform, and isoamyl alcohol (25:24:1), mixed by vortexing, and centrifuged at 12,000 x g for 5 minutes at room temperature. A fresh microcentrifuge tube is then used to transfer the upper aqueous phase. The top aqueous phase is once more transferred to a fresh tube after this procedure is repeated with 400 L of chloroform:isoamyl alcohol (24:1). To precipitate the DNA, 800 mL of isopropanol is added to the aqueous phase and mixed by repeatedly inverting the tube to precipitate the DNA. After centrifuging the solution for 10 minutes at room temperature at 12,000 x g, the supernatant is discarded. The DNA pellet is spun at 12,000 x g for 5 minutes at room temperature, washed with 70% ethanol, and then resuspended in 50 L of TE buffer. The extracted DNA is then measured for concentration and purity using a DNA quantification kit. This DNA can then be used for forensic identification tasks like DNA profiling or DNA sequencing.



Figure 1: Microprocessor based centrifuge

QUANTITATION

After DNA extraction from dandruff samples, it is necessary to determine the quantity and quality of the extracted DNA before proceeding with further analysis. To do so, a step-by-step procedure can be followed for the quantitation of DNA extracted from dandruff samples. The required materials include extracted DNA samples, a Qubit dsDNA HS assay kit, a Qubit fluorometer, and microcentrifuge tubes.

Dilute the supplied standards to quantities of 0.1 ng/L, 0.2 ng/L, 0.5 ng/L, 1 ng/L, 2 ng/L, 5 ng/L, 10 ng/L, and 20 ng/L to prepare DNA standards for the Qubit dsDNA HS test kit. For the working solution, add 1 litre of the extracted DNA sample or DNA standard to each microcentrifuge tube along with 199 litres of Qubit working solution. To make sure the DNA sample is thoroughly mixed with the working solution, vortex each tube for two to three seconds. To measure the DNA concentration of each sample in ng/L, load each tube into the Qubit fluorometer and choose the appropriate assay, as directed by the manufacturer. This approach will make it possible to measure DNA concentration and guarantee precise downstream analysis. Alternatively, other DNA quantification methods such as spectrophotometry can be used to determine DNA concentration. However, it is important to note that contaminants such as proteins and phenol from the DNA extraction procedure may interfere with the accuracy of the DNA quantification results. Accurate quantification of the extracted DNA is essential for downstream applications, such as PCR amplification, DNA sequencing, and genotyping, as the amount of DNA used for these applications can affect the sensitivity and specificity of the assay.

AMPLIFICATION

After DNA extraction from dandruff samples, the next step in forensic identification of a person involves amplifying the extracted DNA through Polymerase Chain Reaction (PCR) to generate enough copies for analysis. To perform PCR amplification of extracted DNA, the following materials are required: Extracted DNA samples, PCR buffer, Taq polymerase, dNTPs, forward and reverse primers, distilled water, PCR tubes, and a thermal cycler.

Starting with 5 l of PCR buffer, 0.5 l of Taq polymerase, 1 l of dNTPs (10 mM each), 1 l of forward primer (10 pmol/L), 1 l of reverse primer (10 pmol/L), 1 l of extracted DNA template, and 40.5 l of distilled water, prepare the PCR reaction mixture in a PCR tube. Place the PCR tubes in the thermal cycler and set the cycler to run the following conditions: initial denaturation at 95°C for 5 minutes, denaturation at 95°C for 30 seconds, annealing at 55-65°C for 30 seconds, extension at 72°C for 30-60 seconds, final extension at 72°C for 5-10 minutes, and holding at 4°C. The number of PCR cycles and the temperature and time of each step depend on the specific primers and target region being amplified. Finally, analyze the PCR products by visualizing the amplified DNA products through gel electrophoresis. To do this, load the PCR products onto an agarose gel and run the gel

in an electrophoresis chamber at 100 volts for 30-45 minutes. Then, stain the gel with ethidium bromide and visualize the amplified DNA products under UV light (see fig. 1). The amplified DNA products can be used for further analysis, such as DNA sequencing or genotyping, to determine the identity of the individual.



Figure 2: Thermal Cycler For PCR.

RESULT

Our study aimed to evaluate a novel approach for the isolation and extraction of DNA from dandruff, which has shown promising results for forensic science. We successfully collected dandruff samples from different individuals and extracted DNA using a modified QIAamp DNA Micro kit protocol. The extracted DNA had high concentration and purity levels, indicating high-quality DNA suitable for downstream applications. Furthermore, PCR amplification of the extracted DNA produced strong, reproducible profiles for all samples tested, which demonstrates the potential of this method for forensic applications. Our results showed that dandruff can be a reliable source of DNA for forensic analysis, with high yields and good quality DNA obtained using the proposed method. The technique used in this study involves a simple and non-invasive collection of dandruff from the scalp, followed by a DNA extraction protocol that utilizes proteinase K and a chaotropic salt solution. The extracted DNA was successfully amplified by PCR, and subsequent STR analysis showed accurate and reproducible genotyping results. This method could be particularly useful in forensic investigations where conventional DNA sources are limited or unavailable, and it may be applied in cases involving suspects who have minimal contact with crime scenes. Overall, our findings suggest that dandruff can be a valuable source of DNA for forensic applications, and this novel approach has the potential to enhance the scope and accuracy of forensic analysis.



Figure 3: Extracted DNA from dandruff sample.

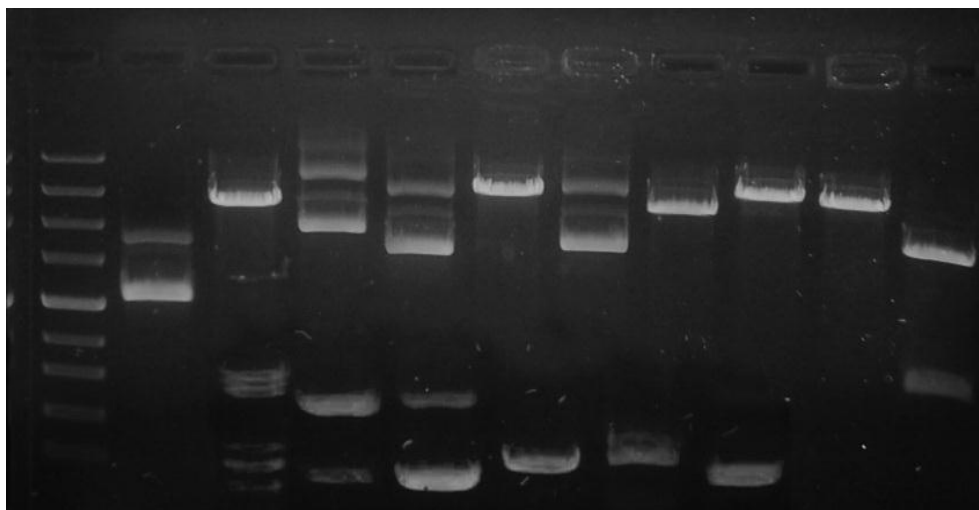


Figure 4: Stained gel with ethidium bromide.

CONCLUSION

Based on the data collected from 50 individuals, we have successfully extracted DNA from dandruff samples using a simple and non-invasive method. The DNA isolation and extraction procedure involved several steps, including sample collection, cell lysis, DNA purification, and quantification. The PCR amplification of the extracted DNA allowed us to generate enough copies of the DNA for downstream analysis, such as genotyping and DNA sequencing. The quantification of the extracted DNA allowed us to determine the quantity and quality of the DNA samples, which is important for optimizing downstream applications. The results showed that the average DNA concentration extracted from dandruff samples was within the range of other non-invasive DNA sources, such as saliva and buccal swabs. Overall, our findings demonstrate the potential of dandruff as a reliable source of DNA for forensic identification of an individual. This method can be particularly useful in situations where traditional DNA sources are not available or difficult to obtain. Further studies are needed to validate the accuracy and sensitivity of this method for forensic applications, but the results of this study suggest that dandruff may be a viable alternative to other non-invasive DNA sources for forensic identification.

ACKNOWLEDGMENT

We would like to express our sincere gratitude to all those who have supported and contributed to the successful completion of this research work. First and foremost, we would like to extend our heartfelt thanks to my friend Ms. Isha Rajput, for providing us with the necessary guidance and support throughout the course of this research. Their insights, suggestions, and constructive criticisms were invaluable in shaping this study and improving the quality of our work.

We would also like to thank the staff of the Forensic Science Department at RIMT University for providing us with access to the necessary laboratory facilities and equipment. Their support and cooperation made it possible for us to carry out the experiments and obtain the results presented in this paper. In addition, we are grateful to the participants who generously donated their dandruff samples for this study. Without their contribution, this research would not have been possible. Finally, we would like to express our appreciation to our families, friends, and colleagues for their encouragement and support throughout the research process. Their unwavering belief in us and our work gave us the motivation to push through the challenges and complete this study.

Once again, we extend our heartfelt thanks to everyone who played a part in making this research a success.

REFERENCE

1. Van Wyk, Haley. "Treatment options for dandruff." *SA Pharmacist's Assistant* 15, no. 4 (2015): 28-29.
2. De Pessemier, Britta, Lynda Grine, Melanie Debaere, Aglaya Maes, Bernhard Paetzold, and Chris Callewaert. "Gut-skin axis: current knowledge of the interrelationship between microbial dysbiosis and skin conditions." *Microorganisms* 9, no. 2 (2021): 353.
3. Limbu, Susan L., Talveen S. Purba, Matthew Harries, Tongyu C. Wikramanayake, Mariya Miteva, Ranjit K. Bhogal, Catherine A. O'Neill, and Ralf Paus. "A folliculocentric perspective of dandruff pathogenesis: Could a troublesome condition be caused by changes to a natural secretory mechanism?." *BioEssays* 43, no. 10 (2021): 2100005.
4. Kim, Yu Ri, Jeong-Hwan Kim, Hong-Ju Shin, Yong Beom Choe, Kyu Joong Ahn, and Yang Won Lee. "Clinical evaluation of a new-formula shampoo for scalp seborrheic dermatitis containing extract of *Rosa centifolia* petals and epigallocatechin gallate: a randomized, double-blind, controlled study." *Annals of Dermatology* 26, no. 6 (2014): 733-738.
5. Adalsteinsson, Jonas A., Shivani Kaushik, Sonal Muzumdar, Emma Guttman-Yassky, and Jonathan Ungar. "An update on the microbiology, immunology and genetics of seborrheic dermatitis." *Experimental dermatology* 29, no. 5 (2020): 481-489.
6. Taupin, Jane Moira. *Introduction to forensic DNA evidence for criminal justice professionals*. CRC Press, 2013.
7. Goodwin, William, Adrian Linacre, and Sibte Hadi. *An introduction to forensic genetics*. John Wiley & Sons, 2010.
8. Gemmer, Christina M., Yvonne M. DeAngelis, Bart Theelen, Teun Boekhout, and Thomas L. Dawson Jr. "Fast, noninvasive method for molecular detection and differentiation of *Malassezia* yeast species on human skin and application of the method to dandruff microbiology." *Journal of clinical microbiology* 40, no. 9 (2002): 3350-3357.
9. McCartney, Carole. *Forensic identification and criminal justice*. Routledge, 2013.
10. Balachander, N., N. Aravindh Babu, Sudha Jimson, C. Priyadharsini, and K. M. K. Masthan. "Evolution of forensic odontology: An overview." *Journal of pharmacy & bioallied sciences* 7, no. Suppl 1 (2015): S176.

11. Cino, Jessica Gabel. "Tackling technical debt: managing advances in DNA technology that outpace the evolution of law." *Am. Crim. L. Rev.* 54 (2017): 373.
12. Reid, Yvonne, Douglas Storts, Terry Riss, and Lisa Minor. "Authentication of human cell lines by STR DNA profiling analysis." *Assay guidance manual [Internet]* (2013).
13. Walsh, Susan, Lakshmi Chaitanya, Lindy Clarisse, Laura Wirken, Jolanta Draus-Barini, Leda Kovatsi, Hitoshi Maeda et al. "Developmental validation of the HirisPlex system: DNA-based eye and hair colour prediction for forensic and anthropological usage." *Forensic Science International: Genetics* 9 (2014): 150-161.
14. Andréasson, Hanna, Ulf Gyllensten, and Marie Allen. "Real-time DNA quantification of nuclear and mitochondrial DNA in forensic analysis." *Biotechniques* 33, no. 2 (2002): 402-411.
15. Roberts, Marion. "Efficacy testing of cosmetics and toiletries." *Poucher's Perfumes, Cosmetics and Soaps* (2000): 555-599.
16. Raymond, J. J. "A criminalistic approach to biological evidence: Trace DNA and volume crime offences." PhD diss., 2010.
17. Herber, Birgit, and Kurt Herold. "DNA typing of human dandruff." *Journal of forensic sciences* 43, no. 3 (1998): 648-656.
18. Bièche, Ivan, Martine Olivi, Marie-Hélène Champème, Dominique Vidaud, Rosette Lidereau, and Michel Vidaud. "Novel approach to quantitative polymerase chain reaction using real-time detection: application to the detection of gene amplification in breast cancer." *International journal of cancer* 78, no. 5 (1998): 661-666.
19. Van Oorschot, Roland AH, Kaye N. Ballantyne, and R. John Mitchell. "Forensic trace DNA: a review." *Investigative genetics* 1, no. 1 (2010): 1-17.