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Extraction of DNA fingerprinting from human hair in alcohol base after potentization

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Abstract

Through this research work extracted the DNA Fingerprinting from Biological sample of Human hair in the alcohol base after potentization. The method which we used is DNA Quantification is PCR in 1% agarose gel, afterwards successfully determination of DNA Fingerprinting in 5x and 10x from Human Hair in alcohol base after potentization.

Keywords: DNA fingerprinting, potentization, alcohol base, biological sample

Introduction

The study of traces of DNA in hair, a crucial piece of evidence in forensic science, has recently been made possible using the polymerase chain reaction (PCR) [1, 2]. Nonetheless, minuscule amounts of DNA are present in hairs [1]. Since nuclear DNAs, especially those from naturally shed hairs or hair shafts, have too little content to be amplified, many investigations have used comparatively abundant mtDNA instead. Compared to hair roots [3-5]. Furthermore, PCR inhibitors may be present in the collected samples because even when enough amounts of DNA are isolated from hair, PCR does not always successfully amp up the DNA. Melanin, the pigment found in hair, has been shown in earlier studies to be a potent inhibitor of the PCR process [6-8]. In particular, PCR is significantly impacted by hair dyeing [6]. In this study, we compare three distinct techniques for obtaining DNA from human hair, including The D1S8 (MS32) locus was amplified using minisatellite variant repeat (MVR)-PCR [10], the Chelex method [9], the QI Aamp DNA Mini Kit method, and the ISOHAIR® method to acquire DNA without PCR inhibitory chemicals. Lastly, the MVR-PCR patterns of DNA extracts from hair roots and buccal swabs were compared to determine if DNA isolated from hairs using the Chelex method, particularly dyed hairs, reflects genomic DNA.

Materials and Methodology

Type of study: DNA fingerprinting analysis

Site of study: Department of Life Sciences, Parul Institute of Applied Sciences, Parul University, Vadodara

Duration of study: 15 days

Sample used: Potentized Alcohol base up to 100 Downwards strokes with 3-5 Human hair

Method for potentization: 100 downwards strokes by Electric potentizer machine

Procedure:

There are following process involved, such as;

1st Step: Collection of Hair sample consist hair bulb 3 to 5

2nd Step: Aseptically cut the hair bulb portion (3mm above the bulb)

3rd Step: Collect hair bulb portion in 1.5 ml MCT with 40 µL of 5% Protinase K

4th Step: Aseptically cut the hair bulb portion (3mm above the bulb)

5th Step: Vortexing & Incubate for overnight at 60 °C

6th Step: add equal volume of P:C:I (25:24:1) & mix by inverting tube for 1 min

7th Step: vortexing & Incubate for overnight at 60 °C

8th Step: Centrifuge at 10,000 g for 10 minutes at 4 °C & Transfer transfer new aqueous layer in MCT

9th Step: 10 µL of 10 mg of RNAase added and kept for 30 minutes at 37 °C

10th Step: Add equal volume of C1 Centrifuge 1000 g for 10 minutes at 4 °C

11th Step: Transfer transfer new aqueous layer in MCT add double volume of IPA & 1/10th of 3M Sodium acetate

12th Step: incubate -20 °C for 1 hour for precipitation Centrifuge 1000 g for 10 minutes at 4 °C

13th Step: Pellets were wash with 250 µL of EtOH & Centrifuge 1000 g for 10 minutes at 4 °C

14th Step: Discard the supernatant and dry the pellets suspend in 20 µL in TE (p H 8)

15th Step: Quantify the DNA (A260/280) & run it on 2% DNA Agarose gel

Results

Table 1: DNA quantification (Nano drop)

Dilution	A260/280	ng/uL	x Dilution factor
5x	2.139	10.088	50.44
10x	1.9	5.154	51.54

1 µg = 19.8µL

Agarose gel run (1%)

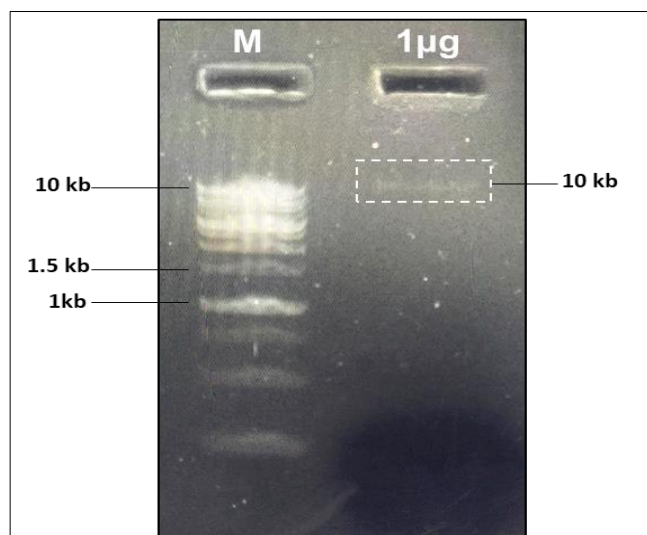


Fig 1: DNA Fingerprinting from Human hair in Agarose gel

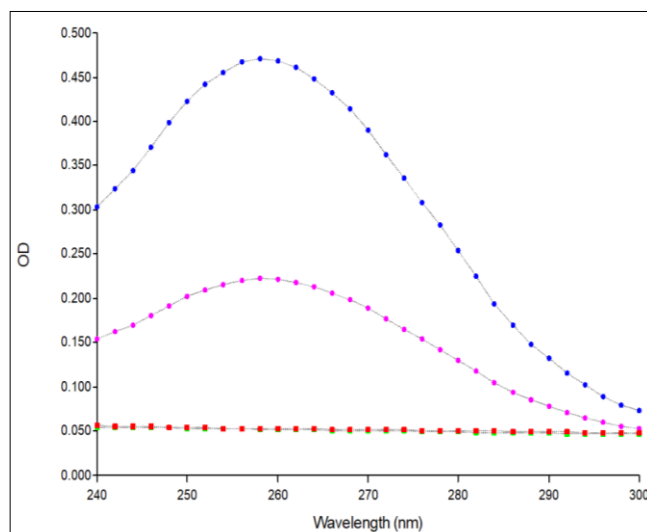


Fig 2: Wavelength

Conclusion

Successfully extracted DNA Fingerprinting from Human hair in alcohol base after potentization up to 100 downwards strokes

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Conflict of interest

No such

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