



A Comparative Study on the Yield of DNA Extracted from Fresh & Condition Exposed Human Scalp Hair Samples

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Abstract:

DNA profiling is a highly conclusive, informative procedure in identification of biological specimens. In the current study, Quantification of DNA was done to detect the concentration of DNA in the samples. Quantity of DNA was calculated by taking the absorbance of samples at 260 nm by Spectrophotometer. In total 100 samples of human scalp hairs were subjected for extraction and quantification of DNA. Samples were exposed to different atmospheric and physical condition like water, stagnant dirty water and detergent for a period of 5 days.

Although the DNA quantification was possible from all samples subjected for analysis, it was found that there was an appreciable difference in the quantity of DNA extracted from sample exposed to different atmospheric and physical condition. When an inter comparison was done among the fresh samples and the samples exposed to different conditions it was found that the highest % difference in concentration was found to be among fresh sample and detergent condition sample. The results of the entire analysis showed the highest t-value between conditions was for the samples exposed to detergent condition and water conditions. The results of the entire analysis showed the lowest p-value between conditions was for the samples exposed to detergent condition and water conditions.

A study in the similar line by considering more sample size and by considering more variables would definitely give greater scope for forensic scientists, DNA analysts & forensic serologist to scientifically validate the data generated through such kind of replicative studies which would in turn help them to conduct analysis, experiments involved in DNA analysis from scalp hair samples having root follicle cells collected from scene of crime, assailant, victims, weapons of assault, clothing, vehicles, cap, scarf, bandana, hat, mask, comb, etc.

Keywords: DNA Quantification, DNA from hair, Hair Evidence, DNA from Condition Exposed hair samples.

Introduction:

Deoxyribonucleic acid, or DNA, is the molecule of life. It is the chemical code specifying our function, appearance and pedigree and is unique for all individuals except identical twins. An individual's DNA is formed by combination of DNA from his or her parents with half coming from the mother and half from the father. For this reason, DNA testing can be used as evidence of paternity of a child.

DNA is found in most cells of the body, including white blood cells, semen, hair roots and body tissue. Traces of DNA can be detected in body fluids such as saliva and perspiration. Mitochondrial DNA, which follows the maternal line of an individual, can be extracted from hair and bone samples. This can be used to examine relatedness and common ancestry between individuals, and to verify the identity of buried remains. This technique was used in the much publicized case of the Romanovs.

Forensic Science utilizes the properties of DNA in several ways. The adage "**every contact leaves a trace**" indicates the importance of a technique able to type trace amounts of genetic material left during the commission of a crime. Hairs or saliva left on a balaclava worn during a robbery, semen located at a rape scene, blood collected from an assault, perspiration on clothing, traces of assailant's skin under a victim's fingernails, can often be DNA profiled. This genetic information can then be used to include or exclude suspects as being the source of the genetic material.

Deoxyribonucleic acid (DNA) contains the genetic information, used in the development and functioning of living organisms. DNA is often called as a '**blueprint of life**', since it contains the hereditary information that an organism require to function. Genes are DNA segments hat carry genetic information. DNA profiling is a highly conclusive, informative procedure in identification of biological specimens, genomic diversity in population studies, characterization and tracing antiquity of ancient DNA and for diagnosis of a plethora of diseases.

Method:

In total 100 samples of human scalp hairs were subjected for extraction and quantification of DNA. These hair samples were collected randomly from the adult human volunteers belonging to age group 21-25 years. In this 10 fresh human scalp hair samples collected were subjected to analysis in the fresh stage itself. Out of the remaining 90 samples, 10 samples each were exposed to different condition like water, stagnant dirty water and detergent for a period of 5 days.

For the extraction of DNA from Hair root follicles, Approximately 1 cm of hair along with the root was cut and placed in microcentrifuge tube. The samples were vortexed. For each sample, one NucleoSpin® Tissue Column was placed into a

collection Tube. It was then centrifuged for 1 min at 11,000 rpm. The flow-through was discarded and the column was placed back into the Collection Tube. The NucleoSpin® Tissue Column was placed into a 1.5 ml microcentrifuge tube and 100 µl pre-warmed Buffer BE (70 °C) was added to it. It was then incubated at room temperature for 1 min and Centrifuged for 1 min at 11,000 rpm. The DNA was thus obtained in the microcentrifuge tubes and was stored at -20 °C.

Quantification of DNA was done to detect the concentration of DNA in the sample. Quantity of DNA was calculated by taking the absorbance of samples at 260 nm by Spectrophotometer. Readings were taken for all the samples.

Observations:

Table 1: Absorbance of DNA obtained from fresh hair samples and hair samples exposed to water at 260 nm, concentration of DNA obtained in µg/ml and percentage difference in concentration

Fresh Absorbance	Fresh Concentration	Condition 1 Absorbance	Condition 1 Concentration	% Difference in Concentration
0.351	70.2	0.229	45.8	34.76
0.345	69	0.225	45	34.78
0.346	69.2	0.224	44.8	35.26
0.343	68.6	0.229	45.8	33.24
0.342	68.4	0.226	45.2	33.92
0.349	69.8	0.228	45.6	34.67
0.366	73.2	0.228	45.6	37.7
0.346	69.2	0.231	46.2	33.24
0.348	69.6	0.227	45.4	34.77
0.344	68.8	0.226	45.2	34.3

Mean of % difference in Concentration= 34.66

- The absorbance of the DNA obtained from fresh hair root follicles was found to be ranging from **0.342 to 0.366**
- The Concentration of the DNA obtained from fresh hair root follicles was found to be ranging from **68.8 to 73.2 µg/ml**.
- The absorbance of the DNA obtained from hair root follicles exposed to water was found to be ranging from **0.224 to 0.231**
- The Concentration of the DNA obtained from hair root follicles exposed to water was found to be ranging from **44.8 to 46.2 µg/ml**.
- The **mean percentage difference** in the concentration of the DNA was found to be **34.66%**.

Table 2: Absorbance of DNA obtained from fresh hair samples and hair samples exposed to dirty stagnant water at 260 nm, concentration of DNA obtained in µg/ml and percentage difference in concentration.

Fresh Absorbance	Fresh Concentration	Condition 2 Absorbance	Condition 2 Concentration	% Difference in Concentration
0.351	70.2	0.158	31.6	54.99
0.345	69	0.152	30.4	55.94
0.346	69.2	0.153	30.6	55.78
0.343	68.6	0.152	30.4	55.69
0.342	68.4	0.155	31	54.68
0.349	69.8	0.163	32.6	53.30
0.366	73.2	0.158	31.6	56.83
0.346	69.2	0.164	32.8	52.60
0.348	69.6	0.166	33.2	52.30
0.344	68.8	0.165	33	52.03

Mean of % difference in Concentration= 54.41

- The absorbance of the DNA obtained from fresh hair root follicles was found to be ranging from **0.342 to 0.366**
- The Concentration of the DNA obtained from fresh hair root follicles was found to be ranging from **68.8 to 73.2 µg/ml**.
- The absorbance of the DNA obtained from hair root follicles exposed to dirty stagnant water was found to be ranging from **0.152 to 0.166**
- The Concentration of the DNA obtained from hair root follicles exposed to dirty stagnant water was found to be ranging from **30.4 to 33.2 µg/ml**.
- The mean percentage difference in the concentration of the DNA was found to be **54.41%**

Table 3: Absorbance of DNA obtained from fresh hair samples and hair samples exposed to detergent at 260 nm, concentration of DNA obtained in µg/ml and percentage difference in concentration.

Fresh Absorbance	Fresh Concentration	Condition 8 Absorbance	Condition 8 Concentration	% Difference in Concentration
0.351	70.2	0.068	13.6	80.63
0.345	69	0.061	12.2	82.32
0.346	69.2	0.066	13.2	80.92
0.343	68.6	0.064	12.8	81.34
0.342	68.4	0.057	11.4	83.33
0.349	69.8	0.055	11	84.24
0.366	73.2	0.063	12.6	82.79
0.346	69.2	0.059	11.8	82.95
0.348	69.6	0.062	12.4	82.18
0.344	68.8	0.074	14.8	78.49

Mean of % difference in Concentration=81.92

- The absorbance of the DNA obtained from fresh hair root follicles was found to be ranging from **0.342 to 0.366**
- The Concentration of the DNA obtained from fresh hair root follicles was found to be ranging from **68.8 to 73.2 µg/ml**.
- The absorbance of the DNA obtained from hair root follicles exposed to detergent was found to be ranging from **0.055 to 0.074**
- The Concentration of the DNA obtained from hair root follicles exposed to detergent was found to be ranging from **11 to 14.8 µg/ml**.
- The mean percentage difference in the concentration of the DNA was found to be **81.92%**

Chart 1: Showing the Mean absorbance and Mean Concentration of DNA extracted from fresh hair root follicles & hair follicles exposed to Stagnant Dirty Water Condition.

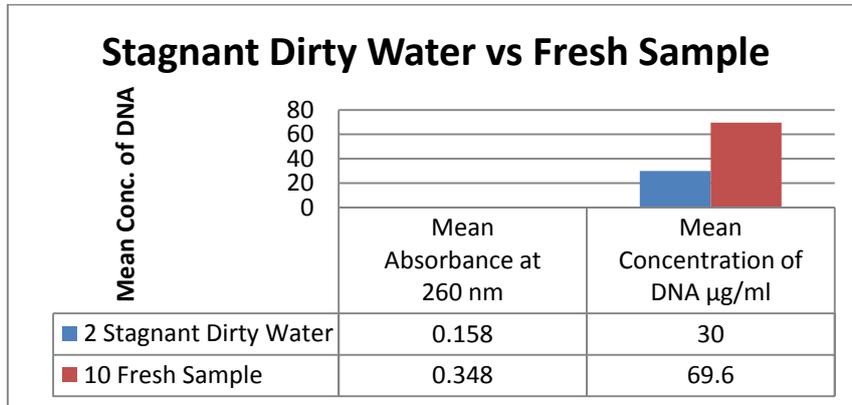


Chart 2: Showing the Mean absorbance and Mean Concentration of DNA extracted from fresh hair root follicles & hair follicles exposed to Detergent Condition.

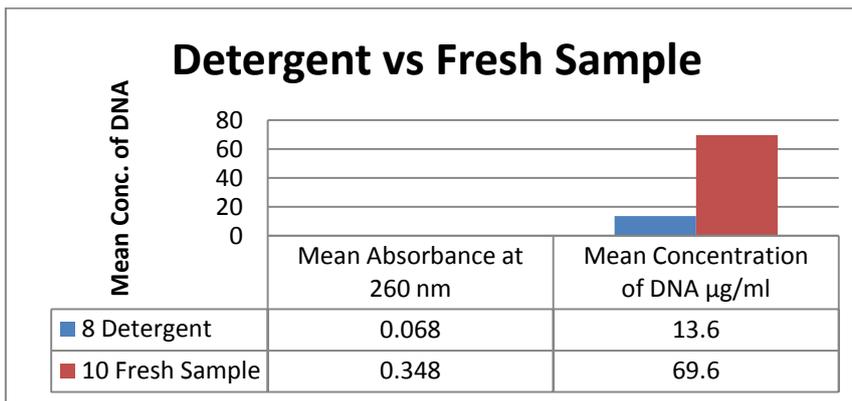
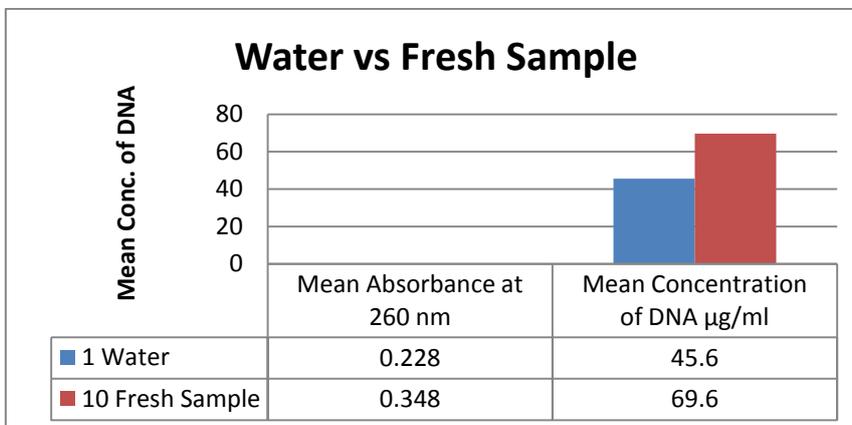


Chart 3: Showing the Mean absorbance and Mean Concentration of DNA extracted from fresh hair root follicles & hair follicles exposed to Water Condition.



Result & Discussion:

From the entire analysis, it was found that the lowest quantity of DNA extracted was from the hair root follicle cells exposed to detergent condition respectively. The results of the entire analysis showed the highest t-value between conditions was for the samples exposed to detergent condition and water conditions. The results of the entire analysis showed the lowest p-value between conditions was for the samples exposed to detergent condition and water conditions. Our studies are in accordance with the study of Amory S et.al., they suggested that the nuclear DNA can be successfully extracted from ancient hair shafts and our studies suggest that in various damaged conditions we can extract ample amount of DNA for further examinations. Our findings were also supported by He BF and Lou DD, they reported that we can perform profiling of DNA from highly degraded and mummified remains.

The results of the entire analysis conducted in this present study, it was possible to extract and quantify the DNA from scalp hair samples with root follicle cells, collected randomly from adult human volunteers, which had been exposed to different atmospheric and physical conditions like water, stagnant dirty water, and detergent. Although the DNA quantification was possible from all samples subjected for analysis, it was found that there was an appreciable difference in the quantity of DNA extracted from sample exposed to different atmospheric and physical conditions.

Conclusion:

The present study results are definitely giving scope for the extraction & quantification of DNA for individualization from the human scalp hairs (with root follicle cells) exposed to different atmospheric and physical conditions. A study in the similar line by considering more sample size and by considering more variables would definitely give greater scope for forensic scientists, DNA analysts & forensic serologist to scientifically validate the data generated through such kind of replicative studies which would in turn help them to conduct analysis, experiments involved in DNA analysis from scalp hair samples having root follicle cells collected from scene of crime, assailant, victims, weapons of assault, clothing, vehicles, cap, scarf, bandana, hat, mask, comb, etc.

References:

1. He BF., Lou DD., A reviewing for profiling of highly degraded remains. *Fa Yi Xue Za Zhi*. 2006 Apr; 22(2):156-8.
2. Amory S., Keyser C., Crubézy E and Ludes B., STR typing of ancient DNA extracted from hair shafts of Siberian mummies. *Forensic Sci Int*. 2007 Mar 2; 166(2-3):218-29. Epub 2006 Jul 12.
3. Just RS., Leney MD., Barritt SM., Los CW., Smith BC., Holland TD., and Parsons TJ., The use of mitochondrial DNA single nucleotide polymorphisms to assist in the resolution of three challenging forensic cases. *J Forensic Sci*. 2009 Jul; 54(4):887-91. Epub 2009 May 26.
4. Köhnemann S., Pennekamp P., Schmidt PF., and Pfeiffer H., qPCR and mtDNA SNP analysis of experimentally degraded hair samples and its application in forensic casework. *Int J Legal Med*. 2010 Jul; 124(4):337-42. Epub 2010 May 25.
5. Allen M, Engström AS, Meyers S, Handt O, Saldeen T, von Haeseler A, Pääbo S, Gyllenstein U., Mitochondrial DNA sequencing of shed hairs and saliva on robbery caps: sensitivity and matching probabilities, *J Forensic Sci*. 1998 May;43(3):453-64.
6. Pfeiffer H., Huhne J., Ortmann C., Waterkamp K and Brinkmann B., /mitochondrial DNA typing from human axillary. *Int J Legal Med*. 1999; 112(5):287-90.
