Original Research Paper

Decay in Intact DNA Recovery in Blood Samples kept at Room Temperature

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Abstract

Blood Samples were kept at room temperature for a period of 3-6 months at room temperature to know the amount of quantitative DeoxyriboNucleic Acid [DNA] recovery from these samples. We are able to recover good amount of DNA for about first 3-6 weeks after which the DNA is decreased drastically and after two months there hardly any chance of intact DNA recovery from these samples. It has been concluded that blood samples recovered from scene of crime after about 1-2 months is a waste. The samples must be recovered as early as possible to recover intact DNA from them. The samples must be collected within 1-2 months from scene of crime until and unless the climate is cold enough to increases decay time. This study is very useful for the investigating authorities which can make errors while collecting blood samples for DNA analysis.

Key Words-Blood Sample, DNA, Scene of Crime

Introduction:

Blood is the most important and most common Forensic Sample recovered from the scene of crime however the time of recovery may differ from fresh blood to many months. Most of the time blood was sent to the Forensic Laboratory for DNA isolation and the investigation agencies thought that it is a minor work to do, which is not so. Our study is based on the fact that the DNA yield decreases with time [1] but what is that time period after which we are unable to isolate the intact DNA from these samples.

Although there is no hard and fast rule that this is the time after which we are unable to recover intact DNA from the samples but our study just show chances of recovery. In the first phase of our study we took only one type of sample [liquid blood] so as to avoid any confusion. In normal conditions blood is found in various states like dried blood, clotted blood, liquid blood etc.

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But only liquid blood is taken so as to guide the investigating agencies about the chances of recovery of intact DNA from blood samples. A total of sixteen samples were taken to study.

Material and Methods:

The Blood samples were obtained from blood bank of J.N. Medical College Hospital where they were stored at 4°Centigrade Positive [3]. Zero Time was the time when the first recovery was done which was same or different from time of Sample collection from the subject or scene of crime. Before this time the time was be calculated as negative value i.e.: minus one hour or minus one day etc. 2.5 ml of blood was taken to isolate the DNA. After storage, it was processed for DNA analysis using phenol extraction method [2]. The optical density of isolated DNA was done by spectrophotometer.

Volume of Blood Taken: 2.5 ml

Conc. of DNA (µg/ml) = OD260 X 50µg/ml X Dilution Factor X Total Vol. in ml

Formula: OD260 x conversion factor = µg/ml of nucleic acid

OD260 Unit = 50µg/ml for dsDNA

Conc. = Concentration OD = Optical Density
Nm = Nano-Meter 50 = constant

Dilution Factor (Note: This is a ratio hence do not have unit) = Amount of water or buffer used for dilution / Amount of Sample (Isolated DNA dissolved in TE Buffer **).**

Observations and Discussion:

The yield of DNA decreases with time as lysis of DNA occurs in the samples as a result of enzymatic action so the amount of recovery is expected to be decreased with time. The question

arises is that what is the time [in Indian environment] after which we are unable to recover DNA from these samples. After a period of 12 weeks there is fewer chances to recover DNA from blood samples kept at room temperature. The curve becomes parallel showing some amount of recovery even after 12 weeks

Summery and conclusion:

The recovery of DNA in blood kept at room temperature decreases with time. After one month there is less chance of recovery of intact DNA from these samples. The study shows marked decrease in Recovery of intact DeoxyriboNucleic Acid [DNA] after a period of 1-2 months so we can conclude here is that there are hardly any chances of intact DNA recovery after a period of 2 months in samples found at room temperature by phenol-chloroform method [2] However there are protocols available to recover DNA from blood even after 3-4 months which are not included in our study. In the light of above study it has been concluded that blood samples recovered from scene of crime after about 1-2 months is a waste. The samples must be recovered as early as possible to recover intact DNA from them. The samples must be collected within 1-2 months from scene of crime until and unless the climate is cold enough to increases decay time.

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Table-I

Showing yield of DeoxyriboNucleic Acid [DNA] blood samples at Room temperature over a period of twelve weeks time

Sr. No.	Time	Mean DNA Yield in µg/ml [Mean ± S.D.]	Percent Decrease in DNA Yield
1	Zero	1027.35±361	
2	One Week	686 ± 309	33
3	Two Weeks	608 ± 158	41
4	Four Weeks	385 ± 151.5	62.5
5	8-10Weeks	378 ±147.75	63.2
6	11-12Weeks	299 ± 59.58	71

Figure I

DeoxyriboNucleic Acid [DNA] recovery at Room Temperature for different incubation period to see effect of Ageing on the intact DNA recovery