



## DEVELOPMENT OF AN OPTIMIZED PROTOCOL FOR THE EXTRACTION OF DNA FROM HUMAN BLOOD

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Extraction of DNA from suitable biological samples is considered to be the most primary and the most crucial step in many research studies related to genetics and molecular biology. Having considered human research, blood is a very typical tissue from which DNA is extracted to be used in a variety of studies involving the application of different techniques in molecular biology. Therefore, it is important that highly optimized protocols are made available in facilitating the extraction of DNA from human blood to ensure the smooth conducting of studies and to generate precise and accurate results. The specific objective of this study was to develop an optimized protocol for the extraction of DNA from human blood which is of sufficient quantity and optimum purity that could be used in further downstream processes and applications with more precision and reliability. DNA was extracted from human blood with the use of FlexiGene<sup>®</sup> DNA extraction kit by QIAGEN<sup>®</sup>. Even though the protocol provided with the commercial kit was followed unchanged, spectrophotometric absorbency results revealed that the DNA extracted did not have sufficient purity required for molecular cloning as it especially consisted of organic compound contamination. The  $A_{260}/A_{230}$  ratio for DNA which needs to be in the range 1.8-2.0 was detected to be only in the range 1.3-1.4. Eventually, the washing step with ethanol to remove the previously added 100% isopropanol to pellet out DNA from other impurities was repeated by doubling the volume of ethanol mentioned in the original protocol and the air-dry time to remove all the ethanol that was used for washing impurities was increased by nearly six times compared to that mentioned in the original protocol, from 5 minutes to 30 minutes. Having done the above modifications, the  $A_{260}/A_{230}$  ratio improved drastically from 1.4 to 2.2. In addition, the protein digestion to remove all the protein contamination happened optimally which is validated by the fact that the  $A_{260}/A_{280}$  ratio obtained was 1.73, which is very close to the ideal value for DNA which is 1.8. The size of the DNA pellet formed was also improved by increasing the amount of 100% isopropanol added, from 100  $\mu$ L to 150  $\mu$ L in order to effectively pellet out the DNA. Accordingly, the concentration of the DNA that yielded increased.



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The optimized protocol for the extraction of DNA was developed this way as an initial step of the study by modifying the original protocol.

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