

EVALUATION OF THE ALUQUANT™ HUMAN GENOMIC DNA QUANTITATION SYSTEM

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Human forensic DNA analysis involves the quantitation of extracted DNA prior to amplification. Currently, the amount of DNA in an extract is determined by hybridizing a higher-primate specific probe to DNA that has been immobilized onto a membrane. This slot blot procedure has been the predominant method used for pre-amplification DNA quantitation in forensic DNA analysis. Slot blot hybridization provides an estimate of the DNA contained within an extract based on a visual comparison of band intensities between DNA from sample extracts and a series of DNA standards. However, visual evaluation of band intensities can be subjective. In contrast, the AluQuant™ system provides an analytical method for quantitating human (or higher primate) DNA. In addition, unlike slot blot methods, the AluQuant™ process is amenable to automation.

As an alternative approach to human-specific DNA quantitation, the AluQuant™ system uses a unique technique based on the polymerase-catalyzed depolymerization of a probe hybridized in solution to repeated human DNA sequences. Depolymerization liberates dNTPs, which are used to generate ATP. Subsequently, ATP is utilized by Luciferase to produce a light signal that is proportional to the amount of human DNA present. The values obtained from the AluQuant™ Human DNA Quantitation system using a luminometer are compared to a standard curve and converted to DNA concentrations. These values can then be used to set up amplification reactions.

Several laboratories in the United States participated in a study of the AluQuant™ system for use in forensic casework to quantitate human genomic DNA samples. We show that the AluQuant™ system is compatible with several methods of DNA isolation from different substrates. Also, degradation of DNA and large excesses of non-human DNA do not interfere with the quantitation.