Estimation of DNA Fragments from PCR Amplification of Microbiome Samples

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ABSTRACT

Polymerase chain reaction (PCR) is a technique that is often incorporated in the workflow of preparing biological samples for sequencing analysis and DNA identification. It is used to amplify fragments of DNA extracted from samples. Using a binomial model for DNA fragment counts, we examine whether initial bacterial load can be determined from counts at end-point of PCR amplification. We present theoretical derivations of the mean and the second moment of the count data based on our model. Several simulation studies are used to compare initial counts of DNA fragments with the end-point of PCR amplification. Our results show that the mean and the second moment of the counts from the end-point of PCR amplification cannot be used to estimate the initial counts of DNA fragments. Also, amplifying samples until saturation result in equal end-point parameters for all samples even when their initial counts differ, except for the second moment which depends on the coefficient of variation of initial counts.