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Comparison Between a Droplet Digital PCR and Real Time PCR Method in Quantification of HBV DNA

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Abstract : HBV infection causes a potential serious public health problem. The ability to detect the HBV DNA concentration is of the importance and improved continuously. By using quantitative Polymerase Chain Reaction (qPCR), several factors in standardized; source of material, calibration standard curve and PCR efficiency are inconsistent. Digital PCR (dPCR) is an alternative PCR-based technique for absolute quantification using Poisson's statistics without requiring a standard curve. Therefore, the aim of this study is to compare the data set of HBV DNA generated between dPCR and qPCR methods. All samples were quantified by Abbott's real time PCR and 54 samples with 2 -6 log10 HBV DNA were selected for comparison with dPCR. Of these 54 samples, there were two outlier samples defined as negative by dPCR. Of these two, samples were defined as negative by dPCR, whereas 52 samples were positive by both the tests. The difference between the two assays was less than 0.25 log IU/mL in 24/52 samples (46%) of paired samples; less than 0.5 log IU/mL in 46/52 samples (88%) and less than 1 log in 50/52 samples (96%). The correlation coefficient was r=0.788 and P-value <0.0001. Comparison to qPCR, data generated by dPCR tend to be the overestimation in the sample with low HBV DNA concentration and underestimated in the sample with high viral load. The variation in DNA by dPCR measurement might be due to the pre-amplification bias, template. Moreover, a minor drawback of dPCR is the large quantity of DNA had to be used when compare to the qPCR. Since the technology is relatively new, the limitations of this assay will be improved.

Keywords : hepatitis B virus, real time PCR, digital PCR, DNA quantification
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