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Comparatively rapid screening tests for diagnosis of hepatitis B virus infection using loop-mediated isothermal amplification (LAMP) paired with lateral flow dipstick (LFD), gold nanoparticles (AuNPs) and real-time turbidimetry

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Abstrak

ABSTRACT
Hepatitis B virus (HBV) infects hepatocytes and causes acute and chronic hepatitis that can lead to cirrhosis and hepatocellular carcinoma (HCC) in both animals and humans. Early detection of HBV infection assists in monitoring the patient's response to anti-HBV therapy, blood donation screening, and disease management, control and eradication. This research focused on development of LAMP assay combined with lateral flow dipstick (LFD), gold nanoparticle (AuNPs) and real-time turbidimetry for screening of the hepatitis B virus. Analytical sensitivity, analytical specificity, diagnostic sensitivity, diagnostic specificity, accuracy and predictive value of each technique were determined and compared to conventional PCR and real-time PCR (gold standard method). The analytical sensitivity of LAMP-LFD and LAMP-AuNPs was 1.24x101 copies /mL, LAMP-real-time turbidimetry was 1.24x102 copies/mL, while that of conventional PCR was 1.24x104 copies/mL. Examination of the analytical specificity of all LAMP-based combinations and conventional PCR showed no cross-reactivity with HCV or human plasma. Upon exploration of one hundred unknown samples, in comparison to real-time PCR, the diagnostic sensitivity and specificity of LAMP-based assays were 100% and 90%, respectively. The accuracy, positive predictive value (PPV) and negative predictive value (NPV) of the LAMP-based assays were 98%, 97.56%, and 100%, respectively. While that of conventional PCR were 60%, 100%, 68%, 100% and 38% of diagnostic sensitivity, diagnostic specificity, accuracy, PPV and NPV, respectively. LAMP-based assays need to be simplified in terms of achieving single-step diagnosis using one master mix solution that is suitable for a point-of-care diagnostic test.