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Development of different diagnostic technologies and platforms for Vector borne diseases

Prof. Qian Han
Department of Biological Sciences School of Life and Pharmaceutical Sciences Hainan University

Abstract

Generally, laboratory diagnosis of vector borne diseases (VBD) can be divided into two categories: direct methods (microscopy, culturing of the causative agent, nucleic acid detection, etc.) and indirect methods (e.g., detection of organism-specific immune responses). Many of the diagnostic tools based on molecular techniques are applied for the finest detection and characterization of VBDs.

We have developed different nucleic acid based diagnostic technologies and platform for the quick and efficient diagnosis of VBDs.

1. **Multiplex PCR for the diagnosis of *Ehrlichia, Babesia, Hepatozoon* and *Mycoplasma* infections:** Several new strategies for multiplex PCR have increased both multiplexing and high-throughput capabilities of detection techniques. When compared with traditional ELISA and PCR, multiplex assays have a number of advantages, including high throughput multiplex analysis, less sample volume and size requirements, efficiency in terms of time and cost. Here, we developed this diagnostic technique which employs the multiplexing methodology for the detection of 4 different pathogens in one single reaction tube. This is standardized for its efficiency, specificity and sensitivity for the diagnosis of multiple pathogens at a time. Unlike the singleplex PCR, the multiplexing technique saves reaction time, less sample handling and also reduces the cost per reaction.
2. **NanoPCR for VBDs.**: This simple technology employs the use of the characteristic ZnO nanoflowers incorporated in our conventional PCR assay, leading to a more efficient, specific, sensitive, cost effective and a quick diagnostic tool. The major highlights of the ZnO nanoflowers based PCR are its turnaround time and high efficiency and quality without compromising on the yield and quality of the DNA. The synthesis of these ZnO nanoflowers is a hassle free method and can be conducted even in the resource limited settings.
3. **Smartphone assisted portable loop mediated isothermal amplification box for diagnosis of VBDs:** Loop-mediated isothermal amplification (LAMP), an isothermal DNA amplification (DNA amplification at a constant temperature without the need of a thermal cycler), caters for several infectious diseases mainly in poor and underdeveloped countries, because of its low cost and relatively user friendly procedure. We developed an efficient and sensitive detection technology for detection of *Ehrlichia* and *Hepatozoon* by linking the LAMP assay to our smartphone by using a simple, inexpensive and portable "LAMP box", altogether connected to each other wirelessly. This LAMP box is made up of an isothermal heating pad which serves as a platform for the reaction tubes and LAMP assay. The entire set up can be connected to the smartphone via an inbuilt Wi-Fi which allows the user to establish and leads to the simple graphical user interface (GUI) to control the LAMP box. A 5V USB power source was used to supply power to the box. The sensitivity of the LAMP assay was estimated to be up to 10^{-6} dilution limit. Our smartphone enabled LAMP box can allow the testing of 5- 6 samples at a time. The developed platform can not only be used for the diagnosis of canine infectious diseases like *Ehrlichia* and *Hepatozoon*, but can also serve as an efficient diagnostic platform for many other infectious diseases and majorly towards field-based diagnostics.

Biography:

Prof. Qian Han, Department of Biological Sciences School of Life and Pharmaceutical Sciences Hainan University 58 Renmin Road, Haikou, Hainan 570228, China.
Email: qianhan@hainanu.edu.cn
Contact Number: 18689881295

Bottom Note:

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