Biotechnology Advances 33 (2015) 178–190

Contents lists available at ScienceDirect

Biotechnology Advances

journal homepage: www.elsevier.com/locate/biotechadv

Research review paper

Recent advances in micro/nanotechnologies for global control of hepatitis B infection

U. Hakan Yildiz [a], Fatih Inci [a], ShuQi Wang [a], Mehlika Toy [b], H. Cumhur Tekin [a], Asad Javaid [c], Daryl T.-Y. Lau [c], Utkan Demirci [a,*]

[a] Demirci Bio-Acoustic-MEMS in Medicine (BAMM) Laboratory, Stanford University School of Medicine, Canary Center at Stanford for Cancer Early Detection, Palo Alto, CA 94304, United States
[b] Department of Surgery, Stanford School of Medicine, Palo Alto, CA, United States
[c] Department of Medicine, Beth Israel Deaconnes Medical Center, Harvard Medical School, Boston, MA, United States

ARTICLE INFO

Article history:
Received 21 August 2014
Received in revised form 3 November 2014
Accepted 4 November 2014
Available online 20 November 2014

Keywords:
Nanotechnology
Hepatitis B
Screening
Diagnosis
Microfluidic
Point-of-care
Nanobiotechnology
Plasmonics
Graphene

ABSTRACT

The control of hepatitis B virus (HBV) infection is a challenging task, specifically in developing countries there is limited access to diagnostics and antiviral treatment mainly due to high costs and insufficient healthcare infrastructure. Although the current diagnostic technologies can reliably detect HBV, they are relatively laborious, impractical and require expensive resources that are not suitable for resource-limited settings. Advances in micro/nanotechnology are pioneering the development of new generation methodologies in diagnosis and screening of HBV. Owing to combination of nanomaterials (metal/inorganic nanoparticles, carbon nanotubes, etc.) with microfabrication technologies, utilization of miniaturized sensors detecting HBV and other viruses from ultra-low volume of blood, serum and plasma is realized. The state-of-the-art microfluidic devices with integrated nanotechnologies potentially allow for inexpensive HBV screening at low cost. This review aims to highlight recent advances in nanotechnology and microfabrication processes that are employed for developing point-of-care (POC) HBV assays.

Contents

* Corresponding author at: Demirci Bio-Acoustic-MEMS in Medicine (BAMM) Laboratory, Stanford University School of Medicine, Canary Center at Stanford for Cancer Early Detection, 3155 Porter, Palo Alto, CA 94304, United States.
E-mail address: utkan@stanford.edu (U. Demirci).

http://dx.doi.org/10.1016/j.biotechadv.2014.11.003
0734-9750/

Introduction

Hepatitis B virus (HBV), the etiological agent of hepatitis B, can cause chronic hepatitis and lead to liver failure, cirrhosis and hepatocellular carcinoma (HCC), which accounts for 1 million deaths annually worldwide (Dienstag, 2008). The chronic HBV infection prevalence varies greatly in ethnicity and region in East and Southeast Asia, Western Pacific, and Sub Saharan Africa, it exceeds 8%, whereas in North America and Western Europe (Lavanchy, 2004) lesser than 1% of the population is infected. Although vaccination reduces occurrence rate of chronic HBV infection (Zanetti et al., 2008), an antiviral therapy emerges as a sole option for control and prevention of progression of disease (Hoofnagle, 2006). Suppression of HBV replication by antiviral drugs may reduce the incidence of cirrhosis to less than 1% year, and decrease the rate of HCC in patients with advanced fibrosis or cirrhosis by 50% (Liaw et al., 2004). In the United States, for example, only 20–30% of CHB carriers know their status, and only 12.5% of CHB carriers who are eligible for treatment under the guidelines receive it (Cohen et al., 2011). An important problem of prolonged therapy with the first generation nucleos(t)ide analogs is the occurrence of drug resistance, which may negate the therapeutic benefit. This problem can now be addressed by using 3rd generation drugs with minimal resistance in treatment-naïve patients (Chien and Liaw, 2008). The well-established serological and nucleic acid screening assays have been successfully implemented for diagnosis and screening, where the infrastructure is available. Despite the availability of HBV testing and potent drugs with minimal resistance in developed countries, access to diagnostics, treatment, and monitoring remains limited in developing countries due to resource constraints and lack of infrastructure. The primary motive of emerging diagnostic technologies remains to eliminate such limitations and facilitate HBV testing that leverages portable, inexpensive, but sensitive and specific diagnostic technologies.

Advances in nanotechnology and emergence of nanomaterials with exceptional electrical, optical and mechanical properties provide invaluable opportunities in developing new generation methodologies for HBV testing. Due to response capability of nanomaterials to minor stimulus, signal transduction at the molecular level is enabled that improves limit of detection and sensitivity by several orders of magnitudes with respect to traditional sensors. The combination of nanomaterials (metal/inorganic nanoparticles, carbon nanotubes) with microfabrication technologies also renders miniaturized sensors for rapid sensing of HBV and other viruses from ultra-low volume of biological samples. HBV nanobiosensors often rely on generalized approach "utilization of biomarker HBsAg" and "interaction with HBV" and subsequent transduction of the binding event to a detectable signal. In contrast to traditional serological methodologies used in virus (or nucleic acid) detection, nanomaterials such as gold nanoparticles provide an ultra-high surface area enabling immobilization of a number of biomarkers and inducing substantial change in plasmonic properties even in the presence of few target molecules. By the advancement of bioconjugation techniques, application of nanomaterials in complex bioassays such as HBV and HIV detection, are profoundly simplified, which lowers the assaying time and cost. The demand to develop fast, easy, preferably inexpensive detection strategies makes nanomaterials an attractive and indispensible component of HBV diagnostics for POC testing that enables quantification, ultra-sensitivity, and on-board-signal-amplification. Such an ideal detection platform can be potentially realized by leveraging several different cutting edge technologies and high performance nanomaterials. Fulfilling this highly demanding task requires, i) understanding the origin and nature of HBV infection, ii) resolving multistep biological mechanisms of infection and transfection, and iii) exploring sensing capability of nanomaterials such as gold nanoparticles, carbon nanotubes or graphene.

The purpose of this review is to highlight recent advances and future perspectives in nanotechnology and microfabrication processes that are employed for developing HBV detection platforms and assay methodologies. First, we present insights into the natural history, epidemiology and treatment of HBV. Next, we discuss monitoring of therapy, cancer relationship and co-infections as well as the public health aspects such as screening, diagnostics, cost-effective tools and assays for HBV. Last advances in nanotechnological tools for diagnosis of HBV and HBV co-infections are discussed in detail highlighting the impact of nanotechnology and nanomaterials on developing novel HBV detection platforms and testing methodologies.

General description of hepatitis B virus and natural history

Hepatitis B virus is a DNA virus in the family of Hepadnaviridae (Tacke et al., 2004). The whole genome of HBV is a complex consisting of four partially protruding reading frames that encode entire viral structure (Tacke et al., 2004). HBV enters into the hepatocyte upon endocytosis process, followed by uncoating, and formation of covalently closed circular DNA (ccc DNA) in nucleus that serves as a viral replication template (Doo and Liang, 2001; Tacke et al., 2004). The replication of HBV is asymmetric by transcription of RNA. Due to lack of proofreading activity of its transcriptase (Pol/Rt), estimated mutations are at rate of one error/10^4–10^5 nucleotides daily (Locarnini and Mason, 2006). It is considered that random mutations may overlap with the antiviral-induced mutations and result in drug-resistant strains. Permanent elimination of HBV is extremely low or not achievable since virus capable of establishing persistent reservoir in the form of ccc DNA in hepatocytes (Delaney, 2013).

Epidemiology

Human body is a sole reservoir for HBV. However, the high replication activity of HBV leads to production of high concentration of viral particles circulating/present in blood and body fluids of infected person, and therefore, makes hepatitis B virus extremely transmissible. Most people become chronically infected at childbirth when the mother is a hepatitis B carrier (vertical transmission), while others become infected by close personal contact (infancy, unprotected sex) or by injections (medical and dental instruments or intravenous drug use) (horizontal transmission). The horizontal transmission risk of chronic of HBV infection incidence is 30% to 50% for the age group between birth and 5 years of age, decreases dramatically to 7–10% by aging. (McMahon, 2009a).

There are eight genotypes of HBV and their distribution varies in different regions of the world. Genotype A is found most frequently in North America, but it is also present globally. In Asia, genotypes B and C are dominant. Genotype D is prevalent in South Asia, Middle East and Southern Europe. Genotype E is frequent in sub-Sahara Africa while Genotype F is mostly found among Native Americans. Genotypes G and H are relatively uncommon (Fig. 1), (Magnius and Norder, 1995). It is generally accepted that HBV genotype and subgenotype are the certain variables to predict outcome of chronic HBV infection (McMahon, 2009a). The cross-sectional studies conducting the examination of relation of genotypes C and B to HCC incidence reveal that genotype C has a potential impact on increasing risk of HCC (Lee et al., 2003) (Fig. 1).

Chronic hepatitis B

Chronic hepatitis B (CHB) is described as necroinflammation in liver originating from prolonged presence of HBV, which is diagnosed by the persistent serum hepatitis B surface antigen (HBsAg) for 6 months or longer (Hollinger and Lau, 2006). The natural stages of CHB are classified as four major clinical phases based on levels of serum aminotransferase (ALT) and hepatitis B virus DNA (HBV DNA), presence of hepatitis Be antigen (HBeAg), and suspected immune status (McMahon, 2009b). These phases are: 1) Immunotolerant phase, 2) immunoelimination or