Biosensors and Bioelectronics 102 (2018) 256–266

Contents lists available at ScienceDirect

Biosensors and Bioelectronics

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

Paper-based fluorogenic devices for in vitro diagnostics

Meirong Wu[a], Qiongyu Lai[a], Qiang Ju[a], Lin Li[a,*], Hai-Dong Yu[a,**], Wei Huang[a,b,**]

[a] Key Laboratory of Flexible Electronics (KLOFE) & Institute of Advanced Materials (IAM), Jiangsu National Synergetic Innovation Center for Advanced Materials (SICAM), Nanjing Tech University (Nanjing Tech), 30 South Puzhu Road, Nanjing 211816, PR China

[b] Key Laboratory for Organic Electronics and Information Displays & Institute of Advanced Materials (IAM), Nanjing University of Posts & Telecommunications, 9 Wenyuan Road, Nanjing 210023, PR China

ARTICLE INFO

Keywords:
Paper-based devices
Fluorogenic detection
In vitro diagnostics
Point of care
Bioassays

ABSTRACT

In vitro diagnostics (IVD) are essential in healthcare systems for detection of diseases, conditions, or infections. Affordable and sensitive methods remain a challenge in the development of IVD. In this article, we give an overview of paper-based fluorogenic devices, an emerging cost-effective analytical method for IVD applications. The paper-based devices are inexpensive, sensitive, selective, user-friendly and equipment-free. Furthermore, the fluorescent detection on paper-based devices has recently attracted enormous attentions due to its high sensitivity and selectivity. We summarize and compare various fluorescent materials that are used in the paper-based fluorogenic devices, including fluorescent dyes, quantum dots, metal nanoclusters, upconversion nanoparticles, and carbon dots. We review a wide range of IVD applications of the paper-based fluorogenic devices, e.g., detection of nucleic acids, proteins, cells, and so on. It may open an avenue to improve the global healthcare systems in the developing countries at point of care settings and in emergency situations.

1. Introduction

In vitro diagnostics (IVD), which are primarily used for detecting targets in samples such as blood, urine, sweats, saliva, interstitial fluid and tissue, are of critical importance in the healthcare systems (Ehrenberg and Ault, 2005). The global IVD market is anticipated to reach $74.46 billion by 2022 (Yetisen et al., 2013). The trend of the development of IVD includes: (1) automation and integration of the equipment and reagents; (2) device miniaturization and point of care; (3) analysis on molecules and to be personalized (Ali et al., 2009). IVD promote the efficiency of diagnostics and enable possible applications in remote settings, which are of particular importance for rapid on-site diagnosis and reducing biological risks of infectious diseases.

Among current IVD technologies, some advanced ones such as polymerase chain reaction (PCR) and enzyme-linked immunosorbent assay (ELISA) have already been implemented in developed countries; however, they cannot be widely used in developing countries because of limited availability of laboratory infrastructure, skilled personnel, and financial supports (Ozcan, 2014). The World Health Organization defined that diagnostics for developing countries should be ASSURED: affordable, sensitive, specific, user-friendly, rapid and robust, equipment free, and deliverable to end-users (Peeling et al., 2006). The rapid and cost-effective detection based on paper-based devices is emerging as a new high-efficiency analytical method to meet this demand (Choi et al., 2015).

The paper-based devices integrated the advantages of the paper with those of the microfluidics, i.e. they are cheap, portable and easy to fabricate, require little sample, enable multiplexed and quantitative analysis, and stable to a wide range of temperature and time (Zhang et al., 2017). Commonly used detection methods for paper-based devices include colorimetry (Shen et al., 2012b; Su et al., 2015a, 2015b; Vashist et al., 2015; Zhou et al., 2014), electrochemistry (Delaney et al., 2011; Dungchai et al., 2009; Ge et al., 2012b), chemiluminescence (Ge et al., 2012a; Liu et al., 2014; Yu et al., 2011), electrochemiluminescence (Li et al., 2014a; Mani et al., 2013), photoelectrochemistry (Ge et al., 2013; Sun et al., 2014a; Wang et al., 2015c), fluorescence (Noor et al., 2013; Velu et al., 2015; Wang et al., 2016; Yamada et al., 2015), etc. The fluorescent detection on paper-based devices has recently attracted increasing attentions, and shown unique advantages for many applications including IVD of cancer (Yildiz et al., 2013), infections (Dou et al., 2014), and other physiology diseases index (Wang et al., 2015a). In this article, we will review recent advances of the paper-based fluorogenic devices and their applications for IVD. We will also outlook the future research direction and envisage more potential applications by integrating the paper-based fluorogenic devices with other emerging technologies.

* Corresponding author.
** Co-corresponding authors.
E-mail addresses: iamlli@njtech.edu.cn (L. Li), iamhdyu@njtech.edu.cn (H.-D. Yu), iamwhuang@njtech.edu.cn (W. Huang).

https://doi.org/10.1016/j.bios.2017.11.006
Received 28 July 2017; Received in revised form 10 October 2017; Accepted 1 November 2017

1.1. Paper-based devices

The paper-based devices are fabricated through creating hydrophilic channels confined by hydrophobic walls on paper (Liana et al., 2012). The advantages of paper substrates in comparison to plastic plates include that they are (1) affordable, (2) user-friendly, (3) ubiquitous, (4) sensitive, (5) biocompatible, (6) easy to fabricate, store, stack, and transport, and (7) require less sample and reagents for conducting assays (Ju et al., 2016). The paper-based devices with hydrophilic/hydrophobic micro-channel networks enable fluid handling and quantitative analysis for their potential applications in medicine, healthcare and environmental monitoring (Hu et al., 2014).

The movement of aqueous fluids within the paper-based devices mainly rely on capillary force, in both horizontal and vertical dimensions depending on various assay designs. The paper-based devices include two-dimensional (2D) (Dutta et al., 2016; Kaushik et al., 2016; Zhang et al., 2016; Zhao et al., 2016) and three-dimensional (3D) (Ariza-Avidad et al., 2016; Fischer et al., 2016; Liu and Crooks, 2011; Zhang et al., 2013) devices. The 2D paper-based devices are fabricated by patterning physical or chemical hydrophobic boundaries to form micro-channels on paper. 3D paper-based devices are fabricated by stacking or folding alternating layers of paper. The 3D devices can distribute fluids within layers of paper and between adjacent layers of paper. Using 3D devices, it is possible to distribute samples from a single entry point into multiple test spots where assays can take place, enabling multiplexed diagnostics. Both 2D and 3D paper-based devices can serve as a substrate for filtering samples, performing chromatographic separations, and taking biochemical reactions provided by the different types of paper to be combined into a single device. Current methods for fabricating paper-based microfluidic devices include (1) wax printing, (2) inkjet printing, (3) photolithography, (4) flexographic printing, (5) plasma treatment, (6) laser treatment, (7) wet etching, (8) screen-printing, and (9) wax screen-printing (Chen et al., 2015; Parolo and Merkoci, 2013; Zhang et al., 2015). Coupled with different fabrication methods and functional diagnostic equipment to fabricate miniaturized portable medical tools, the paper-based devices will gain many new developments as IVD platforms (Li et al., 2015a).

1.2. Detection methods based on paper-based devices

The response signal from paper-based diagnostics can be detected by various detection methods including colorimetry, electrochemistry, chemiluminescence, electrochemiluminescence and fluorescence.

a) Colorimetric detection is generally carried out based on the color changes resulting from the aggregation and/or separation of nanocrystals induced by chemical/biochemical reactions between target analytes and colorimetric probes. It is the simplest analytical detection technique for paper-based devices, because it does not rely on the bulky off-chip detection system and can be used to identify the analytes by naked eyes. However, it is difficult for this method to provide the quantitative conclusion, and its application also face the limitations caused by the diversity of people's eyes, background and/or noise, etc (Yetisen et al., 2013).
b) Electrochemical detection involves interaction of chemical species with electrodes or probes to obtain electrical signals (potential or current). Besides rapid and specific, the most attractive feature is that it enables a digital readout (Rackus et al., 2015). However, the readout of electrical signal requires configured equipment which is not portable. Other disadvantages include cumbersome electrode handling processes, frequent calibration, and electrode fouling problems (Dungchai et al., 2009).
c) Chemiluminescence (CL) assay for the detection of analytes is based on the substrate chemical reactions to cause photochemical emission, which neither needs the help of an enzyme, nor requires excitation light source and emission filters. Reagents for chemiluminescence are typically inexpensive, and the measurement is highly sensitive, making it very attractive (Delaney et al., 2011).
d) Electrochemiluminescence (ECL) in paper-based devices is generally initiated by chemical and electron transfer reactions with co-reactant reagents at the surface of the working electrode under a specific applied potential, thus it can provide highly selective detection in a wide concentration range. The generation of electrochemiluminescence signals needs an outer equipment like an electrochemical workstation and a chemiluminescence analytical testing system (Bertoncello and Forster, 2009).
e) Fluorescent detection is another kind of optical methods. Emitting fluorescence is a key aspect to the design of analytical devices, whether quantifying (semi-quantifying) the concentrations of analyses or determining the mere absence/presence of analyses. Fluorescent detection has recently attracted enormous interests for paper-based diagnostics. We will review recent advances of the paper-based fluorogenic devices and their applications for IVD in the following sections.

2. Fluorescent materials for paper-based devices

In recent years, many examples of fluorescence integrated with paper-based devices have been applied to the detection of bacteria, proteins (Yan et al., 2015), biomarkers (Xu et al., 2016), heavy metals and others that can cause disease on human (Li et al., 2014b). The paper-based fluorogenic devices have the following advantages: (1) high sensitivity, which can reach one over one hundred million; (2) fluorescent probes offer a wide wavelength range, Stokes' shift and spectral bandwidth that meet specific requirements for a variety of applications, making fluorescent probes useful with high selectivity; (3) the fluorescent detection can be used in qualitative/quantitative analysis because it is determined from the fluorescence intensity; (4) the fluorescent detection has a dynamic wide linear range; (5) the operation process is simple; (6) the fluorescent detection has good reproducibility; (7) the fluorescent detection uses little amount of sample and reagent; (8) and the fluorescent detection requires a simple equipment (Qian et al., 2016; Wolfbeis, 2015).

Fluorescence is a form of luminescence. The Stokes' shift leads to the facility for separating symmetrical excitation and emission spectra (Fig. 1a) (Medintz et al., 2005). Perrin-Jablonski diagram illustrates the generation of fluorescence in one-photon absorption (Fig. 1b). The molecules or atoms are initially stable at the ground state (S_0). Upon absorption of a photon with energy quantitatively equal to the energy gap between the lowest level of the ground state and one vibrational level of first/second excited state (S_1/S_2), the molecules are excited to the excited state. Molecules at excited state are not stable and will easily release excess energy in the form of photon (fluorescence) and/or non-radiative relaxation (non-fluorescence).

As the majority of molecules don't exhibit fluorescence in biological systems, the measurement of the non-fluorescent compounds using a fluorescent assay would require the use of "fluorescent probes". Fluorescent materials used on paper-based devices including fluorescent dyes, quantum dots, metal nanoclusters, upconversion nanoparticles, and carbon dots will be discussed and compared in Table 1.

2.1. Fluorescent dyes

Fluorescent dyes have been widely used as fluorescent indicators in the paper-based devices (Lu et al., 2017). Generally, the fluorescent dyes are normally polyaromatic or heterocyclic hydrocarbons with π-bond system (Fig. 2). In general, small molecule fluorescent dyes have features of small size, high fluorescence intensity, good biocompatibility, and easy modifications (Feng et al., 2016; Long et al., 2016).

However, choosing the appropriate fluorescent dye is relatively difficult since many dyes have drawbacks in low fluorescence quantum yield, weak resistance to photo-bleaching, narrow excitation spectra