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Smartphone-based analytical biosensors

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The traditional analytical biosensor instruments are relatively bulky, expensive, and not easy to handle, thus their applications are largely limited in resource-limited settings. The recent development of microfluidic lab-on-a-chip (LOC) technology has provided a possible solution to miniaturize the conventional biosensing system, yet other accessory devices to detect, readout, analyze, transfer, and display results are still required. With the rapid development, mass production, and pervasive distribution of smartphones in recent years, they have provided people with portable, cost-effective, and easy-to-operate platforms to build analytical biosensors for point-of-care (POC) applications and mobile health. Based on the common analytical methods, this paper reviews the recent development of four types of smartphone based analytical biosensory systems at the POC, *i.e.*, smartphone-based microscopic imaging, colorimetric, electrochemical, and electrochemiluminescence biosensor. The different bio-sensing strategies and analytical performance together with future perspectives are discussed.

1. Introduction

The traditional analytical biosensing instruments are relatively bulky, expensive, and not easy to handle, which greatly limit their applications in biomedical diagnosis, especially in resource-limited settings. For example, the flow cytometer is a widely used clinical diagnostic instrument for cell analysis and sorting, which is the current gold standard method to count CD4+ T-cells for HIV/AIDS monitoring.[1,2] Yet, it has the drawbacks of prohibitively high cost, bulky size, and tedious measurement procedures.[3–5] Another gold standard biomedical diagnostic technique, optical microscopy, has been used for a long time for accurate disease monitoring and discovering its underlying causes through high-resolution cells/microorganisms imaging.[6–8] However, to reach highly sensitive measurements and sensing, it relies on bulky and expensive optics as well as professional personnel to operate in established hospitals or clinics. These problems pose significant healthcare challenges especially in low-income and resource-limited areas. Therefore, portable and affordable analytical and biomedical sensing instruments are in high-demand to provide point-of-care (POC) diagnostics.[8–12]

POC diagnostics refer to the diagnosis at the patient site outside a clinical laboratory by a clinician or the patient him/herself. The diagnostics are usually portable and easy-to-operate with short time-to-result so that rapid clinical decision can be made and treatment delay can be alleviated.[13–15] To miniaturize the traditional bulky biosensing system, the recent advancement of microfluidic lab-on-a-chip (LOC) technology has provided a possible solution to integrate biosensors, actuators, microfluidics, and micro-electro- mechanical systems (MEMS) all together on a miniaturized chip-scale platform.[16–22] However, to practically apply such a miniaturized biosensor LOC in clinical biomedical diagnostics, most biosensor LOCs still require other accessory devices to detect, readout, analyze, transfer, or display results. To address such challenges, researchers are recently turning into the smartphone-based analytical biosensors.[23–28]

Smartphones initially came to the consumer market in the late 90s, which turned the conventional feature-phones into hand-held computers. Then after the revolutionary launch of Apple's iPhone in 2007, smartphones quickly became a mainstream popularity. With the continuous increasing penetration rate of smartphones, the number of smartphone users is predicted to increase from 2.1 billion in 2016 to around 2.8 billion in 2019, more than one-third of the world's total population.[29] Typically, a smartphone is equipped with numerous types of sensors such as accelerometer and thermometer to sense data for various purposes, in which the most widely used sensor by consumers is probably the optical camera

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sensor. The smartphone usually also possesses fast multicore processor, large memory, adequate battery, audio and USB port, and touch screen, to provide powerful computation capability, large data storage, portable power supply, and convenient user interface to interact with external, respectively. The wireless data transfer methods such as 4G cellular data service, Bluetooth, Wi-Fi, and near-field communication (NFC) can also enable the acquired smartphone data to be transmitted to professionals or cloud, allowing data management for local medical facilities in remote emergencies.

As the capabilities of smartphones are becoming more and more powerful, researchers only need to design certain accessory attachments on smartphones for analytical biosensing, which can provide illumination sources, signal detectors, or just instrumental interfaces that supply power and process data.[26] Considering such facts of rapid development, mass production, and pervasive distribution for smartphones in recent years, they have provided people with portable, cost-effective, and easy-to-operate platforms to integrate with microfluidics and LOC to build analytical biosensors for POC applications and mobile health. So far, the effectiveness of smartphone-based analytical biosensors has already been demonstrated in numerous applications such as healthcare diagnostics, environment monitoring, and food toxin screening.[30–37]

Therefore, based on the common analytical methods, this paper reviews the most recent development of four types of smartphone-based analytical biosensor systems at the POC, *i.e.*, smartphone-based microscopic imaging sensor, colorimetric sensor, electrochemical sensor, and electrochemiluminescence sensor. The different bio-sensing strategies, analytical performance, and applications are discussed to provide a most recent overview of the smartphone-based analytical biosensor development, with conclusions and future perspectives at the end. Note that this classification is not intended to completely cover all the existing types on smartphone-based biosensors, but mainly considers the wide and common applications of these four biosensing types in the existing literature.

2. Smartphone-based microscopic imaging sensor

Nowadays, almost all smartphones must have a camera on the front and back side. Dual cameras are gradually becoming a new trend due to the market competition for better focusing performance and dynamic range in photography. Inside each optical camera is actually a Complementary Metal Oxide Semiconductor (CMOS) image sensor (CIS), which typically includes a photodiode pixel array and peripherical readout circuits to convert photons (optical images) into electronic signals.[38,39] As the integrated circuit design and fabrication techniques keep advancing, the pixel array size of the smartphone embedded CIS has now increased beyond 40-Mega, meanwhile the pixel pitch has shrunk to around 1 μm.[40] The recent development of back-side illumination (BSI) pixel architecture instead of the conventional front-side illumination (FSI) architecture also enables deep photodiode and short optical path to improve the sensitivity and dynamic range.[41] Other advantages of CIS also include high framerate (due to high-speed readout circuits), low power consumption, on-chip ISP (image signal processor), and scalability for larger pixel array. With the high-resolution imaging capability of CIS, smartphone-based optical image sensing has been the most commonly applied analytical modality, especially in the microscopic imaging application.[42]

Moreover, the rapidly advancing computational power of the embedded Central Processing Unit (CPU) and Graphics Processing Unit (GPU) in smartphone enables the captured images to be analyzed, corrected, and improved in a real time fashion, thus a computational microscopy without the need for external processing on PC can be realized. By attaching external accessories such as optical components, sample tray, and light emitting diode (LED) illumination source, the smartphone-based microscopy can detect the morphological features of samples such as size, shape, brightness, and color, and use them as signal labels for the detection of blood cells, tumor cells, microorganisms, nanoparticles, DNA, and virus.[42]

A. Bright-field microscopic imaging biosensor

(1) Lens-based approach. Like a conventional light microscope, smartphone-based microscopic imaging sensor also has two common sensing modalities, *i.e.*, bright-field and fluorescent. The bright-field microscopic imaging biosensor is discussed first.

Some early attempts built the smartphone-based microscopic sensor on the feature phones when the smartphone was still not very popular. For example, in 2009, Breslauer *et al.* presented a portable smartphone-based microscope as shown in Fig. 1(a), which mounted standard microscope eyepieces and objectives on the camera sensor to adjust magnification and resolution.[43] The system spatial resolution was measured as 1.2 μm and the field of view (FOV) was 180 μm diameter. The system demonstrated successful imaging of P. falciparum-infected red blood cells and *M. tuberculosis*-infected sputum samples in bright field and fluorescence mode, respectively. Ever since, numerous smartphone-based microscopic sensors have been proposed.

The utilization of standard microscope lens is still complex and relatively large. In 2011, Z. J. Smith *et al.* proposed one smartphone-based microscopy by using only a 1 mm ball lens as the magnifying element over the smartphone camera system, as shown in Fig. 1(b).[44] Peripheral blood smears were imaged with 1.5 μm resolution achieved. Although the size of lens was reduced, the side effects of significant flatfield distortions and field-dependent magnification emerged for ball lens magnification.

As an alternative approach, N. A. Switz *et al.* attached a reversed mobile phone camera lens to an intact mobile phone to realize a 10 mm^2 FOV and a resolution ≤5 mm imaging as shown in Fig. 1(c).[45] Their prototype achieved the detection of

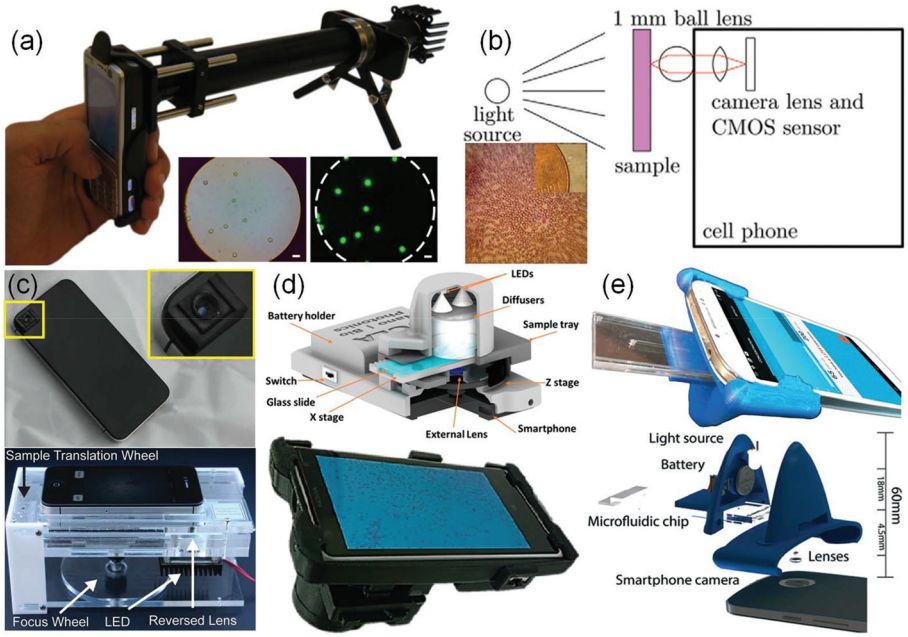

Fig. 1 (a) Mobile phone microscope with standard microscope eyepieces and objectives, reproduced from ref. 43 with permission from the Public Library of Science, copyright 2009; (b) smartphone-based microscopy with a 1 mm small ball lens reproduced from ref. 44 with permission from the Public Library of Science, copyright 2011; (c) smartphone-based microscopy with a reversed mobile phone camera lens reproduced from ref. 45 with permission from the Public Library of Science, copyright 2014; (d) bright field smartphone-based microscope for the detection of Schistosoma haematobium eggs without label, reproduced from ref. 46 with permission from the American Society of Tropical Medicine and Hygiene, copyright 2017; (e) smartphone-based microscopy with a pair of external aspheric lenses for CD4 T-cell detection, reproduced from ref. 47 with permission from the Royal Society of Chemistry, copyright 2017.

red and white blood cells in blood smears as well as soil-transmitted helminth eggs in stool samples. Such approach provides a balance between lens size and imaging quality.

In another recent case, a portable smartphone-based bright field microscope was developed for the detection of *S. haematobium* eggs without label in rural Ghanaian school-based setting, as shown in Fig. 1(d).[46] It employed two white LEDs and an external lens for illumination and magnification to reach a spatial resolution of about 0.87 μm. Two polymer diffusers and a microscope slide tray over an adjustable focal plane were designed to guarantee a uniform illumination over the sample. The prototype demonstrated a sensitivity of 72.1% and a high specificity of 100%.

Another recent work proposed a smartphone-based microscopic biosensor for label-free CD4 T-cell detection as shown in Fig. 1(e).[47] A special designed microfluidic chip with surface modification captured the CD4 cells and was inserted into the optical attachment above the camera sensor for imaging. A pair of external aspheric lenses harvested from pick-up heads of an external DVD drive was positioned adjacent to each other above the camera. The device was evaluated using whole blood and compare with manual and commercial flow cytometry analysis, which showed a good agreement.

(2) **Lensfree approach.** To make the system more compact, another type of bright-field microscopic imaging biosensor is the lensfree approach.[48] In lensfree imaging, there are also two common types. One is the shadow imaging with incoherent light source. The other is the holographic imaging with partially-coherent light source.

Shadow imaging is probably the simplest form of bright-field microscopy technique using a smartphone. The initial work was the On-chip Microscope developed at Yang's group by attaching one microfluidic channel closely above a CIS.[49,50] The basic imaging principle is as follows. When bio-samples in the microchannel flew across the CIS surface, their diffracted images would be projected to the CIS pixel array underneath with the above white light illumination. Based on such principle, as shown in Fig. 2(a), Lee *et al.* demonstrated a smartphone-based lensless microscopy by adopting the ambient illumination such as sunlight as the light source.[51] Due to the diffraction effect without a lens, the directly captured shadow images have a very low resolution. Therefore, image reconstruction was necessary. In this work, they performed a subpixel resolved super-resolution processing with a custom-designed App, which can reconstruct multiple low-resolution images to a high resolution one.[50] The prototype implemented a spatial resolution of 500 nm for the imaging of microorganisms in water and blood smear. However, the throughput limitation may limit its practical usage.

The incoherent smartphone based microscopic image sensing systems can only capture the intensities of the object images. Differently, holographic imaging can acquire both amplitude and phase information of the objects. Based on holographic imaging principle, Ozcan's group reported a

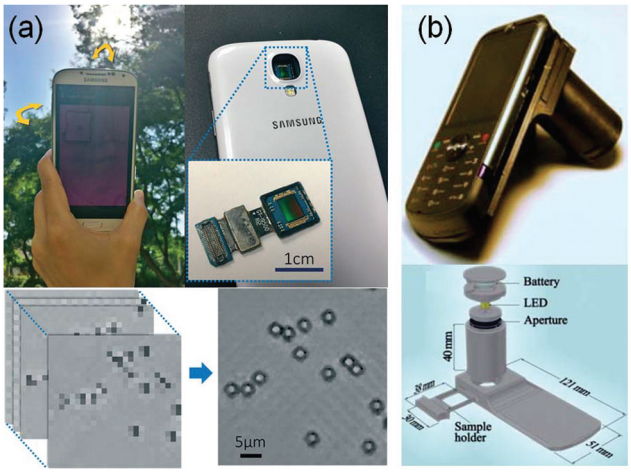

Fig. 2 (a) Smartphone-based lensless microscope utilizing ambient illumination, reproduced from ref. 51 with permission from the Royal Society of Chemistry, copyright 2014; (b) smartphone-based lensless microscope using holographic imaging reproduced from ref. 52 with permission from the Royal Society of Chemistry, copyright 2010.

series of lensless microscopic prototypes.[53–55] One of their initial demonstration on a mobile phone used a light-weight attachment to image blood cells, waterborne parasites, and microparticles as shown in Fig. 2(b).[52] The attachment consisted a 587 nm wavelength LED with a 100 μm pinhole ahead of the source for partially-coherent sample illumination. The scatted light from the sample interferes with the unperturbed background, which created in-line holograms and are then acquired by the mobile phone embedded CIS. Then, iterative holographic reconstruction image processing algorithms were employed to recover the amplitude and phase information. To further improve the spatial resolution, super resolution image processing algorithms were applied with a source array.

B. Fluorescent microscopic imaging biosensor

Compared with bright-field bio-imaging, fluorescence bio-imaging provides enhanced sensitivity and specificity, which is better for detection of biological targets even at nanoscale. In the current fluorescent imaging, LEDs are usually employed to excite the fluorescence and provide enough signal to noise ratio (SNR) level for observation, and/or quantify the fluorescently labeled micro objects, such as blood cells, proteins, bacterium, nucleic acids, and viral antigens.[56,57]

An early fluorescence microscopy demonstration was built on a feature phone by Zhu *et al.*, as shown in Fig. 3(a).[58] Butt-coupled LEDs at the side were used to excite the fluorescent tags, and the emitted fluorescent images of the sample were captured by an external lens attached above the existing lens of the cell-phone camera. The prototype successfully imaged labeled white blood cells and *Giardia Lamblia* cysts in water for quality monitoring. A 10 μm resolution over a FOV of 81 mm^2 was achieved with the help of compressive sampling based digital image processing.

The limit of detection can be further improved to nanoscale for nanoparticle, viruses, or molecular biological samples detection, for example, deoxyribonucleic acid (DNA). As an

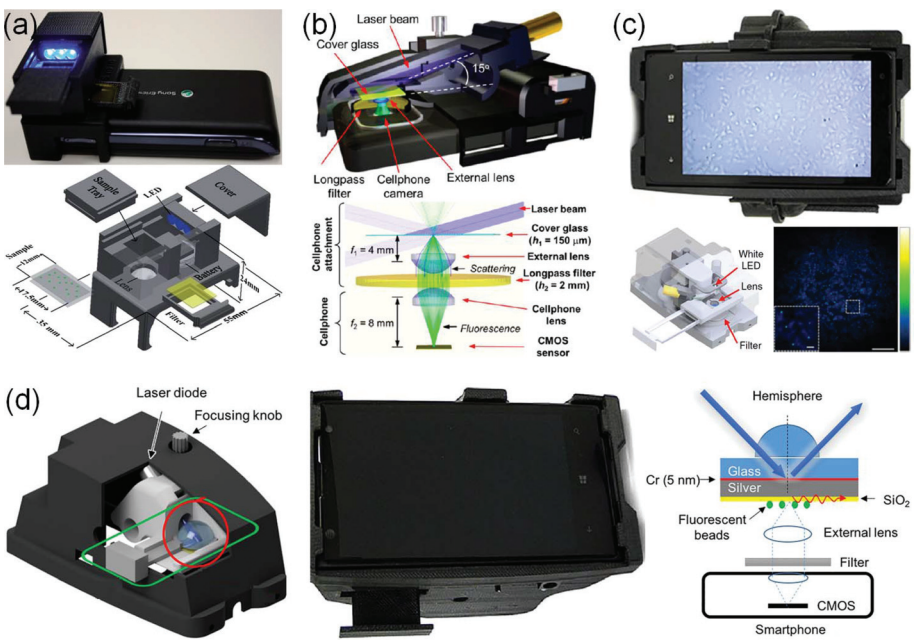

Fig. 3 (a) Smartphone-based fluorescence microscopy with LED butt-coupled from the side, reproduced from ref. 58 with permission from the Royal Society of Chemistry, copyright 2011; (b) smartphone-based fluorescence microscopy for DNA detection, reproduced from ref. 59 with permission from the American Chemical Society, copyright 2013; (c) smartphone-based fluorescence microscopy for DNA sequencing analysis reproduced from ref. 60 with permission from the Nature Publishing Group, copyright 2017; (d) smartphone-based fluorescence microscope with plasmonics enhanced sensitivity reproduced from ref. 61 with permission from the Nature Publishing Group, copyright 2017.

example, Wei *et al.* designed a smartphone-based opto-mechanical attachment which used a 450 nm laser diode to illuminate the sample at an large incidence angle of about 75° and a long-pass interference filter to reject the scattering light as shown in Fig. 3(b).[59] An external low NA lens was utilized to generate a 2× magnification with a translation stage to adjust focus. Such design can efficiently reject the background rejection so that weak fluorescent signal from nanoscale samples can be isolated. By validation with scanning electron microscopy (SEM), the smartphone based microscopic imaging biosensor system demonstrated the capability to image 100 nm fluorescent particles as well as human cytomegalo viruses (HCMV). More recently in 2017, one smartphone-based microscopic biosensor targeting next-generation DNA sequencing and *in situ* point mutation analysis as shown in Fig. 3(c) was also proposed by the same group for cost-effective POC molecular diagnostics.[60] Two laser diodes were used in this prototype with the similar large incidence angle as the previous work.

Moreover, to enhance the fluorescent detection sensitivity, the same group also proposed to use a thin metal-film based surface-enhanced fluorescence (SEF).[61] As shown in Fig. 3(d), the laser diode excited through a glass hemisphere, and the laser beam was filtered with a linear polarizer. After a series of optimization such as film thickness and excitation angle, they achieved ~10-fold improved fluorescence intensity in comparison with a bare glass substrate, which allowed the fluorescent particles with 50 nm diameter as well as single quantum-dots to be imaged.

3. Smartphone-based colorimetric sensor

The previous section reviewed the smartphone-based microscopic imaging biosensor modalities, which offered numerous solutions for high-resolution image based sensing. In this section, another widely applied sensing modality based on color changes and intensity measurement, *i.e.*, smartphone-based colorimetric analytical biosensor, was reviewed.

The reason that CIS of a smartphone can be used for colorimetric sensing is due to the on-chip micro lens and color filter on each CIS pixel.[62,63] The micro lens is formed on color filter to enhance light gathering power to the photodiode and improve sensitivity; the color filter filters the light according to wavelength ranges so that the intensities of filtered light can represent color information of light. The most common pattern for color filter array is the Bayer mosaic, of which each two-by-two Bayer pattern submosaic consists of one red, two green, and one blue filter with each one covering one pixel.[64] Besides this RGB color space, Hue Saturation Value (HSV) and International Commission on Illumination (CIE) are also used sometimes when external parameters such as brightness and color intensity affect the RGB image analysis.[65] Moreover, color correction and white balance are also indispensable to guarantee proper color fidelity of the captured image.[66,67]

When developing smartphone-based colorimetric biosensor, the smartphone camera can be directly used to capture images with colorimetric changes. And sometimes external attachments are required to control the ambient light condition and camera position, and provide a battery, LED illumination or external lens. Yet, it is still not easy to conduct high-accuracy quantitative measurements due to difficulty in small color change detection and different color balancing functions of the smartphone. The color quantification in recent smartphone-based colorimetric biosensor can be performed by smartphone App without extra computer processing.

Numerous biomedical assay applications can cause colorimetric and intensity changes. As for the colorimetric immunoassay, T. S. Park *et al.* proposed one smartphone based *E. coli* detection in field water with a three-channel paper-based microfluidic chip as shown in Fig. 4(a).[68] Anti-*E. coli*-conjugated microparticles were loaded into two *E. coli* detection channels for low and high concentration bacteria detection. And bovine serum albumin (BSA)-conjugated beads were loaded into the control channel for comparison. When field water sample was introduced, the *E. coli* antigens would immunoagglutinate the antibody-conjugated beads. Then the immunoagglutination level could be quantified by examining the intensity of Mie scatter from the images captured by a smartphone installed on an angled holder with an optimized angle and distance. The assay can be finished in 90 s and showed good agreements with the commercial tool.

For chemiluminescence with redox reaction, the smartphone-based biosensor is also used. One most recent work by Shahvar *et al.* as shown in Fig. 4(b) presented a smartphone-based chemiluminescence sensing for thin layer chromatography (TLC) imaging towards the detection of morphine.[69] The potassium permanganate reagent was added to the morphine spot on the TLC plate, and the emitted light image was taken by a smartphone camera attached with a custom-design dark box. With a smartphone App for image processing, the intensity of the red color (R) was used as the analytical signal for morphine concentrations.

One other simple colorimetric sensing technique using a smartphone camera to measure the nitrite concentration and pH based on an inexpensive paper-based microfluidic device was demonstrated by N. Lopez-Ruiz, *et al.*, as illustrated in Fig. 4(c).[70] No external attachment was required except the flash of the smartphone used for light source. The smartphone camera images are processed by a custom-built App to detect the colored sensing areas on the microfluidic paper and extract H (hue) and S (saturation) levels in the HSV space to represent pH and nitrite concentration. The results showed that the pH resolution was 0.04 units with an accuracy of 0.09, while for nitrite, it achieved 0.51% at 4.0 mg L^{-1} of resolution and 0.52 mg L^{-1} limit of detection (LOD).

Similarly, in the forensic application, Shin *et al.* recently proposed a smartphone-based bloodstain age estimation by directly taking its image and perform colorimetric analysis of

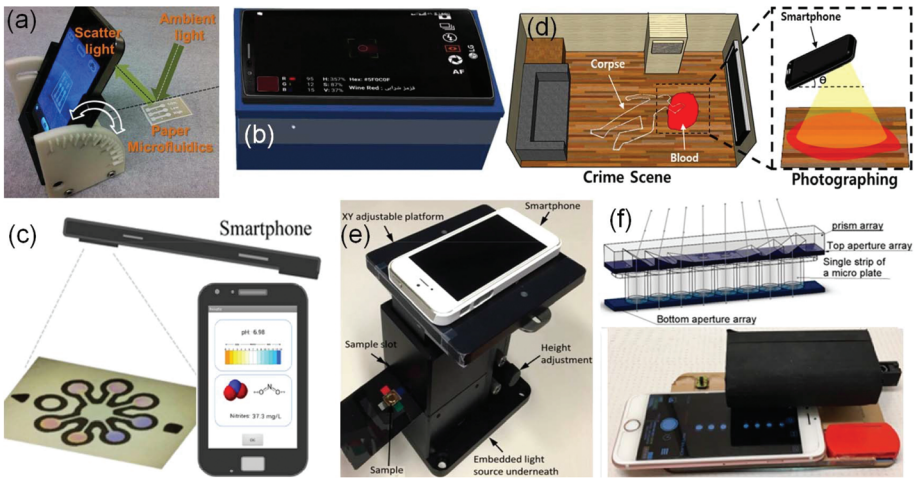

Fig. 4 (a) Smartphone-based colorimetric immunoassay for *E. coli* detection, reproduced from ref. 68 with permission from IEEE, copyright 2015; (b) smartphone-based chemiluminescence sensing for TLC imaging reproduced from ref. 69 with permission from Elsevier, copyright 2018; (c) smartphone-based colorimetric simultaneous detection of pH and nitrite reproduced from ref. 70 with permission from the American Chemical Society, copyright 2014; (d) smartphone-based colorimetric bloodstain age estimation reproduced from ref. 71 with permission from Elsevier, copyright 2017; (e) smartphone-based self-referenced plasmonic sensing reproduced from ref. 72 with permission from the American Chemical Society, copyright 2016; (f) smartphone-based colorimetric biosensor for rapidly herbicides detection reproduced from ref. 73 with permission from the American Chemical Society, copyright 2017.

the RGB intensity without any accessory, as shown in Fig. 4(d).[71] Then the plot of the brightness value of HSV can infer the age of bloodstain. The deficiency is that age of estimation cannot be longer than 42 hours due to no significant color change after that elapsed time.

Another recent inkjet-printed paper-based colorimetric polyion sensor with a smartphone as a detector was proposed by X. Wang, *et al.*[74] The optode paper was affixed at the back of a transparent 96-well microtiter plate, and its picture was directly captured in a custom-designed black box by a smartphone with LED flash as the only light source.

To reduce the error from the light source, X. Wang *et al.* proposed a self referenced smartphone-based plasmonic nanohole sensor for liquid refractive index and optical absorbance enhancement sensor, as shown in Fig. 4(e). The LOD was improved greatly by about one hundred times in a microplate reader format.[72] It was integrated with an internal reference sample and use an App to process images for colorimetric sensing.

One other recent work by Y. Wang *et al.* proposed a smartphone based colorimetric biomedical sensor to rapidly characterize the herbicides in single-stripe microliter plate with a sensitivity in the range of 1 to 80 ppb (part per billion) and good linearity, as shown in Fig. 4(f).[73] The 3D printed attachment includes a prism array and two aperture arrays on the top and at the bottom of the microplate stipe so as to guide light through all wells vertically and block other lights, also allow all the well images to be captured by taking only one picture. No smartphone holder was applied to enable that all smartphone with front cameras could be applied to this device. The device is compact (50 mm × 100 mm × 160 mm), and costs only about $10 for POC.

4. Smartphone-based electrochemical sensor

Electrochemical analysis, with high sensitivity, high specificity and convenience, has been widely applied in quantitative detections of analytes (*e.g.* proteins, nucleic acids and metabolites) for environment monitoring, safety evaluation as well as wellness tracking.[75–79] However, traditional electrochemical biosensors are generally expensive, bulky and limited to the trained personnel. Smartphones therefore offer a promising platform to control, record, and display the real-time and POC detections by electrochemical measurements. Until now, many applications have been exploited that smartphone serves as the data receiver and analyzer of an electrochemical sensor. According to the employed electrochemical apparatus, smartphone-based electrochemical sensors have been classified into three types of amperometric biosensor, potentiometric biosensor, impedimetric biosensor.

A. Smartphone-based amperometric biosensor

Amperometric biosensor is relied on the variation of current resulting from the oxidation or reduction of the electrical mediating species when the potential is constant, which is proportional to the concentration of analytes. Amperometry is the most widely used technique combined with smartphone due to its different methods to detect different analytes, cyclic voltammetry, chronoamperometry, square wave voltammetry and differential pulse voltammetry included.

Wang and Lin *et al.* first demonstrated a paper-based microfluidic smartphone electrochemical biosensor for label-free white blood cell (WBC) counting.[80] As shown in Fig. 5(a)

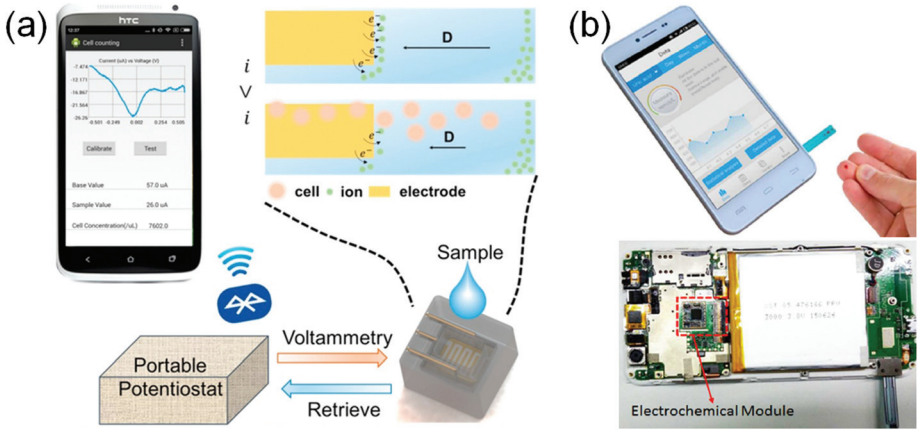

Fig. 5 (a) The system diagram and electrochemical principle for WBC count where *D* is the diffusion coefficient and *i* is the electrochemical current. Reproduced from ref. 80 with permission from Elsevier, copyright 2017; (b) photograph of the proposed medical smartphone with an pre-buried electrometer; side view showed a slot for UA test strip insertion, schematic diagram shows a microcontroller based current measurement circuits; top layout view of the medical-smartphone showed the compact electrochemical module pre-buried within the smartphone, reproduced from ref. 31.

WBCs separated from whole blood was physically captured on the microporous paper with interdigitated gold microelectrodes, which was used to determine the cell concentration *via* the method of differential pulse voltammeter. The quantitative results were collected wirelessly by the smartphone within only one min. This platform could be applied to rapidly quantify the concentration of WBC in its physiological and pathological range with only 10 μL sample and high repeatability. The unique smartphone-based amperometric biosensor provides the opportunity for fast cell quantification to achieve POC diagnosis of WBC for patients with leukemia or other immune diseases.

Another example is that Guo *et al.* developed the world's first medical smartphone, which worked with a disposable test strip for rapid estimation of uric acid (UA) in finger-pricked whole blood.[76] The smartphone was integrated with the electrochemical module connecting to a disposable electrochemical UA test strip as shown in Fig. 5(b). Only 3 μL finger-pricked whole blood was enough to load on the testing strip for UA characterization. A satisfactory agreement was achieved compared to the bulky analyzer in clinical lab. The smartphone was a reader that mapped the current signal to the blood UA level, which could be easily stored and uploaded to a personal-health management center for telemedicine application. This medical smartphone is commercially available by Laya Technology Co., Ltd, Chengdu, China.

B. Smartphone-based potentiometric biosensor

Smartphone-based potentiometric biosensor is also a kind of portable electrochemical biosensor for the POC detections. Potentiometry is suitable for sensing applications in which the cumulative electrical charges result in the differences in electrical potential on top of the dielectric layer. Deng *et al.* developed an on-site gender detection device using smartphone-based electrochemical chip. Based on the difference of creatine kinase (CK) and alanine transaminase (ALT), male and female gender can be differentiated.[81] The smartphone included an embedded electrochemical analysis module and a customized application (APP) for electrochemical biosensing. The enzyme level in biofluids was converted to the consumption of NADH by enzyme cascade reaction, which can be potentiometrically analyzed by the as-designed chip. This device was capable of gender verification within 20 min through on-site detection of serum and serum stains. The device exhibited excellent sensitivity and specificity by gender verification of 39 real samples.

Also, Garcia *et al.* reported a smartphone-based potentiometric biosensor for POC testing of salivary α-amylase (sAA) for personal psychological measurement.[82] The biosensing system was integrated with a smartphone with a sAA-detection application, a sensing chip with preloaded reagents, and a potentiometric reader. The sAA reacts with the preloaded reagents when saliva enters into the reaction zone on the sensing chip, resulting in the conversion of $Fe(CN)_6^{3-}$ to $Fe(CN)_6^{4-}$. Then the sensing chip was pressed to transfer the mixture to the detection zone. The potential read by the smartphone-based potentiometric sensor was sent to the sAA-detection application through the USB port, and converted to the concentration of sAA according to a calibration curve. And the results were in good agreement with that acquired by a reference assay kit. This platform provided an emerging noninvasive biomarker for psychological health for POC testing of sAA.

C. Smartphone-based impedimetric biosensor

The impedance of an electrochemical system is measured by electrochemical impedance spectroscopy (EIS) when the frequency of potential applied is fixed. The smartphone-based impedimetric biosensor was first reported by Broeders *et al.* in 2013.[83] In addition, Zhang *et al.* have demonstrated a portable smartphone-based biosensor for trinitrotoluene (TNT) detec-

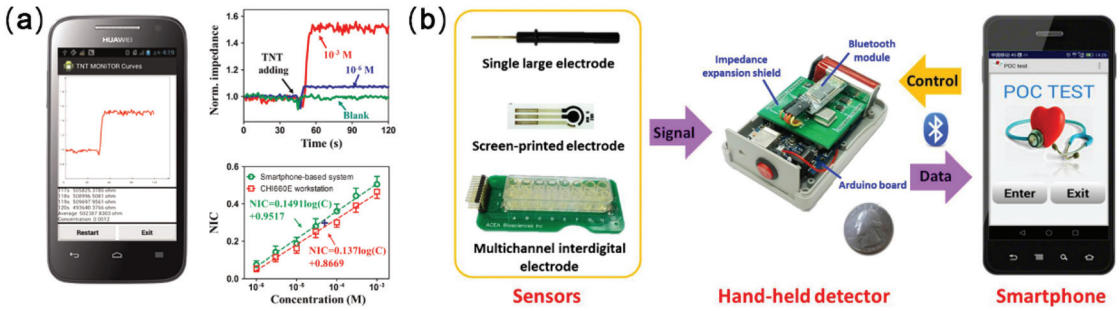

Fig. 6 (a) Picture of smartphone screen showing impedance monitoring curve (the top half) and information about real time impedance values and finally concentrations (the bottom half); real-time monitoring of TNT at different concentrations measured by the system; dose-dependent fitting curves of NIC *versus* concentrations from different devices (the NIC response to as-prepared TNT solution at 5×10^{-5} M was shown as blue cross), reproduced from ref. 84 with permission from Elsevier, copyright 2015. (b) The smartphone-controlled electrochemical biosensor system, including sensors, the hand-held detector and the smartphone, reproduced from ref. 85 with permission from Elsevier, copyright 2016.

tion by impedance monitoring.[84] The disposable biosensor was consisted of the screen-printed electrodes modified with TNT-specific peptides, which produced impedance responses to the biosensor, as shown in Fig. 6a. The impedance responses could be observed and output by the smartphone *via* Bluetooth. Then, the TNT responses and the concentration of TNT were displayed on the smartphone in real time. In this work, the limitation of detection of TNT is as low as 10^{-6} M, providing a portable platform for chemical detections.

Liu's group developed a smartphone-controlled biosensor for on-site detection of proteins by EIS.[85] As is shown in Fig. 6(b), the system was integrated with a miniaturized biosensor, a hand-held EIS detector and a smartphone. The miniaturized biosensor was integrated with printed carbon electrodes and interdigital gold electrodes for protein detection, which were modified with bio-components. Then, special antibodies and peptides were immobilized on the electrodes to quantify the protein binding and enzyme activities of bull serum albumin (BSA) and thrombin. In their study, the limitation of detection of BSA and thrombin the system could approach to 1.78 g mL^{-1} and 2.97 ng mL^{-1}, respectively. The analytical performances of this system indicate that diverse smartphone-controlled biosensor system could be developed for POC quantifications of various proteins.

5. Smartphone-based electrochemiluminescence sensor

Electrochemiluminescence (ECL) is highly sensitive detection technique by combining the electrochemistry and chemiluminescence.[86] Electrochemistry reaction by applying a specific voltage produce the electro-generated substances on the surface of the electrode. Then these substances reacting with some components pre-buried above the electrodes formed an excited state through electronic delivery and returned from the excited stage to the basic stage in which accompanied the luminescence happened. Excellent characteristics, for example, lower-background signal, easy to manipulate with the position, light emission, highly sensitivity makes the ECL widely available in the current clinical market.

Doeven *et al.* demonstrated a low cost platform to perform the electrochemiluminescence analytical platform by utilizing a smartphone. The miniaturized system can obtain power source and interchange the biosensor information from the smartphone's USB 'On-The-Go' (OTG) port which utilized audible tone pulses acted over the audio jack of the smartphone. As Fig. 7 indicated, the smartphone can provide a stable platform for the electrochemiluminescence analytical platform, which paved the way for more immune biosensor with the smartphone as signal readout.[87]

Zhang's group reported a palm-sized paper-based bipolar electrode electrochemiluminescence (P-BPE-ECL) system with a smartphone as the signal reader as shown in Fig. 8.[88] In the

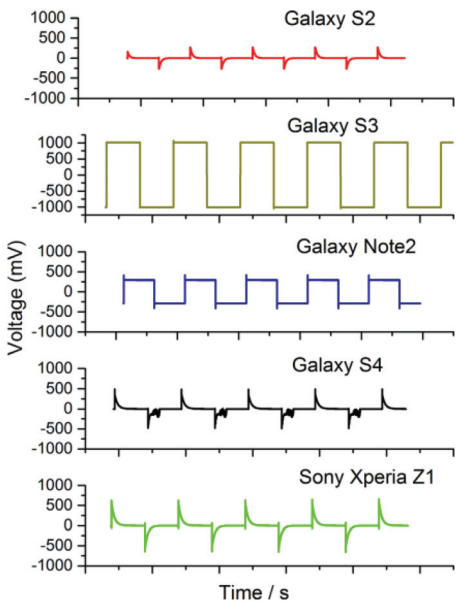

Fig. 7 Audio outputting signal from prevailing smartphones in the markets when 1 Hz square wave was generated at maximum volume, reproduced from ref. 87 with permission from Elsevier, copyright 2015.

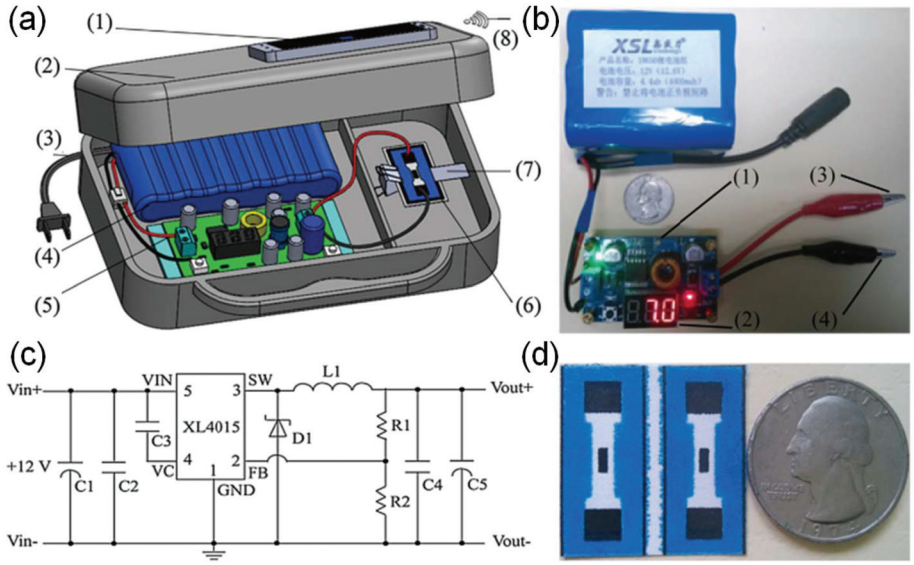

Fig. 8 Scheme of the proposed P-BPE-ECL system reproduced from ref. 88 with permission from Elsevier, copyright 2016. (a) schematic view of the palm-sized system: (1) smartphone; (2) box to load the system; (3) power cable; (4) reusable Li battery; (5) control and display driving part; (6) the proposed BPE-ECL paper device; (7) device holder; and (8) wireless communication of measured result to PC; (b) practical photo of the Li battery and functional circuit board: (1) potentiometer for power management; (2) digital tube for display; (3) positive output-voltage; and (4) negative output-voltage. (c) Design of the circuit function. (d) Photo of the paper-based sensor consisting of two sets of P-BPE-ECL units.

proposed miniaturized system, a lithium battery is utilized for power supply and the smartphone performed as ECL signal reader. For the disposable strip, the screen printed BPE was fabricated as the driving electrodes for ECL biosensor, and the wax screen printing is applied for manufacturing microfluidic channels on electrode patterned paper substrate. The proposed miniaturized system was demonstrated to have the good sensitivity, wide detection range, reliability and reproducibility. Glucose in PBS and artificial urine (AU) solutions were tested to reveal the limits of detection (LOD) of 0.017 mM and 0.030 mM for the proposed system, respectively. In summary, the reported strips with smartphone can provide a fast and reliable platform for mobile health.

6. Conclusions and future perspectives

The recent advancement of smartphone-based analytical biosensor has provided people with numerous portable, cost-effective, and easy-to-operate platforms for POC applications and mobile health. Towards this, this article reviews the most recent development of four types of smartphone based analytical biosensor based on the common analytical methods, *i.e.*, smartphone-based microscopic imaging, colorimetric, electrochemical, and electrochemiluminescence biosensor, with their biosensing strategies and analytical performances discussed. Although not all aspects of the smartphone based analytical biosensor system are covered, the special characteristics and advantages of smartphone based analytical biosensors are of great potentials and are very promising in the future point of care clinical test and personalized healthcare.

Despite the great progress made, challenges still exist for the design and application of smartphone-based analytical biosensors. Existing smartphone-based analytical biosensors are mostly equipped with in-house designed accessories, which are hand-made and have no strict or unified standards. The target is to be compact with not too complicated setup. Thus, the analytical sensitivity, precision and reproducibility should be strictly guaranteed and validated by well-accredited technologies such as the gold standard benchtop instruments so that they can be translated to practical or clinical applications. The performance improvement can be from software perspective. For example, more and more smartphone-based biosensors have adopted machine learning techniques to improve the resolution of image sensing and accuracy of sample classification.[4,8,38,89,90] From hardware, the novel technologies should be utilized to simplify accessories for smartphone by optimizing their structures and coupling. Multimodal sensor can be designed to add more analytical capabilities to the existing sensor. The development of smartphone technology itself is critical fundamentally such as further enlarged pixel array, reduced pixel size, and improved pixel intensity for the smartphone camera, more powerful processor, higher-speed wireless connection such as 5G. The future smartphone may adopt modular design, which would revolutionize the smartphone-based analytical sensor design again. Since a good smartphone-based biosensor is based on the research and technology development of multiple disciplines, they would enlighten the designers of biomedical circuits, MEMS device, analytical chemistry, microsystem, com-

munication, material, optical, and mechanical area to further develop the promising potentials of smartphone-based analytical biosensor in future POC applications, so that they can make positive impact to the future healthcare continuously.

All in all, due to the pervasive penetration rate of smartphone in people's lives, after continuous advance of smartphone-based biosensing technology and fully exploiting the potentials of smartphone-based biosensors, we strongly believe that the existing centralized diagnostic and healthcare systems, environmental monitoring, food safety screening, *etc.*, could be revolutionized.

Conflicts of interest

There are no conflicts to declare.

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References

1 M. Brown and C. Wittwer, Flow cytometry: principles and clinical applications in hematology, *Clin. Chem.*, 2000, **46**(8), 1221–1229.
2 M. E. Piyasena and S. W. Graves, The intersection of flow cytometry with microfluidics and microfabrication, *Lab Chip*, 2014, **14**(6), 1044–1059.
3 X. Huang, X. Wang, M. Yan and H. Yu, A robust recognition error recovery for micro-flow cytometer by machine-learning enhanced single-frame super-resolution processing, *Integration*, 2015, **51**, 208–218.
4 X. Huang, J. Guo, X. Wang, M. Yan, Y. Kang and H. Yu, A contact-imaging based microfluidic cytometer with machine-learning for single-frame super-resolution processing, *PLoS One*, 2014, **9**(8), e104539.
5 J. Guo, X. Huang and Y. Ai, On-demand lensless single cell imaging activated by differential resistive pulse sensing, *Anal. Chem.*, 2015, **87**(13), 6516–6519.
6 E. Betzig, J. Trautman, T. Harris, J. Weiner and R. Kostelak, Breaking the diffraction barrier: optical microscopy on a nanometric scale, *Science*, 1991, **251**(5000), 1468–1470.
7 E. Betzig and R. J. Chichester, Single molecules observed by near-field scanning optical microscopy, *Science*, 1993, **262**(5138), 1422–1425.
8 X. Huang, Y. Jiang, X. Liu, H. Xu, Z. Han, H. Rong, H. Yang, M. Yan and H. Yu, Machine learning based single-frame super-resolution processing for lensless blood cell counting, *Sensors*, 2016, **16**(11), 1836.
9 G. J. Kost, Guidelines for point-of-care testing. Improving patient outcomes, *Am. J. Clin. Pathol.*, 1995, **104**(4), S111–S127.
10 P. B. Luppa, C. Müller, A. Schlichtiger and H. Schlebusch, Point-of-care testing (POCT): Current techniques and future perspectives, *TrAC, Trends Anal. Chem.*, 2011, **30**(6), 887–898.
11 S. K. Vashist, P. B. Luppa, L. Y. Yeo, A. Ozcan and J. H. Luong, Emerging technologies for next-generation point-of-care testing, *Trends Biotechnol.*, 2015, **33**(11), 692–705.
12 X. Huang, H. Yu, X. Liu, Y. Jiang and M. Yan, A Single-Frame Superresolution Algorithm for Lab-on-a-Chip Lensless Microfluidic Imaging, *IEEE Des. Test*, 2015, **32**(6), 32–40.
13 S. K. Sia and L. J. Kricka, Microfluidics and point-of-care testing, *Lab Chip*, 2008, **8**(12), 1982–1983.
14 P. Yager, G. J. Domingo and J. Gerdes, Point-of-care diagnostics for global health, *Annu. Rev. Biomed. Eng.*, 2008, **10**, 107–144.
15 P. Yager, T. Edwards, E. Fu, K. Helton, K. Nelson, M. R. Tam and B. H. Weigl, Microfluidic diagnostic technologies for global public health, *Nature*, 2006, **442**(7101), 412–418.
16 C. D. Chin, V. Linder and S. K. Sia, Lab-on-a-chip devices for global health: past studies and future opportunities, *Lab Chip*, 2006, **7**(1), 41–57.
17 R. Daw and J. Finkelstein, Lab on a chip, *Nature*, 2006, **442**(7101), 367–367.
18 D. Figeys and D. Pinto, Lab-on-a-Chip: A Revolution in Biological and Medical Sciences, *Anal. Chem.*, 2000, **72**(9), 330–335.
19 D. Mark, S. Haeberle, G. Roth, F. von Stetten and R. Zengerle, Microfluidic lab-on-a-chip platforms: requirements, characteristics and applications, *Chem. Soc. Rev.*, 2010, **39**(3), 1153–1182.
20 X. Huang, U. Farooq, J. Chen, Y. Ge, H. Gao, J. Su, X. Wang, S. Dong and J. K. Luo, A surface acoustic wave pumped lensless microfluidic imaging system for flowing cell detection and counting, *IEEE Trans. Biomed. Circuits Syst.*, 2017, **11**(6), 1478–1487.
21 X. Huang, H. Yu, X. Liu, Y. Jiang, M. Yan and D. Wu, A dual-mode large-arrayed CMOS ISFET sensor for accurate and high-throughput pH sensing in biomedical diagnosis, *IEEE Trans. Biomed. Eng.*, 2015, **62**(9), 2224–2233.
22 H. Yu, M. Yan and X. Huang, *CMOS integrated lab-on-a-chip system for personalized biomedical diagnosis*, John Wiley & Sons, Singapore, 2018.
23 S. Kanchi, M. I. Sabela, P. S. Mdluli and K. Bisetty, Smartphone based bioanalytical and diagnosis applications: A review, *Biosens. Bioelectron.*, 2017, **102**, 136–149.
24 K. E. McCracken and J.-Y. Yoon, Recent approaches for optical smartphone sensing in resource-limited settings: a brief review, *Anal. Methods*, 2016, **8**(36), 6591–6601.

25 G. Rateni, P. Dario and F. Cavallo, Smartphone-based food diagnostic technologies: a review, *Sensors*, 2017, **17**(6), 1453.
26 A. Roda, E. Michelini, M. Zangheri, M. Di Fusco, D. Calabria and P. Simoni, Smartphone-based biosensors: A critical review and perspectives, *TrAC, Trends Anal. Chem.*, 2016, **79**, 317–325.
27 X. Xu, A. Akay, H. Wei, S. Wang, B. Pingguan-Murphy, B.-E. Erlandsson, X. Li, W. Lee, J. Hu and L. Wang, Advances in smartphone-based point-of-care diagnostics, *Proc. IEEE*, 2015, **103**(2), 236–247.
28 D. Zhang and Q. Liu, Biosensors and bioelectronics on smartphone for portable biochemical detection, *Biosens. Bioelectron.*, 2016, **75**, 273–284.
29 Smartphones – Statistics & Facts. https://www.statista.com/statistics/330695/number-of-smartphone-users-worldwide/.
30 Z. S. Ballard, C. Brown and A. Ozcan, Mobile Technologies for the Discovery, Analysis, and Engineering of the Global Microbiome, *ACS Nano*, 2018, **12**(4), 3065–3082.
31 J. Guo, Uric Acid Monitoring with a Smartphone as the Electrochemical Analyzer, *Anal. Chem.*, 2016, **88**(24), 11986–11989.
32 S. K. Jain and B. Bhaumik, An energy efficient ECG signal processor detecting cardiovascular diseases on smartphone, *IEEE Trans. Biomed. Circuits Syst.*, 2017, **11**(2), 314–323.
33 H. Karlsen and T. Dong, Smartphone-Based Rapid Screening of Urinary Biomarkers, *IEEE Trans. Biomed. Circuits Syst.*, 2017, **11**(2), 455–463.
34 P.-S. Liang, T. S. Park and J.-Y. Yoon, Rapid and reagentless detection of microbial contamination within meat utilizing a smartphone-based biosensor, *Sci. Rep.*, 2014, **4**, 5953.
35 A. Sun, A. Venkatesh and D. A. Hall, A multi-technique reconfigurable electrochemical biosensor: enabling personal health monitoring in mobile devices, *IEEE Trans. Biomed. Circuits Syst.*, 2016, **10**(5), 945–954.
36 Q. Wei, R. Nagi, K. Sadeghi, S. Feng, E. Yan, S. J. Ki, R. Caire, D. Tseng and A. Ozcan, Detection and spatial mapping of mercury contamination in water samples using a smart-phone, *ACS Nano*, 2014, **8**(2), 1121–1129.
37 D. Xu, X. Huang, J. Guo and X. Ma, Automatic smartphone-based microfluidic biosensor system at the point of care, *Biosens. Bioelectron.*, 2018, **110**, 78–88.
38 X. Liu, X. Huang, Y. Jiang, H. Xu, J. Guo, H. W. Hou, M. Yan and H. Yu, A Microfluidic Cytometer for Complete Blood Count With a 3.2-Megapixel, 1.1-um-Pitch Super-Resolution Image Sensor in 65 nm BSI CMOS, *IEEE Trans. Biomed. Circuits Syst.*, 2017, **11**(4), 794–803.
39 E. R. Fossum, CMOS image sensors: electronic camera-on-a-chip, *IEEE Trans. Electron Devices*, 1997, **44**(10), 1689–1698.
40 Y. Kim, W. Choi, D. Park, H. Jeoung, B. Kim, Y. Oh, S. Oh, B. Park, E. Kim, Y. Lee, T. Jung, Y. Kim, S. Yoon, S. Hong, J. Lee, S. Jung, C. R. Moon, Y. Park, D. Lee and D. Chang, In A 1/2.8-inch 24Mpixel CMOS image sensor with 0.9um unit pixels separated by full-depth deep-trench isolation2018 IEEE International Solid-State Circuits Conference (ISSCC), 11-15 Feb. 2018, 2018, pp. 84–86.
41 G. Taverni, D. P. Moeys, C. Li, C. Cavaco, V. Motsnyi, D. S. S. Bello and T. Delbruck, Front and Back Illuminated Dynamic and Active Pixel Vision Sensors Comparison, *IEEE Trans. Circuits Syst. II, Exp. Briefs*, 2018, 1–1.
42 J. C. Contreras-Naranjo, Q. Wei and A. Ozcan, Mobile phone-based microscopy, sensing, and diagnostics, *IEEE J. Sel. Top. Quantum Electron.*, 2016, **22**(3), 1–14.
43 D. N. Breslauer, R. N. Maamari, N. A. Switz, W. A. Lam and D. A. Fletcher, Mobile phone based clinical microscopy for global health applications, *PLoS One*, 2009, **4**(7), e6320.
44 Z. J. Smith, K. Chu, A. R. Espenson, M. Rahimzadeh, A. Gryshuk, M. Molinaro, D. M. Dwyre, S. Lane, D. Matthews and S. Wachsmann-Hogiu, Cell-phone-based platform for biomedical device development and education applications, *PLoS One*, 2011, **6**(3), e17150.
45 N. A. Switz, M. V. D'Ambrosio and D. A. Fletcher, Low-Cost Mobile Phone Microscopy with a Reversed Mobile Phone Camera Lens, *PLoS One*, 2014, **9**(5), e95330.
46 I. I. Bogoch, H. C. Koydemir, D. Tseng, R. K. Ephraim, E. Duah, J. Tee, J. R. Andrews and A. Ozcan, Evaluation of a mobile phone-based microscope for screening of Schistosoma haematobium infection in rural Ghana, *Am. J. Trop. Med. Hyg.*, 2017, **96**(6), 1468–1471.
47 M. K. Kanakasabapathy, H. J. Pandya, M. S. Draz, M. K. Chug, M. Sadasivam, S. Kumar, B. Etemad, V. Yogesh, M. Safavieh and W. Asghar, Rapid, label-free CD4 testing using a smartphone compatible device, *Lab Chip*, 2017, **17**(17), 2910–2919.
48 A. Ozcan and E. McLeod, Lensless imaging and sensing, *Annu. Rev. Biomed. Eng.*, 2016, **18**, 77–102.
49 X. Heng, L. R. Baugh, Z. Yaqoob, P. W. Sternberg, D. Psaltis and C. Yang, Optofluidic microscopy—a method for implementing a high resolution optical microscope on a chip, *Lab Chip*, 2006, **6**(10), 1274–1276.
50 G. Zheng, S. A. Lee, S. Yang and C. Yang, Sub-pixel resolving optofluidic microscope for on-chip cell imaging, *Lab Chip*, 2010, (22), 3125–3129.
51 S. A. Lee and C. Yang, A smartphone-based chip-scale microscope using ambient illumination, *Lab Chip*, 2014, **14**(16), 3056–3063.
52 D. Tseng, O. Mudanyali, C. Oztoprak, S. O. Isikman, I. Sencan, O. Yaglidere and A. Ozcan, Lensfree microscopy on a cellphone, *Lab Chip*, 2010, **10**(14), 1787–1792.
53 O. Mudanyali, D. Tseng, C. Oh, S. O. Isikman, I. Sencan, W. Bishara, C. Oztoprak, S. Seo, B. Khademhosseini and A. Ozcan, Compact, light-weight and cost-effective microscope based on lensless incoherent holography for telemedicine applications, *Lab Chip*, 2010, **10**(11), 1417–1428.
54 W. Bishara, U. Sikora, O. Mudanyali, T.-W. Su, O. Yaglidere, S. Luckhart and A. Ozcan, Holographic pixel super-resolution in portable lensless on-chip microscopy using a fiber-optic array, *Lab Chip*, 2011, **11**(7), 1276–1279.

55 Y. Wu and A. Ozcan, Lensless digital holographic microscopy and its applications in biomedicine and environmental monitoring, *Methods*, 2017, **136**, 4–16.
56 H. Yu, Y. Tan and B. T. Cunningham, Smartphone Fluorescence Spectroscopy, *Anal. Chem.*, 2014, **86**(17), 8805–8813.
57 W.-I. Lee, S. Shrivastava, L.-T. Duy, B. Y. Kim, Y.-M. Son and N.-E. Lee, A smartphone imaging-based label-free and dual-wavelength fluorescent biosensor with high sensitivity and accuracy, *Biosens. Bioelectron.*, 2017, **94**, 643–650.
58 H. Zhu, O. Yaglidere, T.-W. Su, D. Tseng and A. Ozcan, Cost-effective and compact wide-field fluorescent imaging on a cell-phone, *Lab Chip*, 2011, **11**(2), 315–322.
59 Q. Wei, H. Qi, W. Luo, D. Tseng, S. J. Ki, Z. Wan, Z. n. Göröcs, L. A. Bentolila, T.-T. Wu and R. Sun, Fluorescent imaging of single nanoparticles and viruses on a smart phone, *ACS Nano*, 2013, **7**(10), 9147–9155.
60 M. Kühnemund, Q. Wei, E. Darai, Y. Wang, I. Hernández-Neuta, Z. Yang, D. Tseng, A. Ahlford, L. Mathot and T. Sjöblom, Targeted DNA sequencing and in situ mutation analysis using mobile phone microscopy, *Nat. Commun.*, 2017, **8**, 13913.
61 Q. Wei, G. Acuna, S. Kim, C. Vietz, D. Tseng, J. Chae, D. Shir, W. Luo, P. Tinnefeld and A. Ozcan, Plasmonics enhanced smartphone fluorescence microscopy, *Sci. Rep.*, 2017, **7**(1), 2124.
62 J. Nakamura, *Image sensors and signal processing for digital still cameras*, CRC press, 2017.
63 G. Agranov, V. Berezin and R. H. Tsai, Crosstalk and microlens study in a color CMOS image sensor, *IEEE Trans. Electron Devices*, 2003, **50**(1), 4–11.
64 R. Lukac, K. N. Plataniotis and D. Hatzinakos, Color image zooming on the Bayer pattern, *IEEE Trans. Circ. Syst. Video Technol.*, 2005, **15**(11), 1475–1492.
65 N. A. Ibraheem, M. M. Hasan, R. Z. Khan and P. K. Mishra, Understanding color models: a review, *ARPN J. Sci. Technol.*, 2012, **2**(3), 265–275.
66 Y.-C. Liu, W.-H. Chan and Y.-Q. Chen, Automatic white balance for digital still camera, *IEEE Trans. Consum. Electron.*, 1995, **41**(3), 460–466.
67 A. Rizzi, C. Gatta and D. Marini, A new algorithm for unsupervised global and local color correction, *Pattern Recognit. Lett*, 2003, **24**(11), 1663–1677.
68 T. San Park and J.-Y. Yoon, Smartphone detection of Escherichia coli from field water samples on paper microfluidics, *IEEE Sens. J.*, 2015, **15**(3), 1902–1907.
69 A. Shahvar, M. Saraji and D. Shamsaei, Smartphone-based chemiluminescence sensing for TLC imaging, *Sens. Actuators, B*, 2018, **255**, 891–894.
70 N. Lopez-Ruiz, V. F. Curto, M. M. Erenas, F. Benito-Lopez, D. Diamond, A. J. Palma and L. F. Capitan-Vallvey, Smartphone-based simultaneous pH and nitrite colorimetric determination for paper microfluidic devices, *Anal. Chem.*, 2014, **86**(19), 9554–9562.
71 J. Shin, S. Choi, J.-S. Yang, J. Song, J.-S. Choi and H.-I. Jung, Smart Forensic Phone: Colorimetric analysis of a bloodstain for age estimation using a smartphone, *Sens. Actuators, B*, 2017, **243**, 221–225.
72 X. Wang, T.-W. Chang, G. Lin, M. R. Gartia and G. L. Liu, Self-referenced smartphone-based nanoplasmonic imaging platform for colorimetric biochemical sensing, *Anal. Chem.*, 2016, **89**(1), 611–615.
73 Y. Wang, M. M. Zeinhom, M. Yang, R. Sun, S. Wang, J. N. Smith, C. Timchalk, L. Li, Y. Lin and D. Du, A 3D-printed, portable, optical-sensing platform for smartphones capable of detecting the herbicide 2, 4-dichlorophenoxyacetic acid, *Anal. Chem.*, 2017, **89**(17), 9339–9346.
74 X. Wang, M. Mahoney and M. E. Meyerhoff, Inkjet-printed paper-based colorimetric polyion sensor using a smartphone as a detector, *Anal. Chem.*, 2017, **89**(22), 12334–12341.
75 L. Yi, J. Li, C. Guo, L. Li and J. Liu, Liquid metal ink enabled rapid prototyping of electrochemical sensor for wireless glucose detection on the platform of mobile phone, *J. Med. Devices*, 2015, **9**(4), 044507.
76 J. Guo, Smartphone-powered electrochemical dongle for point-of-care monitoring of blood β-ketone, *Anal. Chem.*, 2017, **89**(17), 8609–8613.
77 A. Sun, T. Phelps, C. Yao, A. Venkatesh, D. Conrad and D. A. Hall, Smartphone-based pH sensor for home monitoring of pulmonary exacerbations in cystic fibrosis, *Sensors*, 2017, **17**(6), 1245.
78 A. J. Bandodkar, S. Imani, R. Nuñez-Flores, R. Kumar, C. Wang, A. V. Mohan, J. Wang and P. P. Mercier, Re-usable electrochemical glucose sensors integrated into a smartphone platform, *Biosens. Bioelectron.*, 2018, **101**, 181–187.
79 J. Guo, Smartphone-powered electrochemical biosensing dongle for emerging medical IoTs application, *IEEE Trans. Ind. Informat.*, 2018, 1–1.
80 X. Wang, G. Lin, G. Cui, X. Zhou and G. L. Liu, White blood cell counting on smartphone paper electrochemical sensor, *Biosens. Bioelectron.*, 2017, **90**, 549–557.
81 W. Deng, Y. Dou, P. Song, H. Xu, A. Aldalbahi, N. Chen, N. N. El-Sayed, J. Gao, J. Lu and S. Song, Lab on smartphone with interfaced electrochemical chips for on-site gender verification, *J. Electroanal. Chem*, 2016, **777**, 117–122.
82 P. T. Garcia, L. N. Guimarães, A. A. Dias, C. J. Ulhoa and W. K. Coltro, Amperometric detection of salivary α-amylase on screen-printed carbon electrodes as a simple and inexpensive alternative for point-of-care testing, *Sens. Actuators, B*, 2018, **258**, 342–348.
83 J. Broeders, D. Croux, M. Peeters, T. Beyens, S. Duchateau, T. J. Cleij, P. Wagner, R. Thoelen and W. De Ceuninck, Mobile application for impedance-based biomimetic sensor readout, *IEEE Sens. J.*, 2013, **13**(7), 2659–2665.
84 D. Zhang, J. Jiang, J. Chen, Q. Zhang, Y. Lu, Y. Yao, S. Li, G. L. Liu and Q. Liu, Smartphone-based portable biosensing system using impedance measurement with printed electrodes for 2, 4, 6-trinitrotoluene (TNT) detection, *Biosens. Bioelectron.*, 2015, **70**, 81–88.
85 D. Zhang, Y. Lu, Q. Zhang, L. Liu, S. Li, Y. Yao, J. Jiang, G. L. Liu and Q. Liu, Protein detecting with smartphone-

controlled electrochemical impedance spectroscopy for point-of-care applications, *Sens. Actuators, B*, 2016, **222**, 994–1002.

86 L. Hu and G. Xu, Applications and trends in electrochemiluminescence, *Chem. Soc. Rev.*, 2010, **39**(8), 3275–3304.

87 E. H. Doeven, G. J. Barbante, A. J. Harsant, P. S. Donnelly, T. U. Connell, C. F. Hogan and P. S. Francis, Mobile phone-based electrochemiluminescence sensing exploiting the 'USB On-The-Go'protocol, *Sens. Actuators, B*, 2015, **216**, 608–613.

88 L. Chen, C. Zhang and D. Xing, Paper-based bipolar electrode-electrochemiluminescence (BPE-ECL) device with battery energy supply and smartphone read-out: a handheld ECL system for biochemical analysis at the point-of-care level, *Sens. Actuators, B*, 2016, **237**, 308–317.

89 H. Ceylan Koydemir, S. Feng, K. Liang, R. Nadkarni, P. Benien and A. Ozcan, Comparison of supervised machine learning algorithms for waterborne pathogen detection using mobile phone fluorescence microscopy, in *Nanophotonics*, 2017, vol. 6, pp. 731.

90 Y. Rivenson, H. Ceylan Koydemir, H. Wang, Z. Wei, Z. Ren, H. Günaydın, Y. Zhang, Z. Göröcs, K. Liang, D. Tseng and A. Ozcan, Deep learning enhanced mobile-phone microscopy, *ACS Photonics*, 2018, **5**(6), 2354–2364.