Blood glucose monitoring techniques: recent advances, challenges and future perspectives

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Abstract

Blood glucose monitor is a fundamental tool to maintain and control blood glucose levels for routine management of diabetes and complications associated with it. There have been continuous scientific advances and achievements in biosensors for the development of comprehensive glucose monitoring systems since last few decades. Portable, accurate, and reliable glucose monitoring devices with advanced features have been developed in recent years. However, invasive glucose monitors remained the most dominant devices on the market. Commercialization of clinically accurate and dependable non-invasive blood glucose monitoring devices still require rigorous works and further studies in sensor technologies. The aim of this article is to bring current updates and recent progress in electrochemical glucose sensing and non-invasive glucose monitoring techniques along with their merits and limitations. We present their key challenges and address future prospects of non-invasive glucose monitoring.

Keywords

Glucose, Electrochemical, Glucose monitoring, Non-invasive, Glucose monitors, Non-enzymatic.

1. Introduction

The recent scientific advancements in glucose sensing technologies have revolutionized the development and commercialization of glucose monitoring devices. With current technological improvements in glucose biosensors, accurate, comprehensive and integrated glucose monitors have been developed. However, the current available glucose monitoring devices rely on invasive monitoring techniques that require finger prick to withdraw blood samples which cause pain, inconvenience and prone to infections. Hence, there is a need to develop accurate and reliable non-invasive blood glucose monitor that improves convenience and comfort for people with diabetes.

Blood glucose monitoring techniques are broadly classified into invasive, minimally invasive and non-invasive based on their detection modality [1, 2] (as shown in Figure 1).

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Enzymatic electrochemical glucose sensors are the most utilized sensing techniques in the blood glucose monitoring [3, 4]. Through advances in nanotechnology and bio-sensing, the study of non-enzymatic electrochemical glucose sensors became of great interest [3]. Optical glucose sensing methods are among the widely studied non-invasive monitoring techniques to develop glucose monitors. The aim of this paper is to present and discuss the latest advances and developments in the blood glucose monitoring techniques. First, we briefly discuss the current advances in electrochemical blood glucose sensing in two categories: enzymatic and non-enzymatic electrochemical sensing techniques along with their advantages and limitations. Then, we present the recent progress in non-invasive blood glucose monitoring techniques with their merits and drawbacks. Finally, we will explain the key challenges of current blood glucose monitoring techniques and highlight their future perspectives focusing on non-invasive blood glucose monitoring.
Figure 1 An overview of blood glucose monitoring techniques

2. Invasive blood glucose monitoring techniques
2.1 Enzymatic electrochemical sensing

Electrochemical biosensors have revolutionized and dominated the markets of blood glucose monitoring devices since their evolution by Clark and Lyons in 1962. They were initially based on the glucose oxidase enzyme (GOx) for catalytic oxidation of glucose in the presence of oxygen that monitors consumption of oxygen or production of hydrogen peroxide [5, 6]. Electrochemical glucose sensors are the most utilized commercially available devices due to their higher sensitivity, simplicity, robustness, good reproducibility, low cost, accurate and faster time responses [7, 8]. Electrochemical biosensors utilize amperometric, potentiometric, impedance and conductometric approaches to detect electrochemical changes during biorecognition event in glucose sensing. Enzymatic amperometric glucose biosensors are the most widely used sensors whose principle is based on monitoring current generated by electrons exchange between biological systems and electrodes [8].

Extensive scientific contributions have been made in the development of enzymatic electrochemical glucose sensors, through three generations. In the first generation, enzymatic glucose biosensor used oxygen (O2) as a mediator. The glucose level is estimated from the amperometric signal generated via electrochemical oxidation of hydrogen peroxide (H2O2) or electrochemical reduction of O2 [5]. An amperometric measurement of H2O2 requires high potential for high selectivity and oxygen deficit
occurs due to variations in oxygen tension and lower concentration which forced an evolution of second generation enzyme based biosensors [9, 10]. Here, oxygen was replaced by non-physiological electron-redox mediator where the amperometric signal generated via oxidation of the mediator is used to estimate glucose levels [5, 9]. Redox-mediated glucose biosensor faced problems in maintaining mediator and enzyme near the electrode due to small and diffusive molecules [11]. The third generation enzymatic electrochemical biosensors do not require mediator where electrons are directly transferred between enzymes and electrodes [11]. Recent advances in nanostructures and nanotubes such as graphene and carbon nanotubes are promising for the development of nano electrodes and enzymatic electrochemical nano sensors used to develop convenient blood glucose monitors [12–14]. Enzymatic electrochemical biosensors are characterized by shorter stability; relatively higher fabrication costs, complicated modification and limitations associated with the nature of enzymes such as irreversibility, and signal drift [6, 15, 16].

Table 1 provides a summary of few commercially available enzyme based electrochemical glucose monitors.

<table>
<thead>
<tr>
<th>Brand name</th>
<th>Enzyme</th>
<th>Mediator</th>
<th>Test time (S)</th>
<th>Sample volume (µL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abbott Freestyle® Lite</td>
<td>GDH-FAD(2)</td>
<td>Os complex</td>
<td>5</td>
<td>0.3</td>
</tr>
<tr>
<td>Accu-Chek®Aviva Plus</td>
<td>GDH</td>
<td>Ferricyanide</td>
<td>5</td>
<td>0.6</td>
</tr>
<tr>
<td>LifeScan OneTouch® Ultra 2</td>
<td>GOx</td>
<td>Ferricyanide</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>Bayer Contour™ Next USB</td>
<td>GDH</td>
<td>Ferricyanide</td>
<td>5</td>
<td>0.6</td>
</tr>
<tr>
<td>Precision Xtra™</td>
<td>GDH</td>
<td>Ferricyanide</td>
<td>5</td>
<td>0.6</td>
</tr>
</tbody>
</table>

2.2 Non-enzymatic electrochemical sensing

Non-enzymatic electrochemical sensors bring the beginning of the fourth generation of glucose biosensors [5]. The method involves direct oxidation of glucose into gluonic acid at enzyme-less solid electrodes [9]. These electrodes possess a higher surface area and electro catalytic activity with better sensitivity and selectivity over enzymatic glucose sensors. The trends towards the development of non-enzymatic glucose electrodes based on metals and their composites, alloys, and bimetals, metal-metallic oxides, carbon materials, and layered double hydroxides were reviewed in [17]. The study of nanomaterial for non-enzymatic glucose sensors was illustrated [18] and details on recent advances in non-enzymatic glucose sensors were described in [3].

The fabrication of non-enzymatic glucose sensor using copper oxide nanowires (CuO NWs) as in [19] and copper nanowires with multi-walled carbon nanotubes [20] that provided high sensitivity, and fast response time [19,20] were studied. The development of surfactant template assisted platinum nanoparticles (PtNPs) glucose biosensor [21], Ni/multi-walled carbon nanotubes (MWNTs) electrodes [22], graphene-Schottky junction based glucose biosensor [15], Ni3S2 nanosheet array glucose electrodes [23], Copper nanoparticle modified graphene sheets electrode for novel non-enzyme glucose biosensors [24], and hollow Co3O4 microsphere assembled with nanocrystals non-enzymatic biosensor [25] were reported in research publications indicating the possibilities of their commercialization.

The advancements in nanotechnologies, such as nanomaterial, nano porous metals, carbon nanotubes and graphene allowed the developments of non-enzymatic glucose biosensors with higher surface areas, higher sensitivity, and better selectivity [5,18]. Despite the great efforts in scientific publications, commercialization of non-enzymatic biosensors have been a challenging issue. Non-enzymatic glucose sensors hardly catalyse glucose oxidation under normal physiological conditions and lack a perfect match between normal concentrations with analytical ranges of the fabricated sensor, which is difficult for clinical uses [17,18].

3. Non-invasive blood glucose monitoring techniques

3.1 Optical methods

Optical glucose sensing is the most widely studied method, for the development of non-invasive blood glucose measurement. Even though none of the optical based biosensors that meet clinical accuracy are commercialized yet, several researches are undergoing. The Table 2 below provides a brief summary of extensively studied optical based blood glucose monitoring techniques in recent years.
Table 2 Summary of recent advances in optical based blood glucose monitoring techniques

<table>
<thead>
<tr>
<th>Optical technique</th>
<th>Descriptions of principles</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluorescence spectroscopy</td>
<td>Depends on fluorescence where an irradiation of a substance with lower wavelength ultraviolet light source results in an emission of light with different energy and frequency [26–28]. It contains a chemical compound called fluorophores, a component that causes a molecule to absorb the energy of a specific wavelength light and re-emit different energy at specific wavelengths where a change in fluorescence is proportional to glucose concentration [29].</td>
</tr>
<tr>
<td>Raman spectroscopy</td>
<td>Based on an inelastic scattering of laser light passed through human tissue which shifts its frequency and changes wavelength through Raman effect [1]. The shift in frequency is used to observe vibrational, rotational and low-frequency transitions. Molecular vibration affects an emission of scattered light which is used to estimate the concentration of glucose [30].</td>
</tr>
<tr>
<td>Photoacoustic spectroscopy</td>
<td>Measures an acoustic pressure wave produced by an interaction of laser light with tissues. An excitation energy of infrared laser passes through the aqueous glucose solution, where it absorbs light and releases heat energy [31]. This causes volumetric expansion in the light illuminated cylindrical region, generating photoacoustic pressure wave correlated with glucose level [31,32].</td>
</tr>
<tr>
<td>Absorption spectroscopy</td>
<td>Based on absorption, reflection and scattering properties of light by passing infrared light through human tissue with wavelength ranges of near infrared (750nm-2500nm), mid-infrared (2500nm-100,000 nm), or far infrared. The light-tissue interaction produces absorption, reflection, and scattering of the irradiated light source [31,33]. The change in light characteristics correlates with glucose concentration [34].</td>
</tr>
<tr>
<td>Thermal emission spectroscopy</td>
<td>Employs a principle of tympanic membrane thermometers that measure signals of emitted infrared light produced in human tissues due to changes in the concentration of blood glucose [34,35].</td>
</tr>
<tr>
<td>Optical coherence tomography</td>
<td>Utilizes low coherent light source to measure optically scattered signal from human tissue. It involves a combination of backscattered light from tissue with light reflected from the reference arm of the interferometer. The delay correlation between backscattered light from the sample and light reflected from reference arm are measured [30]. An increase in glucose concentration increases the refractive index and causes a change in scattering of light [34].</td>
</tr>
<tr>
<td>Polarimetry</td>
<td>Measures rotation of polarized light as it passes through optically active solutes such as glucose. The polarization of light is dependent on thickness, temperature, and concentration of solutes which is used to determine glucose levels in the blood. It was employed in aqueous humor of the eye [36].</td>
</tr>
<tr>
<td>Ocular spectroscopy</td>
<td>Employs electrochemical biosensor based contact lens to measure glucose concentration in tears [34,37]. A change in wavelength of reflected light determines glucose concentration [38].</td>
</tr>
<tr>
<td>Occlusion spectroscopy</td>
<td>Measures the scattering property of red or near-infrared light passed through a human finger after applying pressure that occludes blood flow [1,39].</td>
</tr>
<tr>
<td>Metabolic Heat Conformation</td>
<td>Involves multivariate mathematical analysis of heat dissipation, the rate of blood flow and degree of oxygen saturation in blood where an oxidation of glucose generates heat whose quantity is correlated with the amount of dissipative oxygen and glucose [40]. It uses humidity, thermal and optical sensors [41].</td>
</tr>
<tr>
<td>Conservation of Energy</td>
<td>It is an extension of metabolic heat conformation that additionally measures basal metabolic rates and heart rates [42].</td>
</tr>
</tbody>
</table>

In the Table 3 below, the advantages and limitations of optical based blood glucose monitoring techniques are described.

Table 3 Summary of advantages and drawbacks of optical based blood glucose sensors

<table>
<thead>
<tr>
<th>Optical technique</th>
<th>Merits</th>
<th>Limitations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluorescence spectroscopy</td>
<td>Extremely sensitive and requires less calibration [27].</td>
<td>Depends on skin colour, thickness, and pigmentation. The fluorophore dye may cause toxicity to tissues [1, 27].</td>
</tr>
<tr>
<td>Raman spectroscopy</td>
<td>Less sensitive to temperature, water, and interference from other light sources [30].</td>
<td>Instability of laser source, poor signal to noise ratio, and longer spectral acquisition time are major drawbacks [30,33].</td>
</tr>
<tr>
<td>Photoacoustic spectroscopy</td>
<td>Has a higher sensitivity [30], and wide wavelength range laser light from Ultraviolet to NIR.</td>
<td>Sensitive to environmental factors. Susceptible to interferences from physiological substances [33].</td>
</tr>
<tr>
<td>Absorption spectroscopy</td>
<td>Less expensive. Possesses good tissue penetration property [34].</td>
<td>Sensitive to environmental factors. Has poor signal to noise ratio (SNR) and affected by tissue compositions [34].</td>
</tr>
</tbody>
</table>
Optical technique | Merits | Limitations
--- | --- | ---
Thermal emission spectroscopy | Requires less calibration. Has good reproducibility [34,35]. | Has poor accuracy. Affected by temperature, movement, and thickness of tissue [35].
Optical coherence tomography | Characterized by good SNR, high resolution, and high penetration. | Sensitive to motion and skin temperature.
Polarimetry | Independent of temperature and PH variations. Easy for miniaturization. | Sensitive to scattering of tissues, and motion. Has poor specificity to glucose molecules.
Ocular spectroscopy | Performed at eye cornea where a scattering of light is low [38]. | There is a time lag between glucose in blood and tear. The lens is uncomfortable for people with diabetes.
Occlusion spectroscopy | Has a good signal to noise ratio. | Requires compensation for signal drift [1].
Metabolic Heat Conformation | Feasible and less expensive. | Suffers from environmental interference.
Conservation of Energy | Has good glucose correlation coefficient [42]. | Affected by environmental variations.

### 3.2 Electromagnetic method

Electromagnetic sensing involves measurement of blood’s dielectric properties detected through changes in eddy currents using electromagnetic coupling between two inductors [43]. The change in blood glucose concentration results in the variation of blood’s dielectric properties such as permittivity and conductivity [44]. This causes changes in the electromagnetic coupling of nearby inductors affecting their impedances which results in variations in resonant frequency. The variations in frequency shift helps to determine blood’s dielectric properties correlated with the concentration of glucose [43, 45]. It involves an application of a signal with a specified frequency at primary inductor and measuring an output from the secondary inductor [35, 43]. This method does not require ionization of substances in the body, and utilization of specified frequency range signal helps suppress interference from biological components in the tissues [30].

The feasibility of electromagnetic sensing for non-invasive blood glucose monitoring was demonstrated in research papers. An electromagnetic non-invasive blood glucose measurement with preliminary results based on a small frequency band of 45MHz was studied in [43]. Ultra-wideband (UWB) microwave blood glucose detection technique using 6.5 GHz signal was proposed by [46] where experimental measurements were performed using realistic earlobe phantom with blood glucose range of 0 mg/dl-400 mg/dl. UWB imaging with a signal having a center frequency of 4.7GHz was used to measure glucose concentration in blood plasma, which exhibited an accuracy of 81% [47]. The design of novel and miniaturized microwave based non-invasive blood glucose monitoring was proposed in [48–50]. Susceptibility of optimal frequency investigation to the temperature and the dependence of dielectric parameters on other components of blood are the main drawbacks of this technique [51].

### 3.3 Bioimpedance spectroscopy

This technique is based on the measurement of impedance as a function of frequency in response to low-intensity current applied across tissue [52, 53]. The change in plasma’s glucose concentration causes an increase in potassium ion concentration and a decrease in sodium ion concentration in red blood cells [54] which eventually results in variations of membrane potential. This variation can be estimated by determining permittivity and conductivity of cell membrane through the dielectric spectrum [52, 55]. Bioimpedance spectroscopy based blood glucose measurement on the human tissue was experimentally demonstrated by [52] which was done by measuring the impedance between frequency ranges of 1MHz and 200MHz to suppress sensitivity of glucose sensor to body’s electrical changes. The design of bioimpedance spectroscopy based non-invasive wearable blood glucose monitor was studied [56]. This method is susceptible to temperature, movement, skin moisture, sweating, and body dehydration.

### 3.4 Reverse iontophoresis

This method uses low electrical current across the skin to withdraw a small amount of glucose with electrochemical glucose sensors worn on the skin. Then electrochemical sensors determine the levels of glucose in the blood [33, 57].

Development of tattoo-based non-invasive glucose sensor with reverse iontophoresis was indicated in [58]. Reverse iontophoresis requires finger prick for periodic calibration, causes skin irritation, has poor accuracy, and environmental variations [1, 59].
3.5 Ultrasound

The low-frequency ultrasound wave is applied across the skin to extract concentration of glucose determined by an electrochemical amperometric biosensor or optical sensor. The feasibility of an amperometric biosensor placed over ultrasonically permeated sites was evaluated by [60]. The drawback of this technique is associated with an amount of extracted glucose concentration. Ultrasound enhances the permeability of skin which extracts only minute volume of glucose. Table 4 provides summary of few non-invasive blood glucose monitors which are based on transdermal and optical methods.

### Table 4 Summary of transdermal non-invasive blood glucose devices

<table>
<thead>
<tr>
<th>Techniques</th>
<th>Device description</th>
<th>Target site</th>
<th>Approvals and status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bioimpedance spectroscopy</td>
<td>Pendragon Medical Ltd, Switzerland: Pendra©</td>
<td>Wrist skin</td>
<td>CE approved in 2003; withdrawn from the market due to poor accuracy in the post-marketing validation study.</td>
</tr>
<tr>
<td>Ultrasound, Electromagnetic and Heat capacity</td>
<td>Integrity Applications, Inc., Israel: GlucoTrack™</td>
<td>Earlobe skin</td>
<td>CE Approved in 2013; claims market availability in few countries [61].</td>
</tr>
<tr>
<td>Radio wave Spectroscopy</td>
<td>MediWise Ltd., London, United Kingdom (UK): Glucowise™</td>
<td>Skin area between thumb and forefinger or earlobe</td>
<td>It is under development. The company plans to launch in late 2018.</td>
</tr>
<tr>
<td>Reverse iontophoresis</td>
<td>Animas Technologies LLC: GlucoWatch© G2 Biographer</td>
<td>Wrist skin</td>
<td>CE and FDA approved; withdrawn from the market due to poor accuracy and skin irritation in 2007.</td>
</tr>
<tr>
<td>Raman spectroscopy</td>
<td>Nemaura Medical Inc., UK: SugarBeat™</td>
<td>Leg, Arm</td>
<td>CE approved in 2016; the company plans to begin its initial launch in the UK due mid-2018.</td>
</tr>
<tr>
<td>Near -infrared spectroscopy</td>
<td>C8 Medisensors, USA</td>
<td>Abdomen skin</td>
<td>CE approved in 2011; the company was closed due to financial problems.</td>
</tr>
<tr>
<td></td>
<td>Cnoga Medical, Israel: TensorTip Combo Glucometer (COG)</td>
<td>Fingertip skin</td>
<td>CE approved in 2014. The post-market evaluation indicated high correlation with reference to invasive devices [62, 63].</td>
</tr>
</tbody>
</table>

4. Major challenges in blood glucose monitoring techniques

Despite great achievements and improvements in the development of blood glucose monitors, several challenges still persist. An invasiveness of currently available glucose monitors decreases user’s compliance, may cause infections, irritation, skin puncture and reduce the frequency of use.

Accuracy is a major challenge in developing blood glucose biosensors. A system accuracy of CE approved invasive blood glucose monitors was evaluated where some devices failed to meet minimum accuracy requirements of DIN EN ISO 15197:2003 and ISO 15197:2013 standards [64, 65]. Most of CE approved non-invasive blood glucose monitors do not meet these standards.

The periodic recalibrations of glucose monitors is another challenging factor. Most of the non-invasive blood glucose monitoring techniques need withdrawal of blood samples and follow complex procedures to calibrate glucose measurement results prior to use [59]. Blood glucose monitors requiring invasive capillary calibration are associated with an increase in cost, discomfort, inconvenience, and complex procedures [9, 66].

Poor signal to noise ratio is another challenging factor in detection and development of non-invasive glucose sensors. Non-invasive glucose monitors lack good linearity, sensitivity, and specificity to glucose molecules, which weakens the signal and an accuracy of glucose estimation. Portability, complexity, reliability, durability, cost effectiveness, and user experience are also determinant parameters in the development of blood glucose monitors.

5. Future perspectives of non-invasive blood glucose monitoring

The growing advances in nanotechnology and biomaterials will transform the future of non-invasive blood glucose monitoring [67]. Glucose biosensors based on nanometals, polymer nanocomposites, carbon nanotubes and graphene are being studied [12–15, 19, 20, 22]; which are promising for the development of portable and convenient painless blood glucose monitors.
Multi-sensing can improve the signal to noise ratio for an estimation of glucose concentration. This was indicated in GlucoTrack® which utilized three non-invasive detection methods: electromagnetic, thermal and ultrasonic [59]. Researchers indicated that multi-sensing integrated with proper estimation algorithm enhances accuracy, improves glucose prediction performance, and reduces time delays in measurement [68–73].

Employing appropriate glucose estimation algorithms and self-calibration models improve calibration required in blood glucose monitors. Partial least-square regression (PLS) and radial basis function (RBF) [74, 75], artificial neural networks (ANN) [76, 77], and pseudo-linear regression with ARMAX model [78, 79], were a few calibration models used for estimation of glucose levels in non-invasive glucose monitors. For instance, FDA has approved calibration free Abbot’s Freestyle Libre Pro™ and Dexcom™ G6 blood glucose monitors. Dexcom™ G6 was launched on June 4, 2018.

6. Conclusion
Through rigorous studies and scientific breakthroughs, sensing and monitoring of blood glucose levels are getting better and more convenient. The trends of recent progress in nanotechnology and miniaturization of biosensors enabled the development of reliable, more accurate, and comprehensive glucose monitoring devices.

Despite the remarkable advances and achievements in the blood glucose monitoring technologies, there are still challenges in the development of non-invasive glucose monitors. Due to lack of precision and clinically acceptable accuracy, non-invasive blood glucose biosensors have never achieved the requirements for market commercialization. There is a high market demand for calibration-free non-invasive blood glucose monitors. To meet this need, an elegant and ingenious technological innovation is highly appreciated for the future development and commercialization of reliable, convenient, clinically accurate, and pain-free blood glucose monitors.

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Conflicts of interest
The authors have no conflicts of interest to declare.

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