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Comparing the Light Response of D-Glucose in Polyacrylamide Hydrogel and Water in NIR Spectral Region by Using an LED Based Portable Device

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ABSTRACT

The spectral properties of the molecules depend on the matrix in which it is present. The interaction of the molecule with the solute molecules affects the vibrational and rotational modes of the molecule. In this study, an absorption-based system was designed to show how the absorbance properties of glucose change in polyacrylamide hydrogel, and the measurements were performed at different wavelengths; 960 nm, 1450 nm, 1550 nm, and 1950 nm. It was observed that the system is sensitive to glucose at 1450 nm and 1950 nm wavelengths in polyacrylamide hydrogel, whereas it is only sensitive at 1450 nm in water which is due to the high absorbance of water at 1950 nm. In polyacrylamide hydrogel, water molecules mostly gather around the polymer chains via electrostatic interactions and the absorbance of water decreases which results in an increasing absorbance of glucose. According to the results, the responsivity of the system at 960 nm and 1550 nm, which are the wavelengths commonly used LED-based systems for measuring glucose in literature, is not high enough for reliable glucose measurements when compared with 1450 nm and 1950 nm.

Keywords: Glucose, infrared absorption, polyacrylamide hydrogel, portable device

INTRODUCTION

Light response of the molecules directly depends on the properties of the matrix (environment) in which the molecules are present. The rigidity, viscosity, polarity, temperature are some of the important parameters which define the matrix. For example, the number of possible vibrational states of a molecule increases with the increasing temperature or decreasing viscosity (Lakowicz, 2006). Therefore, by controlling the parameters of the matrix, the light response of the molecules can be changed and/or adjusted.

Blood measuring devices work with two different methods; these are photometric and electrochemical methods. In the photometric method, sugar in the blood and a particular reagent interact, and the new color formed is analyzed optically by the device (Demitri and Zoubir, 2017). In the electrochemical method, glucose interacts with the test strip and generates an electric current, this current is measured, and the sugar value is calculated (Salacinski *et al.*, 2014).

Blood measurements have played an essential role in the treatment of diabetic patients until now, but continuous measurements make this method painful and uncomfortable for patients considering they need blood samples for measurements (Dalvi, 2013; Ahmad, Kamboh and October, 2015; Javid, Faranak and Zakeri, 2018). Measurements from the blood became a bigger problem in children with diabetes. Thus, more modern measurement methods have become necessary for diabetic patients, enabling comfortable and frequent tests (Fei *et al.*, 2004; Formosa, 2013). In light of the same idea, optical systems that work without taking blood samples from the patient have begun to be studied. Systems that measure optical absorbance, reflectance, and measurements from intercellular fluid (reverse iontophoresis) can be given as examples (Leboulanger, Guy and Delgado-Charro, 2004; Poddar *et al.*, 2008; Zhang *et al.*, 2016). Among these systems, measurement by optical absorbance method was preferred in this paper due to its low cost and easy application (Jung and Hwang, 2013; Xue *et al.*, 2014; Ionescu and Doctorala, 2019). Because it is optically based, it provides excellent ease of use. The low production cost allows better access to people in need (Haxha and Jhoja, 2016). With the increased awareness, the value and the number of optical-based systems are rising every day (Gonzales, Mobashsher and Abbosh, 2019). Optical measurement systems are an active field where research is continuing.

It is aimed in this paper to test and compare the wavelength of the infrared light to be used in different matrices, which are aqueous and PAAm gel, and present it to the literature to enable

further advancement in the area of non-invasive glucose measurements. Therefore, the wavelength at which the absorbance is highest for the glucose molecule will be determined in the artificially designed system that may simulate the tissue. The experiment consists of three stages; (i) preparation of samples in different concentrations, (ii) design and installation of the experimental setup, and (iii) performing the measurements. After the experiment structure was planned, a circuit was designed accordingly. Then a device was designed to hold the electronic circuit and sample together and was printed using a 3D printer. The wavelengths selected in this study were 960 nm, 1450 nm, 1550 nm, and 1950 nm according to the spectrum of the glucose given in (Hotmartua *et al.*, 2015). It was observed that the system is sensitive to glucose at 1450 nm and 1950 nm wavelengths for PAAm gel, whereas it is only sensitive at 1450 nm for water which is due to high absorbance values of free water. According to the results, the responsivity of the system at 960 nm and 1550 nm, which are the wavelengths commonly used in led-based systems for measuring glucose in literature, is not high enough for reliable glucose measurements when compared with 1450 nm and 1950 nm.

MATERIALS and METHODS

Chemicals and Synthesis of PAAm Gels

The monomer (acrylamide, AAm), the initiator (ammonium persulfate, APS), and glucose were supplied by Merck (Darmstadt, Germany). All chemicals were used as received. Distilled water was used when preparing the glucose solutions and for polymerization. At five different concentrations, the aqueous glucose solutions, 20, 40, 60, 80, 100 mg/ml, were prepared in the polystyrene spectrophotometer cuvettes. The polymer concentration was adjusted to 4M and was synthesized via free radical polymerization of AAm. The pre-polymer solutions were prepared by including five different glucose concentrations, 20, 40, 60, 80, 100 mg/ml, and the polymerization was performed in the presence of glucose in the heat bath. The reaction temperature was adjusted to 60 °C, and the reaction took 30 minutes. Since the monomer concentration is high, we obtain a physical gel at the end of the reaction. The polymer samples also were prepared in the polystyrene spectrophotometer cuvettes. The SEM image of the PAAm gel was taken in freeze-dried condition by using Zeiss EVO LS 10 model SEM device.

Experimental Setup

The block diagram of the measurement setup was given in Figure 1. The light source and the photodiode compartments are modular, and they can easily be changed depending on the wavelength. The collimators were used to obtain an almost parallel light beam before and after

the sample. A transimpedance amplifier with an adjustable gain was used in the data acquisition section, as seen in Fig. 2.

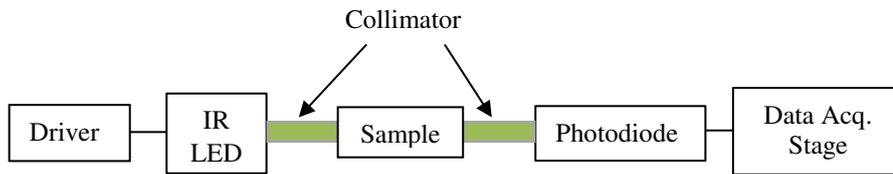


Fig. 1. The block diagram of the experimental setup.

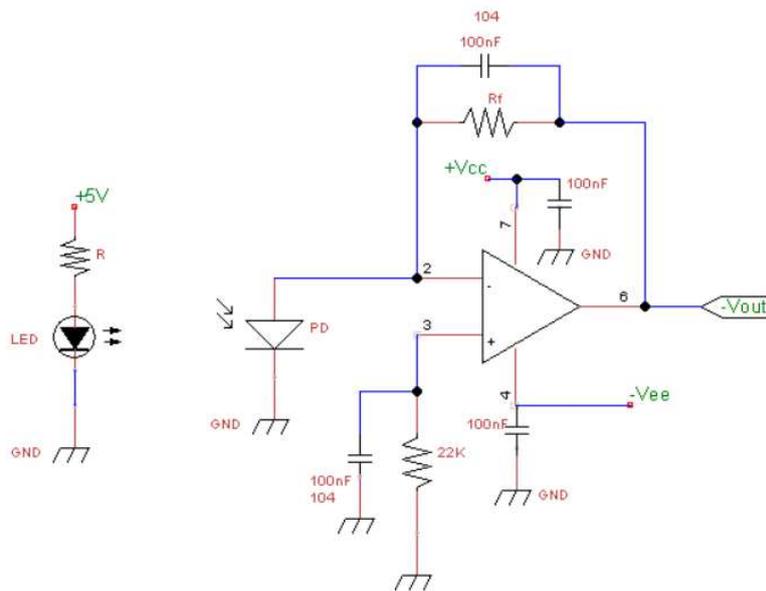


Fig. 2. Circuit scheme of the transimpedance amplifier used in this study.

Due to changing light intensity of the LED and different photodiode responses, circuit gain has been adjusted to the wavelength in which the measurements were performed. Therefore, to obtain accurate results, the gain, which is $-R_f$, must be optimized. The photodiode was used in the photovoltaic mode for the low noise response.

The operational amplifier was selected as TL081CP (ST Microelectronics, General purpose JFET OPAMP), the output voltage is recorded on the computer by using a data logger (UNI-T 71C), and the system is powered by using a low-noise power source (Rigol-DP831A).

The properties of the LEDs and photodiodes used in this study were tabulated in Table 1.

Table 1. Devices used in this study.

Device	Manufacturer	Model	Wavelength (nm)	Radiant Flux (mW)	Active area (mm ²)	R (A/W)
LED	Vishay	TSUS5202	950	15.0	-	-

LED	OSA Opto.	OIS-150 1450P	1400-1500	1.20	-	-
LED	OSA Opto.	OIS-330 1550P	1500-1600	1.05/sr	-	-
LED	Thorlabs	LED1900P	1800-2050	1.00	-	-
PD	Vishay	BPV23F	870-1050	-	4.4	0.6
PD	OSI Opto.	FCI-InGaAs-1000	1100-1620	-	0.79	~ 0.9
PD	Thorlabs	FD05D	900 - 2600	-	0.20	1.3

The LED, PD, and sample holder were designed by SolidWorks 3D design program and printed by FlashForge Inventor model 3D printer. The printed holders were shown in Fig. 3 and it is clearly seen that the system is designed for 10 mm spectrometer cuvettes by which all measurements were performed except the PAAm gel at 1950 nm. At this wavelength, the AAm gel was sandwiched between two glass slides with a light path of 0.4 mm due to the very high absorption in 10 mm cuvette which makes the system unresponsive.

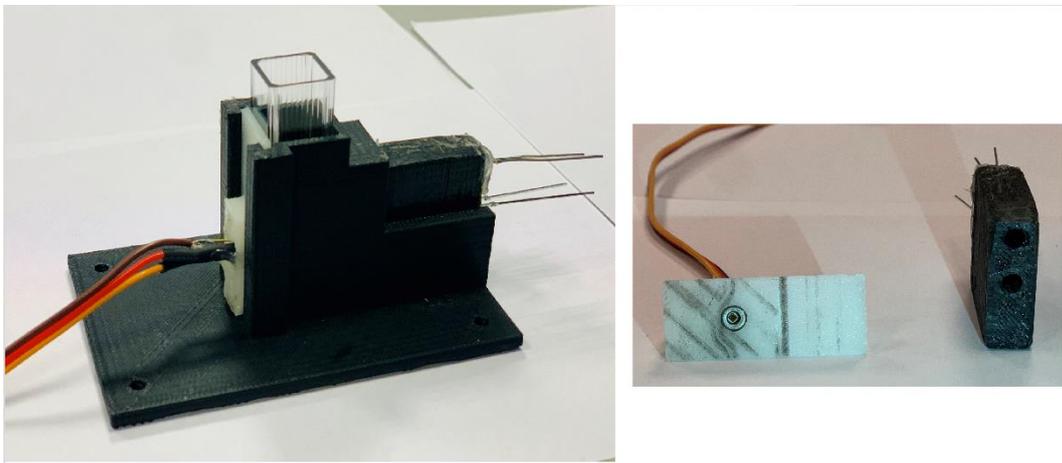


Fig. 3. The holder designed in this study (left) and modular led and PD holders (right).

Measuring NIR Spectra

The NIR spectra of the glucose in water and PAAm gel were taken by using Shimadzu UV-3600 Spectrometer to compare the results obtained by the LED-based system. The spectra of the water samples were taken in quartz cuvettes with a 1 mm light path, and the PAAm gel samples were sandwiched between two glass slides with a light path of 0.4 mm.

Theoretical Model

In this study, the glucose absorption is measured using Beer-Lambert Law which is given in Eq. 1.

$$\frac{I}{I_0} = e^{-\epsilon Cd} \quad (1)$$

here I_0 is the initial light intensity, I is the light intensity after passing the sample, Σ , C and d are molar absorption coefficient, the concentration, and the sample's thickness, respectively.

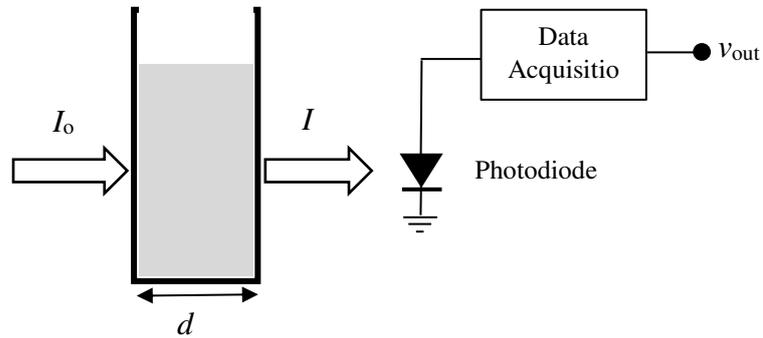


Fig. 4. Schematic representation of the measurement model.

The output voltage of the amplifier, which was given in Fig. 2, depending on the photodiode current is given in Eq. 2.

$$v_{out} = -i_{pd}R_f \quad (2)$$

where V_{out} , i_{pd} , and R_f are the output potential, the photodiode current, and the feedback resistor, respectively. As seen in this equation, the output potential is directly proportional to the photodiode current.

The photocurrent, i_{pd} , induced in the photodiode is proportional to the light intensity [20]. According to Eq. 2, measured potential (v_{out}) depends on this photocurrent. Therefore, Eq. 1 can be written in terms of the potential given in Eq. 3.

$$\frac{v_{out}(C)}{v_{out}(0)} = e^{-\epsilon Cd} \quad (3)$$

where $v_{out}(0)$ and $v_{out}(C)$ are the output potential measured without and with glucose, respectively. By taking the logarithm of both sides of Eq. 3, the absorbance can be obtained as given in Figs. 8 and 10.

$$A = \log_{10}\left(\frac{v_{out}(0)}{v_{out}(C)}\right) = \epsilon Cd \quad (4)$$

The molar absorption coefficient of the glucose for each wavelength was calculated by fitting the measured data to Beer-Lambert's law given in Eq. 4.

RESULTS and DISCUSSION

The glucose absorption measurements were performed at four different wavelengths, 960 nm, 1450 nm, 1550 nm, and 1900 nm, to carry out the most sensitive wavelength for detecting the glucose in water and in PAAm gel. The wavelengths were selected based on the data given in reference (Hotmartua *et al.*, 2015) and the spectra given in Figs. 5 and 6. In fact, glucose has higher absorbance at higher wavelengths, but the photodiode and light source for this region is costly; therefore, it is not cost-effective for a wearable device.

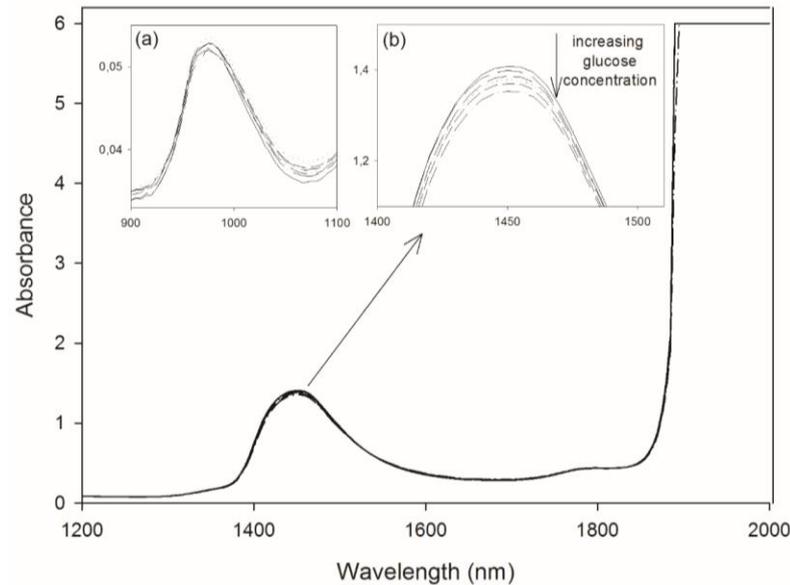


Fig. 5. NIR spectra of D-Glucose in water at different concentrations. The absorption at 960 nm and 1450 nm were magnified as seen in inset figures (a) and (b), respectively.

When the NIR spectra given in Figs. 5 and 6 were compared, it is clearly seen that the overall absorption in water is much higher than in PAAm gel and it saturates in water above 1900 nm. This is due to the high absorption property of water in this wavelength region and the absorption is dominated by water which decreases the response of the system against glucose. In PAAm gel, the overall absorption is lower and the responsivity of the system against glucose is higher, that is the change in the absorption is higher for changing concentration of the glucose (Fig. 6). In addition, the absorption peak of glucose at 1950 nm is visible for glucose in PAAm gel which is the most efficient wavelength for glucose measuring.

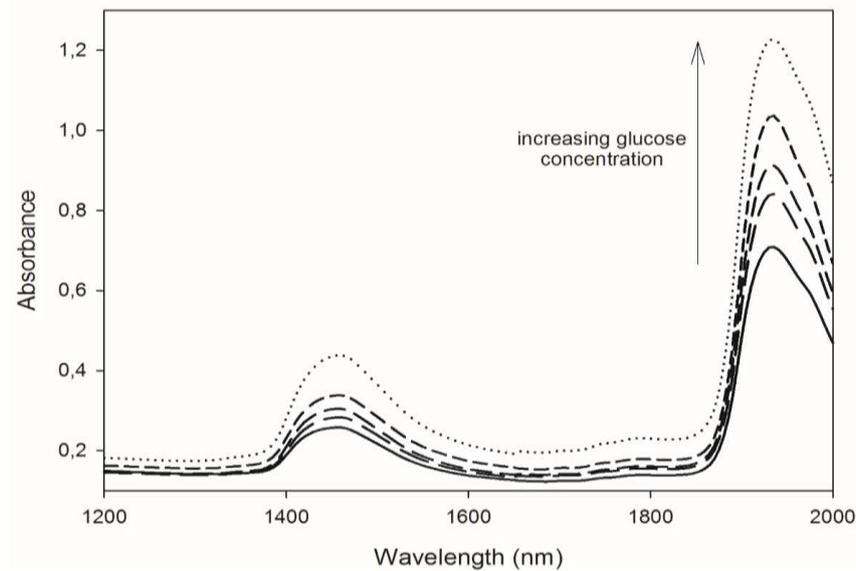


Fig. 6. NIR spectra of D-Glucose in PAAm gel at different concentrations.

Before starting the absorbance measurements for glucose, The aging of glucose in water was studied first. For this purpose, five glucose samples with different concentrations, 20, 40, 60, 80, and 100 mg/mL were prepared, and the output potential for each sample was measured for 20 days at 23 °C. The slopes of the concentration-output voltage graph were given in Table 2. It is seen from the values in this Table that the slope did not change considerably in the first ten days, but it changes considerably at the end of the 20 days. Therefore, it was concluded that the aqueous glucose solution should not be used after ten days.

Table 2. The slope of the concentration-output voltage graph with respect to time.

Time (Day)	$\Delta V / \Delta C$ (Volt. mL/mg)
1	0.0003
3	0.0004
5	0.0003
10	0.0002
20	0.0019

The background-subtracted potential values, which were measured by using an LED-based system for glucose in water, at 960 nm, 1450 nm, 1550 nm, and 1950 nm were given in Fig. 7 as a function of glucose concentration. The potential changes linearly with the concentration of the glucose in water and the slopes were calculated as 0.0287 mV/mM, 0.0422 mV/mM, and 0.0071 mV/mM for 960 nm, 1450 nm, and 1550 nm, respectively. The highest slope was

obtained for 1450 nm, and it is seen clearly from Fig. 7 that the system's response to the glucose is high at this wavelength. It can be concluded that this wavelength is more suitable for designing a system for measuring the glucose for wearable technology. This is also clearly seen from the absorption spectrum of the glucose in water in the NIR region given in Fig. 5 where the glucose absorbs the light more efficiently at 1450 nm. Additionally, the LED-based system does not respond to the concentration changes of the glucose in water at 1950 nm due to the high absorption of water at this wavelength which is in accordance with the results given in Fig. 5 where the absorption reached saturation above 1900 nm.

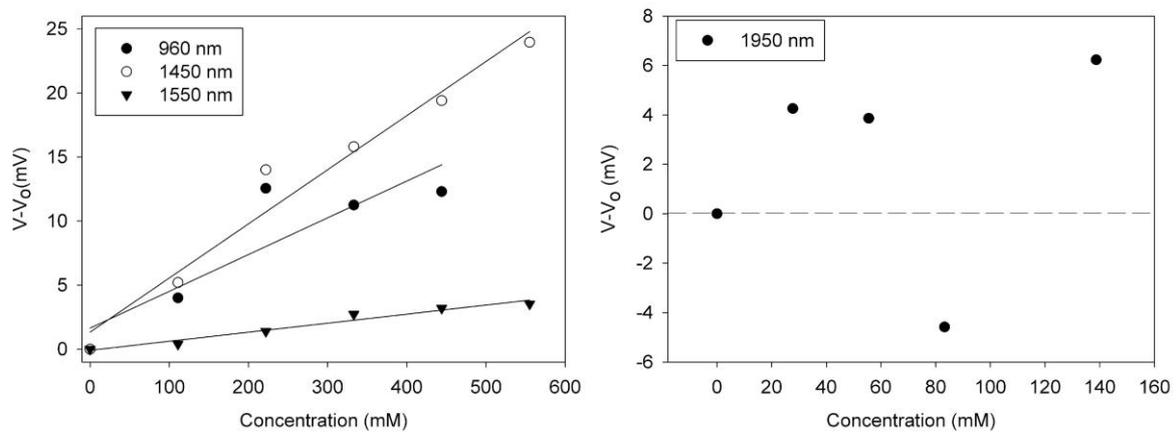


Fig. 7. The background-subtracted potential change as a function of glucose concentration in water at different wavelengths. Left: 960 nm, 1450 nm, 1550 nm, right: 1950 nm. The measurements were performed by using an LED-based system designed in this study.

The absorbances as a function of glucose concentration at 960 nm, 1450 nm, and 1550 nm were given in Fig. 8. The absorbance changes linearly with the concentration and decreases with increasing glucose concentration contrary to the expectation. However, this unexpected behavior was also observed by different groups in literature and it was explained as the waveguide effect of the glucose molecules in water (Javid, Faranak and Zakeri, 2018). That is, when the concentration of the glucose increases in water, they are aligned to construct a waveguide-like structure for electromagnetic waves and leads to an increase in the transmitted light intensity through the sample.

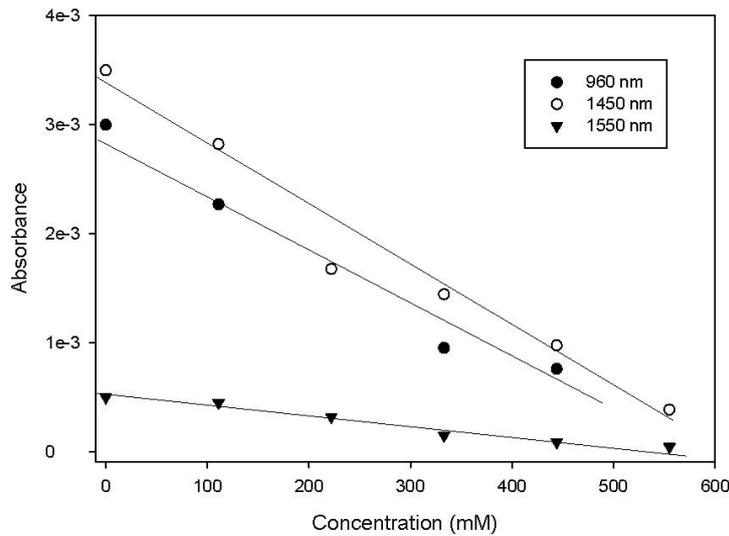


Fig. 8. The absorbance of glucose in water as a function of glucose concentration at different wavelengths. The measurements were performed by using an LED-based system designed in this study.

In water, since the absorbance dependence on the glucose concentration is completely different, enhancement factors, v , were calculated from the slopes of Fig. 8 instead of molar absorption coefficients and they were found to be 5.22×10^{-3} , 5.49×10^{-3} , and $1.01 \times 10^{-3} \text{ mM}^{-1}$ for 960 nm, 1450 nm, and 1550 nm, respectively. The enhancement factor is higher at 1450 nm than the other wavelengths. When the absorption spectrum given in Fig. 5 is examined in detail, it is seen that the absorption is higher at 1450 nm, which shows that the system designed in this study gives results that are in accordance with the commercial spectrometer.

The potential changes, which were measured by using an LED-based system, as a function of glucose concentration in PAAm gel were given in Fig. 9 at different wavelengths. The system's responsivity to the glucose was found to be 0.0549 mV/mM, 0.1563 mV/mM, 0.0939, and 0.3242 mV/mM for 960 nm, 1450 nm, 1550 nm, and 1950 nm, respectively. This result clearly shows that the system's responsivity to the glucose is highest at 1950 nm which couldn't be observed in water due to the high absorption.

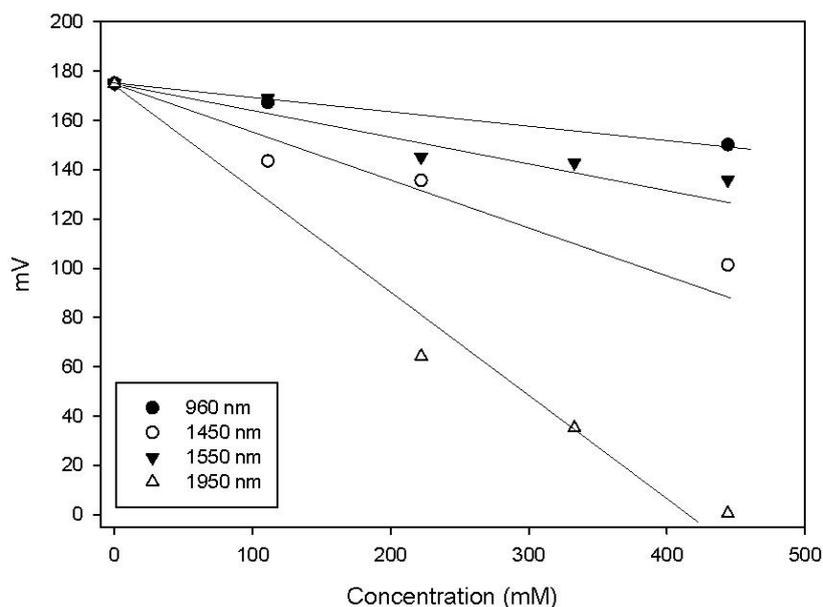


Fig. 9. The background-subtracted potential change as a function of glucose concentration in PAAM gel at different wavelengths. The measurements were performed by using LED based system designed in this study.

The absorbance variations as a function of glucose concentration in PAAM gel were given in Fig. 10 at different wavelengths. Here, it is seen that the change at 1950 nm is more significant which is in accordance with the spectra given in Fig. 6. When the absorbance values given in Figs. 8 and 10 were compared, it is clearly seen that the glucose absorbs light more in PAAM gel than in water.

The responsivities and calculated molar absorption coefficients were tabulated in Table 3. The responsivity of the LED system is higher at all wavelengths in PAAM gel and it is highest at 1950 nm in gel. This is due to the high absorption of the free water when compared with gel. In addition, the molar absorption coefficients calculated by using the LED based system and the NIR Spectrometer are close to each other which shows that the LED based system designed in this study works correctly and gives reliable results.

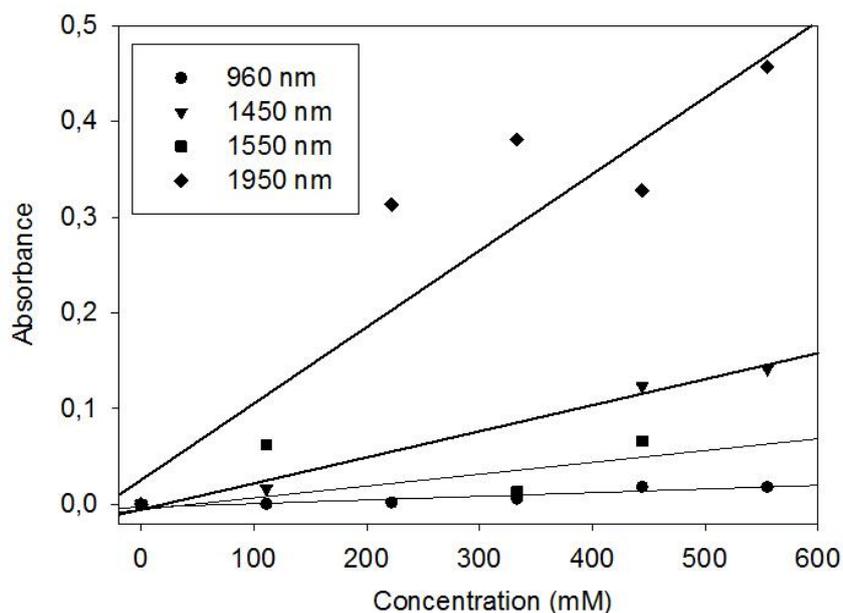


Fig. 10. The absorbance of glucose in PAAm gel as a function of glucose concentration.

The SEM image of the PAAm gel was shown in Fig. 11 and it is clearly seen that it has a very porous structure. The glucose molecules can easily be trapped in the pores of the gel. It is well known in literature that PAAm gel is highly hydrophilic and the positively charged amine groups on the polymer chains interact with the water electrostatically which gathers the water molecules around the polymer chains (Yilmaz *et al.*, 2009).

Table 3. The responsivity (R) and the molar absorption coefficients (ϵ) of glucose at different wavelengths. (NM: Couldn't be measured)

Wavelength (nm)	R_{water} (mV/mM)	R_{gel} (mV/mM)	LED based system	NIR Spectrometer
			$\epsilon_{gel} \times 10^{-3}$ ($cm^{-1}mM^{-1}$)	$\epsilon_{gel} \times 10^{-3}$ ($cm^{-1}mM^{-1}$)
960	0.0287	0.0549	1.74 ± 0.25	2.32 ± 0.37
1450	0.0422	0.1563	5.21 ± 0.62	5.88 ± 0.39
1550	0.0071	0.0939	3.57 ± 0.76	3.37 ± 0.50
1950	NM	0.3242	26.45 ± 2.16	27.73 ± 1.56

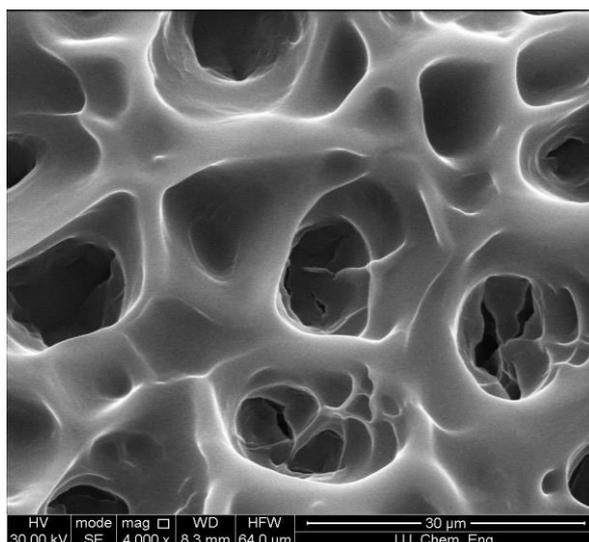
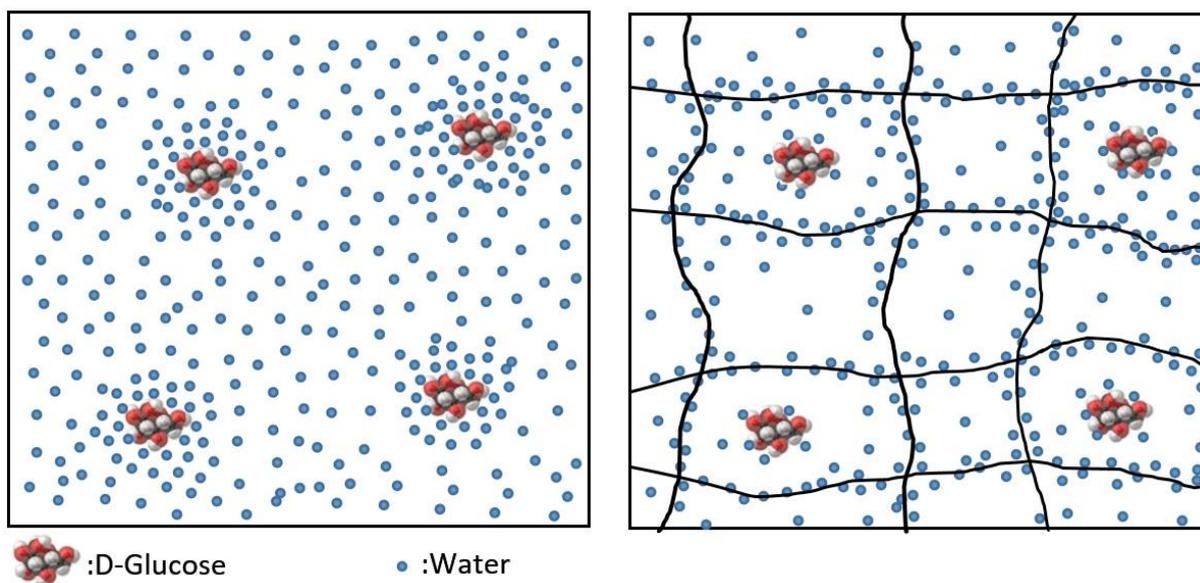


Fig. 11. SEM image of the PAAm gel. The pores are clearly visible in the gel.

Possible mechanism which affects the light response of the glucose in water and in PAAm gel can be depicted as seen in Scheme 1. In the left scheme, the distribution of the glucose dissolved in water was shown. Here, the water in the left scheme was named as free water to be able to distinguish from the water in PAAm gel. In the left scheme, water molecules are distributed almost homogeneously and the glucose molecules are mostly surrounded by water. During the light absorption experiments performed in the free water system, while the most of the light is absorbed by water molecules only small amount of light is absorbed by the glucose due to the shielding effect of water. This description is consistent with the results given in Fig. 5 where the absorption of water is very high when compared with glucose. In the right scheme, water molecules are mostly gathered around the polymer chains due to the electrostatic interactions between water and positively charged amine groups of polymer chains. In the pores of the gel, since some part of the water molecules are gathered around the polymer chains, the number density of the glucose molecules increases relatively when compared with water molecules. During the light absorption experiments performed in the PAAm gel, the overall absorption decreases since some part of the water molecules are shielded by the polymer chains and the absorption by the glucose molecules increases due to the decreased number of water molecules which shield the glucose molecules before. This description is also consistent with the results given in Fig. 6 where the overall absorption decreased and the glucose absorption became more visible.



Scheme 1. The schematic representation to describe the effect of the PAAm gel on the light response of glucose in free water (left) and in PAAm gel (right). Lines in right indicate the polymer chains.

Conclusions

It was concluded that the water plays a crucial role in the measurement of the glucose by using an optical system based on absorption/transmission of light. Water absorbs light in NIR region considerably and makes the optical system blind against the glucose as observed in the experiments performed in this study. To be able to increase the responsivity of the optical system against glucose, water molecules should be passivated. In this study, we showed that the PAAm gel can be used for passivating the water molecules where the water molecules are gathered around the polymer chains due to electrostatic interactions and polymer chains prevent the water molecules to absorb light.

According to the results, it was concluded that the 1950 nm wavelength is the most efficient wavelength. However, this wavelength cannot be used in water due to the very high absorption of water at this wavelength and the light at 1450 nm wavelength should be used for the system used for water.

It can be said that the system designed in this study is appropriate for detecting glucose. Therefore, the results of this study will provide a basis for future works about wearable non-invasive glucose measuring devices.

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Statements and Declarations

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Competing Interests

The authors have no relevant financial or non-financial interests to disclose.

Author Contributions

All authors contributed to the study conception, design and analysis. The 3D design of the system and the electronic control card design and production were performed by Ali Gelir, Onur Burak Ozdemir, Omer F. Kadi, and Kadir B. Yildirim, polymer hydrogel synthesis and material preparation were performed by Ali Gelir, Onur Burak Ozdemir and Sinem N. Seyhan, absorption measurements were performed by Ali Gelir, Onur Burak Ozdemir and Sedat Ozdemir. All authors read and approved the final manuscript.

Data availability. The datasets generated during the current study are available from the corresponding author on reasonable request..