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Detection of HbA1c in Blood Using Diode Laser (491) nm

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ABSTRACT
Laser radiation is widely used in several applications like medicine, biology and industry. Laser biosensor is one of the tools in the biological applications. A laser biosensor is designed with different lengths of solid core photonic crystal fibers (LMA-10) (1.5cm, 1cm and 0.5cm) to be used for different tests. For the blood test, the highest sensitivity of 153 ABS/RIU was found for the (1.5cm) length. Moreover, the biosensor was used for the detection of HbA1c for the diagnosis of diabetes being a recognized valuable tool in diabetes management. Blood samples been taken from healthy people and others suffering from diabetes people. The results showed that the presence of diabetes led to increasing the output intensity of the laser beam as a results of enhancing the nonlinearity . The sensitivities of diabetes is 28.934 ABS/ RIU.

Keywords: HbA1c; Diode Laser; diabetes; LMA-10 Photonic crystal fiber

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INTRODUCTION

Parts of the laser biosensor architecture are less costly, more reliable, and more precise than test strips. There are a range of biosensors that use methods of laser-based-detection, such as SPR , waveguides, optical fiber, etc. In many fields, laser-based biosensors, including immunoassays and drug screening, play an important role because of their high sensitivity and accuracy(1). By using optical fibers, most important laser-based-detection approach utilized in many instruments coud be miniaturized. In biosensors, the laser detection method depend on evanescent waves is usualy used(2). Laser biosensor has become very useful instruments in different fields in the last years, like pharmaceutical research, analytical biochemistry, and food-environmental expermints(3), and diagnostic methods(4). In applications such as disease observations, drug detection, pollutant detection, disease by microorganisms and biomarkers which are markers of a disease in body fluids (urine, saliva, blood, sweat), biosensors are used(5).

HbA1c is a small part of adult hemoglobin, that is formed from hemoglobin and glucose slowly and non-enzymatically. HbA1c is produced over the lifetime of the erythrocyte cause erythrocytes are permeable to glucose; the rate of formation of its is proportional directly to the amount of ambient glucose(6). The quantities of HbA1c therefore give a 'glycemic past' of the past 120 days, which would be the average life of erythrocytes. Whilst also HbA1c reflects glycemia over the previous Twelve weeks, it is weighted to the most recent Four weeks. Blood glucose and blood or urine ketone monitoring provide helpful info for day to day diabetes management, where HbA1c gives crucial data on average glycemic regulatory oversight. It is an important component in blood of diabetes patient and is used to track long-term glycemic regulation and to assess the risk of developing complications of diabetes(7).This work aims to construct of a laser biosensor based on in-line Mach-Zehnder interferometer (Micro-Holes Collapsing) using a solid core photonic crystal fiber (LMA-10). The laser biosensor is used for Sensing for HbA1c concentration measurement.

Sample Collection:

The HbA1c concentrations were estimated by using (4 ml) of fasting venous blood and then performed by Mach-Zehnder interferometer (Micro-Holes Collapsing) using a solid core photonic crystal fiber (LMA-10).

Optical Properties Measurments:

Using a UV-Vis spectrophotometer, the transmission spectrum was collected. By the z-scan measurments presented by Sheik Bahae et al.(8), the nonlinear optical tests were carried out. As aresult of the occurrence of optical nonlinearity, the Z-scan technique relies on the spatial beam profile and reduction of the Gaussian beam in the far-field. In Z-scan experiment, the nanosecond SHG-Nd: YAG laser was utilized. With a beam diameter of < 1.5 mm, a beam divergence of 0.711 mrad and a maximum energy of 120 mJ, this laser has a Gaussian profile. The intensity of the laser also can be calculated, at the focal point being around 17 GW/cm^2. For closed aperture measurments, an aperture with a pinhole size was set.The schematic diagram of the Z-scan technique is depicted in Fig.1. The alteration in the transmitted laser intensity is verified utilizing an optical detector (RJ-7610) as a function of the sample location.

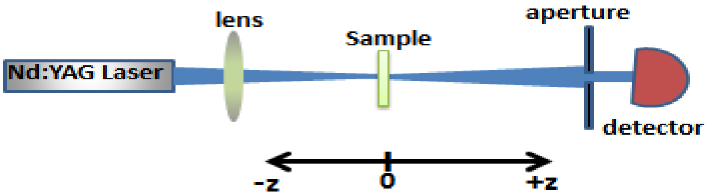

Fig.1: Z-scan experimental setup of z-scan.

Biophotonic Sensor Set-up:

In-Line PCF Mach-Zehnder Modal Interferometer with Mico-Hole Collapsing: Micro-hole collapsing is a simple kind of MZI because its fabrication only need to cleave and splicing the optical fiber(9). the LMA-10 photonic crystal fiber spliced using fusion splicer type (FSM 60 S) to the SMF-28. Collapsing technique can be implemented in this sensor. PCF sensor used with length 1.5cm because it proved to be the most sensitive sensor for changing the refractive index of the biological sample. The light source been used in this experiment is green laser (SHG Nd:YAG laser) with λ=532nm and output power=12.2nw which is shown in figure (2).The beam of the light emitted from the laser source toward the spectrum analyzer transmitting within the PCFs, in which the blood sample exists above. The detected signal could be obtained from the spectrometer.

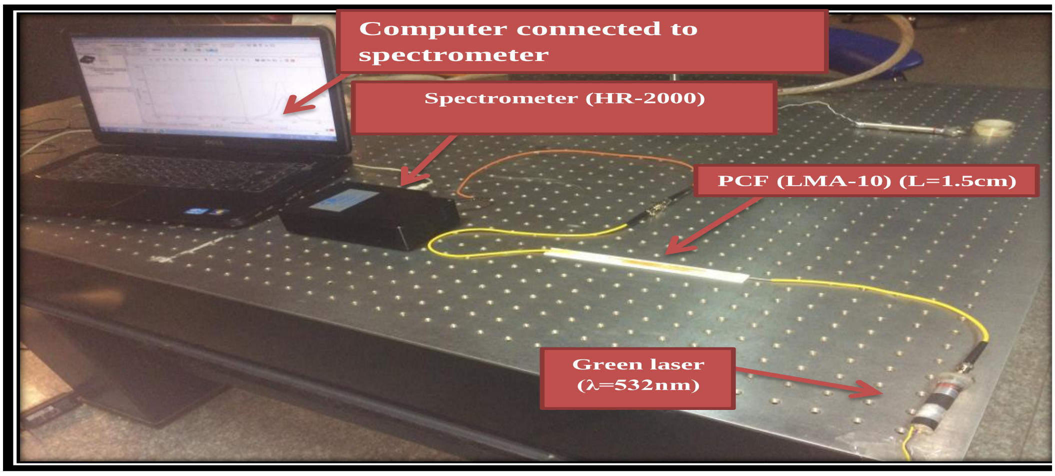

Fig. 2. The experimental setup of biophotonic sensor

RESULTS AND DISCUSSION

Optical properties of Blood Samples:

The absorption spectra for blood samples are measured using (T60 UV-VIS-Near IR) spectrophotometer to select the suitable wavelength of laser source for the detection set-up. Figure (3) show the absorption spectra for the blood samples.

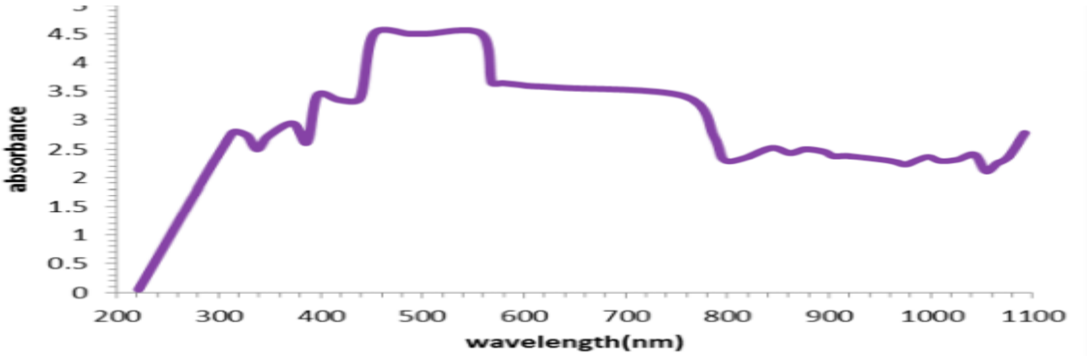

Figure (3): Absorption spectrum for normal blood sample.

By using the closed-aperture Z-scan experiment, the nonlinear refractive index(n_2) alterations can be estimated, when the blood sample is moved along the z and the transmitted intensity is reported in the far-field via an aperture. The normalized transmittance (Fig.4) is plotted against the approximate z sample position for the focal plane. The sign of the nonlinear refractive index n2 relating to self-focusing or defocusing, where the signature of the sample can be shown, is taken from this plot. The sample generates both the focusing and the defocusing in the laser beam, depending on the nonlinear refraction of the matter. By using equ.(1) (n_2) calculated [9]:

$$n_2 = \frac{2\pi \cdot |\Delta\emptyset_0|}{\lambda I_0 L_{eff}} \quad \text{............... (1)}$$

where I_0 is the intensity of laser beam at focusing (Z=0), and L_{eff} means to the effective length inside the sample which is given by[9]:

$$L_{eff} = (1 - \exp -\alpha_0 L)/\alpha_0 \quad \text{...............(2)}$$

where *L* is the geometrical length of blood sample. The term $\Delta\emptyset_0$ means phase shift, and it is calculated by[9,10]:

$$|\Delta\emptyset_0| = \frac{\Delta T_{p-v}}{0.406(1 - S_T)^{0.25}} \quad \text{...............(3)}$$

ΔT_{p-v} is the transmittance difference between the maximum value and the minimum values. The resulting behaviour of the closed aperture Z-scan are shown in Fig.4. From these measurements, blood samples are found to be self-focusing material.

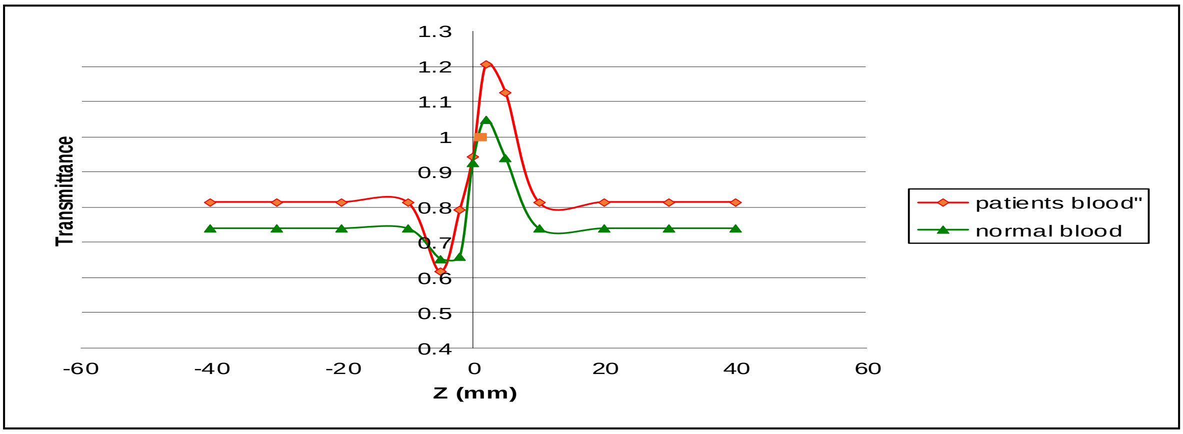

Fig.(4): closed aperature z-scan

An open-aperture Z-scan experiment is measured by removed the aperture in Fig.4; thus the transmitted beam enters the photodetector after passing the blood sample. For the blood samples, the normalized transmittance is plotted against the z-location of the sample shown in Fig.5. The nonlinear optical response of the blood sample to the light beam was confirmed by the occurrence of a peak or a valley in an open aperture[11]. By fitting the experimental data using the following equ.(4) [9], the magnitude of the nonlinear absorption coefficient can be calculated:

$$T(Z) = \sum_m \frac{\left(\frac{\beta I_0 L_{eff}}{1+\left(\frac{Z}{Z_0}\right)^2}\right)^m}{(m+1)^{3/2}} \quad ....(4)$$

In addition, the theoretical fitting is carried out on the basis of the nonlinear equation of propagation concerning two-photon absorption (2PA) or the saturable absorption given by[9]:

$$\alpha(I) = \frac{\alpha_0}{1+\left(I_0/I_s\right)} + \beta I_0 \quad ....(5)$$

$$T_{SA}(Z) = 1 + \frac{I_0}{I_s}\left(1+\frac{Z^2}{Z_0^2}\right) \quad ....(6)$$

where $\alpha(I)$ the intensity dependent absorption, and I_s is the saturation intensity [9].

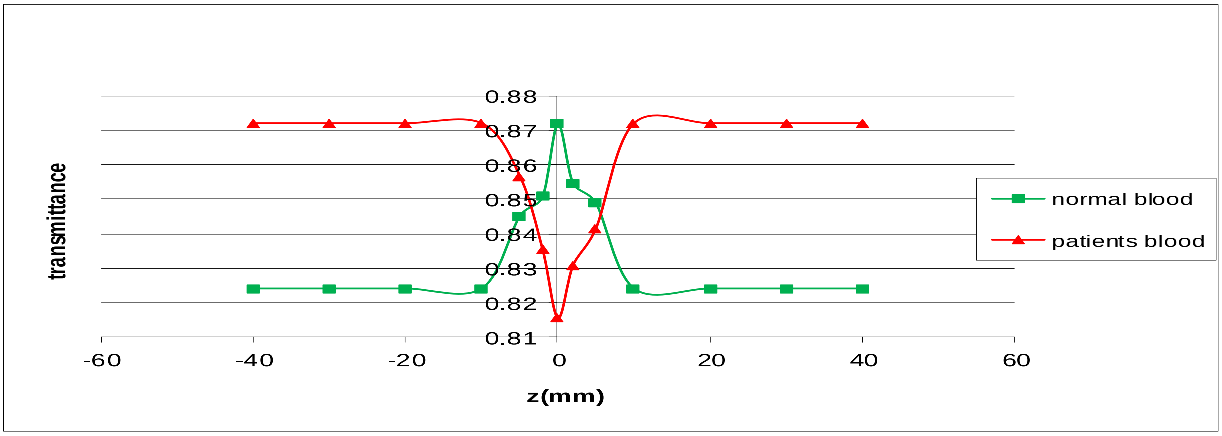

Fig. 5: open aperature z-scan

The open-aperture Z-scan curves for samples excited at an energy of approximately 120 mJ are shown in Fig.5. With two photon absorption or free-carrier absorption, the reverse saturable absorber effect can be considered. The third-order susceptibility results of the blood samples. Table 3 illustrates the measured effects. The susceptibility of the 3rd is determined directly from the following equation:

$$\chi^{(3)} = \sqrt{(Re\chi^{(3)})^2 + (Im\chi^{(3)})^2} \quad ....(7)$$

where,

The real part of third-order susceptibility:

$$Re\chi^{(3)} = 10^{-4}\frac{\varepsilon_0 c^2 n_0^2 n_2}{\pi} \quad ....(8)$$

The imaginary part of third-order susceptibility:

$$Im\chi^{(3)} = 10^{-2}\frac{\varepsilon_0 c^2 n_0^2 \lambda \beta}{4\pi^2} \quad ....(9)$$

The (real and imaginary) parts of third order nonlinear susceptibility have been calculated as about 10^{-10} esu and 10^{-9} esu, respectively. So, the total $\chi^{(3)}$ is in the order of 10^{-9} esu, as presented in Table 1.

Table1: The nonlinear properties of the normal and patients blood sample.

Samples	n_2 (cm²/W)	β (cm/W)	$\chi^{(3)}$ (e.s.u.)
Patients blood sample	0.0551	4034.28	3.9*10^-9
Normal blood sample	0.0393	3434.28	1.6*10^-9

Biophotonic Sensor Results:
The laser biosensor with (1.5cm) PCF length and with 532nm wavelength is used to measuring the Hb concentration. Different blood samples with different Hb concentration had been taken from several people. Transmission spectra of 1.5cm LMA-10 PCF laser biosensor for sensing blood sample with different HbA1C concentration can be shown in fig.(6). The values of Hb concentration have been known by the blood picture examination in laboratory of pathological analyzes. The refractive indices of different values of Hb concentrations are shown in table (2).

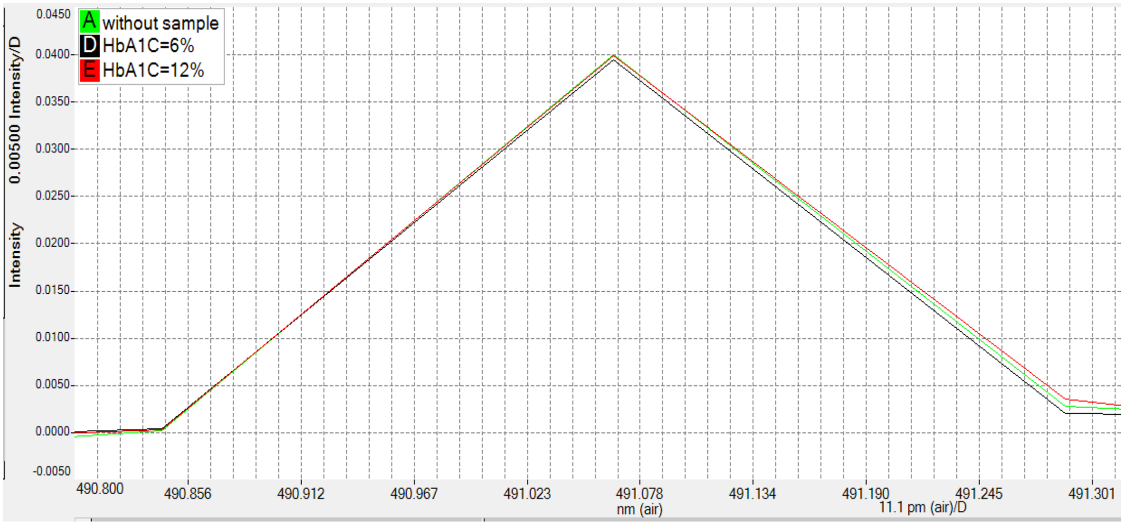

Fig. 6. Transmission spectra of 1.5cm LMA-10 PCF laser biosensor for sensing blood sample with different HbA1C concentration

Table 2. Represented the optical response for different HbA1C concentration.

N0. Of sample	HbA1C concentration %	Central wavelength nm	Wavelength shift $|\Delta\lambda=\lambda_{air}-\lambda_{sample}|$
1	**6**	**491.0690622**	**0.00142**
2	**12**	**491.0692908**	**0.003708**

CONCLUSION

In summary, the linear, and nonlinear optical responses of normal and patients blood samples have been investigated using spectrophotometer, and Z-scan techniques. From these studies, the samples reveal an nonlinear behaviour, broad optical absorption peak within the wavelength region of 300–900 nm, with a high nonlinear optical response. blood behaves as nonlinear refraction and absorption which the competition of self focusing and self-defocusing with saturable and reverse saturable absorption have been studied.

ETHICAL CLEARANCE

The study have the acceptance of Minstory of Health in Iraq, Institutional Human Ethics Committee.

CONFLICT OF INTEREST
There were no conflicts of interest.

SOURCE OF FUNDING
Authors declared that a self-fund have been utilized for this research.

REFERENCES

1. Shriver-Lake LC, Breslin KA, Charles PT, Conrad DW, Golden JP, and Ligler FS .Detection Of Tnt In Water Using An Evanescent-Wave Fiberoptic Biosensor . Anal. Chem.1995; 67: 2431–2435.
2. Dhadwal HS, Kemp P, Aller J, and Dantzler MM.Capillary waveguide nucleic acid based biosensor. Analytica Chimica Acta.2004; 501(2): 205–217.
3. Ciminelli C, Campanella CM, Dell'Olio F, Campanella CE, and Armenise MN.Label-free optical resonant sensors for biochemical applications.Progress in Quantum Electronics. 2013; 37(2): 51–107.
4. Banerjee A., Mukherjee S, Verma RK, Jana B, Khan TK, Chakroborty M, Das R, Biswas S, Saxena A, Singh V, Rajput RS, Tewari P, Kumar, S Saxena, Ghosh AK, John J, and Gupta-Bhaya P.Fiber optic sensing of liquid refractive index.Sensors and Actuators, B: Chemical. 2007; 123(1):594–605.
5. Zhang X, Ju H, and Wang J. Biosensors and Their Biomedical Applications. J. Electrochemical Sensors . 2008; 53(9).
6. Prasad PN. Introduction to Biophotonics. Optical Biosensors J. 2004; 356: 311–356.
7. Zhang Y, Zhang B, and White HS.Electrochemistry of nanopore electrodes in low ionic strength solutions. Journal of Physical Chemistry B.2006; 110(4):1768–1774.
8. Sheikbahae M, Said AA, Wei TH, Wu YY, Hagan DJ, Soileau MJ and Vanstryland EW.*Z-Scan – A simple and sensitive technique for non-linear refraction measurements*.Spie - Int Soc Optical Engineering, Bellingham;1990.
9. Belal Sara J, Alameri Layla M , Rashid Fareed F and Mansour Tahreer S, Laser Biosensor as for Pregnancy Test by Using Photonic Crystal Fiber.International Journal of Medical Research & Health Sciences.2019; 8(2): 41-49.
10. Rawaa A Faris, Zainab F Al-Bawi; Optical Properties for Prepared Silver Nanocomposite. 1st edition LAP LAMBERT Academic Publishing;2017.
11. Rawaa A Faris, Zainab F Mahdi, Mohammad DAH. Immobilised Gold Nanostructures on Printing Paper for Lable-Free Surface-enhanced Raman Spectroscopy.IOP Conference Series: Materials Science and Engineering.2020;871(1):012- 019.