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NIR Photoacoustic Spectroscopy for Non-Invasive Glucose Measurement

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***Abstract*— The use of near infra red (NIR) photoacoustic spectroscopy (PAS) for continuous non-invasive glucose measurement is outlined in the paper. A photoacoustic (PA) measurement apparatus was constructed and PA measurements were made on glucose solutions at multiple NIR excitation wavelengths. A variety of time and frequency domain features, including amplitude and area based features, were extracted from the PA measurements. These features were observed to be proportional to the glucose concentration of the sample. PA measurements from samples of whole blood at different glucose concentrations showed similar results. Subsequently, in vivo PA measurements made on a cohort of 30 volunteers were calibrated using a quadratic fit, and the results were compared to reference glucose concentrations made using a regular blood glucose meter. A comparison of 196 measurement pairs of predicted and reference glucose concentrations using a Clarke Error Grid gave a point distribution of 87.24% and 12.76% over zones A and B of the grid, with no measurement pairs falling in unacceptable zones C-E of the error grid. The predicted measurements had a mean absolute difference (MAD) of 12.57 $\pm$ 13.90 mg/dl and a mean absolute relative difference (MARD) of 9.61% $\pm$ 10.55%. This is an improvement over previous results obtained using PAS and other non-invasive techniques, validating the potential of PAS for continuous non-invasive glucose monitoring.**

I. Introduction

Diabetes afflicts nearly 387 million people worldwide and caused an estimated 4.9 million deaths in 2014[1]. Diabetes results in hyperglycaemia, which leads to impaired vision and renal function in addition to a range of micro- and macro- vascular complications. Regular glucose monitoring combined with intensive insulin therapy to keep blood glucose levels in the normal physiological range can help delay onset and progression of complications[2]. Currently available glucose monitors use electrochemical testing strips to analyse blood drops drawn by the diabetics at regular intervals throughout the day. These methods have a high cost per measurement and are painful to implement, which leads to irregular testing and poor treatment outcomes.

Non-invasive glucose monitoring can provide sample-free glucose measurements without any discomfort to the patient, thus allowing for continuous monitoring and improved outcomes. Presently, a range of analytical techniques are being investigated for non-invasive blood glucose monitoring[3], [4]. We have selected photoacoustic spectroscopy (PAS) for investigation as it is capable of measuring glucose-induced minute changes in the optical absorption coefficient of a sample, and is relatively unaffected by optical scattering by tissues. It is also non-destructive in nature and needs little or no sample preparation before measurement.

The phenomenon of generation of acoustic waves in matter due to excitation by modulated optical radiation is known as the photoacoustic (PA) effect. Irradiation of a sample with optical radiation leads to excitation of the sample, followed by non-radiative relaxation and heat release. This results in thermoelastic expansion and generation of an acoustic wave in the sample. The PA effect was first reported by Bell[5], but gathered pace after the discovery of the laser, which provided an intense optical source capable of inducing PA waves in matter. Rosencwaig demonstrated the similarity of optical and PA spectra of biological molecules, and went on to give an analytical description of PA generation in solids[6], [7].

By selecting excitation wavelengths which are absorbed in particular by glucose, we can utilize the PA response from a sample to predict its glucose concentration. The fundamental absorption bands of glucose lie in the mid infra red, but high absorption by water in this spectral region makes it impractical for measurements on tissues. To allow for greater optical penetration into tissues, we selected two near infra red excitation wavelengths at 905 nm and 1550 nm for the current study[8]. These are close to the 939 nm and 1536 nm overtone bands of glucose[9]. In addition, pulsed laser sources at the two wavelengths were also readily available.

The apparatus for measuring PA signals is described in Section II and the methods utilized for verifying the measurement technique are given in Section III. Sections IV and V discuss the results obtained and conclude the paper.

II. PA Measurement Apparatus

Figure 1 shows a block diagram representation of the experimental apparatus used for making PA measurements. The apparatus used pulsed laser diode (PLD) modules as optical excitation sources. Two PLDs operating at 905 nm (LS9-Series, LaserComponents GmbH) and 1550 nm (LS5-Series, LaserComponents GmbH) were used as optical excitation sources. A set of control circuits was used to regulate the PLD output. The control circuits provided a TTL trigger pulse at a pulse repetition frequency of 100 Hz for PLD operation, and control voltages to adjust the pulse power and pulse width of the PLDs during operation.

The samples are taken in a cuvette (Type G, OptiGlass Limited) and a Lead Zirconate Titanate (PZT) based piezoelectric transducer (SP-5A, Sparkler Ceramics Pvt. Ltd.) is fixed inside the cuvette to receive the PA signal. These

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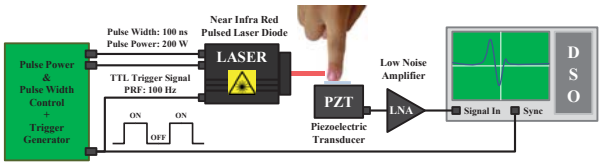


Fig. 1. Block diagram of the apparatus used for PA measurements.

piezoelectric transducers have high sensitivity and are suitable for detecting weak PA pressure signals. A low noise amplifier (LNA)(Model 351A, Analog Modules, Inc.) is used to amplify the output of the PZT elements. The amplified PA signal is acquired using a digital storage oscilloscope (DSO)(Model 54622A, Agilent Technologies). The random noise present in the signal was reduced by averaging the PA signal over 1024 frames using the DSO to get an SNR improvement of 30 dB[10]. The averaged PA signal was saved to a computer for processing and feature extraction.

III. Methods

The pressure, p_{PA}, generated in a sample after exposure of the laser pulse is related to the sample properties and the glucose concentration in the sample, c_g, as follows[4], [11]:

$$p_{PA} = \frac{1}{\pi R^2}\left[\frac{\alpha\beta\upsilon^2}{C_P}\right]E_0 \tag{1a}$$

$$\alpha, \beta, \upsilon \propto c_g \qquad C_P \propto \frac{1}{c_g} \tag{1b}$$

$$p_{PA} \propto \frac{\alpha\beta\upsilon^2}{C_P} \propto c_g \tag{1c}$$

where,
α = optical absorption coefficient of the sample
β = thermal expansion coefficient of the sample
υ = sound velocity in medium
C_P = specific heat of the sample
E_0 = energy incident on the sample
R = radius of PA source

Glucose affects the physical properties of a sample in addition to its optical properties, as given in 1b, resulting in a stronger PA response with increase in the glucose concentration. This was verified through in vitro and in vivo PA measurements, followed by feature extraction and calibration of the measured PA signals.

A. In Vitro Measurements

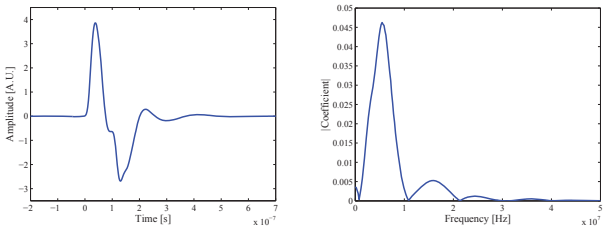
To verify the measurement principle, measurements were initially performed on glucose solutions and whole blood at different concentrations within the physiological glucose concentration range (0-500 mg/dl).

Glucose Solutions: The solutions were made by combining varying amounts of Dextrose Injection I.P. 5% (D5) with distilled water in different proportions. Glucose solutions with concentrations varying from 0 to 500 mg/dl in steps of 50 mg/dl were made and a fixed quantity (4 ml) of each solution was taken in a cuvette for measurement. The cuvette was placed in a sample holder to eliminate variations in the cuvette-laser and cuvette-transducer positions between measurements. The sample was irradiated using the PLD and the resulting PA signal was measured and saved to a computer for processing. PA measurements were taken separately at 905 and 1550 nm excitation wavelengths, with an optical pulse power of 200 W and a pulse width of 100 ns. The PA response was measured and saved to a computer, and the entire procedure was repeated for the next sample.

Whole Blood: Following in vitro measurements on glucose solutions, PA measurements were performed on whole blood drawn from a human volunteer. Blood was drawn from the vein of the volunteer into a blood collection tube (BD Vacutainer, BD). The glucose concentration in blood was measured using a regular glucose meter (OneTouch Ultra 2, LifeScan, Inc.) following which the blood sample was divided into portions and mixed with different proportions of Dextrose Injection I.P. 5% (D5) to get samples at different glucose concentrations. PA measurements were taken on these samples at an excitation wavelength of 905 nm, with a pulse power of 200 W and pulse width of 100 ns. Variations in the measured PA signals are observed with change in the sample glucose concentration.

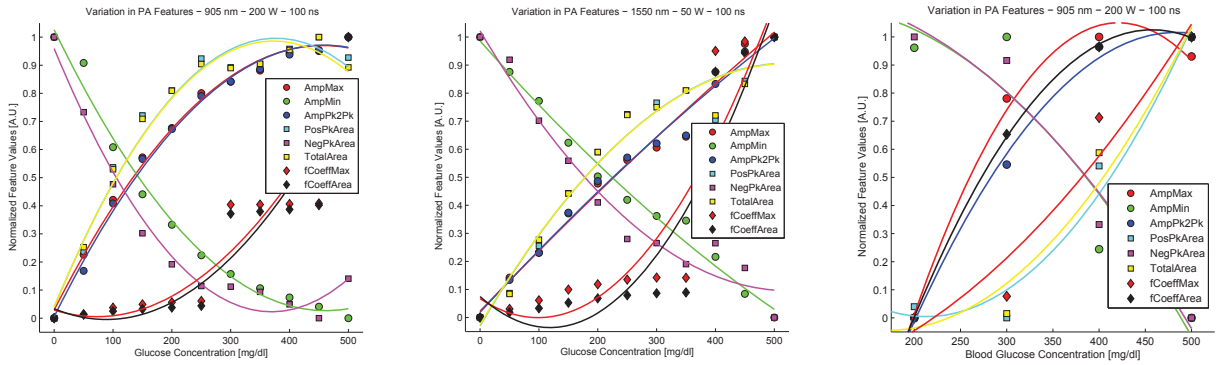
B. PA Signal Features

Figure 2 shows a typical PA pressure waveform measured using the apparatus along with its frequency constituents. To estimate the sample glucose concentration from PA measurements, features representing the PA response must be selected and used for calibration of the system. From 1c, it is known that the PA pressure varies with glucose concentration. Hence, the amplitude of the PA signal and the area under it can be taken as measures of glucose concentration. With the PA waveform being bipolar in nature, as shown in Figure 2a, the maximum, minimum, and peak-to-peak PA amplitude were selected as features of the PA signal. The area under the positive and negative peaks of the PA waveform and total area of the PA signal were also selected as features for calibration. In addition to time-domain features, signal features were also obtained from the amplitude spectrum of the PA signal, as shown in Figure 2b. Here, the maximum coefficient value and total area under the amplitude spectrum were selected as features of the PA signal, as both these quantities represent the strength of the PA signal in the frequency-domain.



(a) Time-Domain (b) Frequency-Domain

Fig. 2. PA Signal acquired using the experimental apparatus.



(a) PA Features on Solutions at 905 nm (b) PA Features on Solutions at 1550 nm (c) PA Features on Whole Blood at 905 nm

Fig. 3. Variations in Features of the PA Signal with Glucose Concentration in Solutions and Whole Blood. List of Features: **1.** Maximum Amplitude (AmpMax) **2.** Minimum Amplitude (AmpMin) **3.** Peak-to-Peak Amplitude (AmpPk2Pk) **4.** Positive Peak Area (PosPkArea) **5.** Negative Peak Area (NegPkArea) **6.** Total Area (TotalArea) **7.** Maximum Frequency Coefficient (fCoeffMax) and **8.** Total Spectrum Area (fCoeffArea)

C. In Vivo Measurements

Following in vitro measurements, PA and blood glucose measurements were performed on human volunteers over the course of glucose/meal tolerance tests. The volunteers initially fasted for 8 hours, following which they consumed a 75 g load of glucose or had a meal to elevate their blood glucose levels. PA and blood glucose measurements were performed simultaneously at intervals of 15-20 minutes over the 2-hour duration of the test. PA measurements were made only at 905 nm keeping the laser power within safety limits[12], [13], while blood glucose was measured using a regular glucose meter (OneTouch Ultra 2, LifeScan, Inc.). The peak-to-peak amplitude of the PA signal was measured and correlated with reference glucose measurements. The system was calibrated using regression based methods, with the PA amplitude being used to obtain the glucose concentration value. The accuracy of the proposed method was tested by comparing glucose concentration estimates to reference glucose measurements using a Clarke Error Grid (CEG)[14]. This experimental study was approved by the Institutional Ethical Committee and informed consent was taken from all study participants.

IV. Results and Discussion

The measured PA waveforms are bipolar in nature, as shown in Figure 2, and have a width of nearly 225 ns. This is in good agreement with theoretical estimates (2.35 τ_{pulse}, $\tau_{pulse} = 100$ ns) and gives confidence in the PA generation scheme[11]. PA measurements taken on aqueous solutions and whole blood at different glucose concentrations were saved to a computer and processed to extract features.

Figures 3a and 3b show the variation in features of PA measurements made on glucose solutions at 905 nm and 1550 nm. The amplitude of the PA signal was found to increase gradually with an increase in the glucose concentration. This is consistent with the theoretical basis given in 1c earlier. The maximum PA amplitude rose and the minimum PA amplitude fell with an increase in glucose concentration. The peak-to-peak amplitude thus increased with the glucose concentration of the solution. The change in amplitude features is not linear and tends to saturate at high glucose concentrations. In addition to signal amplitude, it was noted that area under the PA signal waveform varied significantly with changes in the sample glucose concentration. The area under the positive and negative peaks of the PA signal was found to increase with glucose concentration of the solution. The total area under the PA waveform was also found to increase with rise in sample glucose concentration. Furthermore, this increase was also observed to be non-linear in nature, saturating at high glucose concentrations.

The amplitude spectrum of the PA signal was obtained and the value of frequency domain coefficients was observed to increase with the sample glucose concentration. The maximum coefficient value was found to be at 5 MHz, the center frequency of the PZT transducer, and it increased the glucose concentration. In addition, the area under the entire frequency spectrum of the PA signal also increased with rise in the glucose concentration. Unlike time domain features such as amplitude and area, which varied gradually with glucose concentration, the variation in frequency domain features was not smooth in nature. However, these might still serve as valuable features for calibration of PA measurements. While feature values obtained at 905 nm were found to saturate at high glucose concentrations, the variation in PA signal features at 1550 nm was found to be near-linear.

Similar variations were observed in features obtained from PA measurements taken on whole blood samples at different glucose concentrations. The maximum, minimum and peak-to-peak PA amplitude increased with an increase in the glucose concentration, saturating at higher sample concentrations. The area under the peaks of the PA signal also increased with rise in the sample concentration. The area under the frequency spectrum also followed a similar pattern, increasing with the glucose concentration and saturating at higher concentration values. Figure 3c shows the variations in the PA signal features obtained from whole blood at different glucose concentrations.

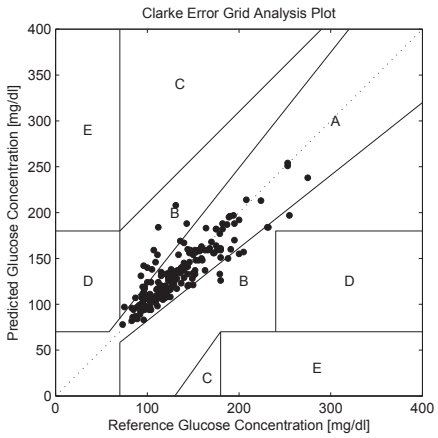


Fig. 4. A Clarke Error Grid Analysis of In Vivo PA Measurements

Finally, in vivo PA measurements made on a group of 30 human subjects were calibrated to reference glucose measurements made simultaneously. A quadratic fit, as observed in amplitude measurements taken on glucose solutions, was used for individual calibration of PA measurements. Figure 4 shows a CEG plot of estimated glucose values and reference glucose measurements from all 30 subjects. The CEG divides the correlation plot of the glucose measurement pairs into 5 zones from A - E depending upon the suitability of use of predicted measurements for treatment. While measurement pairs falling in zones A and B are acceptable, pairs falling in zones C - E are dangerous for treatment. Of the 196 measurement pairs obtained, 171(87.24%) pairs were found to lie in zone A with the remaining 25 (12.76%) pairs lying in zone B of the CEG. There were no measurement pairs in zones C-E of the grid. All measurement pairs fell within acceptable zones A and B of the error grid, with the estimated glucose measurements having a mean absolute difference (MAD) of 12.57 $\pm$ 13.90 mg/dl and a mean absolute relative difference of 9.61% $\pm$ 10.55%. This outcome is an improvement over results reported in literature using PAS and other non-invasive measurement methods as shown in Table I [15], [16], [17], [18], [19].

V. Conclusions and Future Directions

The use of NIR PAS for continuous non-invasive glucose monitoring was demonstrated on glucose solutions and whole blood. An apparatus for making PA measurements was constructed, and time and frequency domain features of PA measurements at different glucose concentrations were examined for calibration. The features were found to increase smoothly with the glucose concentration and saturated at high concentration values. In vivo PA measurements were performed on individuals and a quadratic fit was used for calibration. The calibration accuracy obtained was superior to earlier studies using PAS or other methods. This can be improved further by using multiple sensors, different pulse excitation modes, and multi-variate calibration techniques.

TABLE I
Performance Comparison of Non-invasive Glucose Measurement Methods

	Zone A	Zones A & B	Zones C - E	MARD (%)
Proposed (PAS)	87.24	100.00	0.00	9.61
Raman Spectroscopy[15]	86.66	-	-	-
Sentris (OCT)[16]	83.00	99.00	1.00	11.50
Aprise (PAS)[17]	66.50	94.60	5.40	-
GlucoTrack[18]	60.00	96.00	4.00	22.40
Solianis[19]	56.00	93.00	7.00	-

References

[1] International Diabetes Federation, *IDF Diabetes Atlas*, 6th ed. International Diabetes Federation, 2014.

[2] The Diabetes Control and Complications Trial Research Group, "The Effect of Intensive Treatment of Diabetes on the Development and Progression of Long-Term Complications in Insulin-Dependent Diabetes Mellitus," *The New England Journal of Medicine*, vol. 329, no. 14, pp. 977–986, 1993.

[3] D. D. Cunningham and J. A. Stenken, *In Vivo Glucose Sensing*. John Wiley & Sons, Inc., 2010.

[4] V. V. Tuchin, *Handbook of Optical Sensing of Glucose in Biological Fluids and Tissues*, V. V. Tuchin, Ed. CRC Press, 2008.

[5] A. G. Bell, "The Production of Sound by Radiant Energy," *Science*, vol. 2, no. 48, pp. 242–253, May 1881.

[6] A. Rosencwaig, "Photoacoustic Spectroscopy of Biological Materials," *Science*, vol. 181, no. 4100, pp. 657–658, Aug. 1973.

[7] A. Rosencwaig and A. Gersho, "Photoacoustic Effect with Solids: A Theoretical Treatment," *Science*, vol. 190, no. 4214, pp. 556–557, 1975.

[8] A. N. Bashkatov, E. A. Genina, V. I. Kochubey, and V. V. Tuchin, "Optical properties of human skin, subcutaneous and mucous tissues in the wavelength range from 400 to 2000 nm," *Journal of Physics D: Applied Physics*, vol. 38, no. 15, pp. 2543–2555, Aug. 2005.

[9] O. S. Khalil, "Spectroscopic and Clinical Aspects of Noninvasive Glucose Measurements," *Clinical Chemistry*, vol. 45, no. 2, pp. 165–177, Feb. 1999.

[10] R. G. Lyons, "Signal Averaging," in *Understanding Digital Signal Processing*, 2nd ed. Pearson Education, 2009, ch. 11, pp. 429–456.

[11] A. C. Tam, "Applications of Photoacoustic Sensing Techniques," *Reviews of Modern Physics*, vol. 58, no. 2, pp. 381–431, 1986.

[12] International Electrotechnical Commission, *International Standard - IEC 60825-1 - Safety of Laser Products*. IEC, 2001.

[13] American National Standards Institute, *ANSI Standard for Safe Use of Lasers in Health Care Facilities*. Laser Institute of America, 2005.

[14] W. L. Clarke, D. J. Cox, L. A. Gonder Frederick, W. Carter, and S. L. Pohl, "Evaluating Clinical Accuracy of Systems for Self-Monitoring of Blood Glucose," *Diabetes Care*, vol. 10, no. 5, pp. 622–628, 1987.

[15] N. C. Dingari, I. Barman, J. W. Kang, C.-R. Kong, R. R. Dasari, and M. S. Feld, "Wavelength selection-based nonlinear calibration for transcutaneous blood glucose sensing using Raman spectroscopy," *Journal of Biomedical Optics*, vol. 16, no. 8, pp. 087 009–1–087 009–10, Aug. 2011.

[16] R. A. Gabbay and S. Sivarajah, "Optical Coherence Tomography-Based Continuous Noninvasive Glucose Monitoring in Patients with Diabetes," *Diabetes Technology & Therapeutics*, vol. 10, no. 3, pp. 188–193, 2008.

[17] R. Weiss, Y. Yegorchikov, A. Shusterman, and I. Raz, "Noninvasive Continuous Glucose Monitoring Using Photoacoustic TechnologyResults from the First 62 Subjects," *Diabetes Technology & Therapeutics*, vol. 9, no. 1, pp. 68–74, Feb. 2007.

[18] I. Harman Boehm, A. Gal, A. M. Raykhman, E. Naidis, and Y. Mayzel, "Noninvasive Glucose Monitoring: Increasing Accuracy by Combination of Multi-Technology and Multi-Sensors," *Journal of Diabetes Science and Technology*, vol. 4, no. 3, pp. 583–595, 2010.

[19] A. Caduff, F. Dewarrat, M. Talary, G. Stalder, L. Heinemann, and Y. Feldman, "Non-invasive glucose monitoring in patients with diabetes: A novel system based on impedance spectroscopy," *Biosensors and Bioelectronics*, vol. 22, no. 5, pp. 598–604, Dec. 2006.