Multivariate Regression and Classification Models for Estimation of Blood Glucose Levels Using a New Non-invasive Optical Measurement Technique Named "*Pulse-Glucometry*"

Yasuhiro Yamakoshi[1], Mitsuhiro Ogawa[*,2,3] and Toshiyo Tamura[1]

[1]*Graduate School of Engineering, Chiba University, Chiba, Japan*

[2]*Graduate School of Natural Science and Technology, Kanazawa University, Kanazawa, Japan*

[3]*Yu.sys Corporation, Kanazawa, Japan*

Abstract: An optical method recently proposed for non-invasive *in vivo* blood glucose concentration (BGL) measurement, named "*pulse glucometry*", was implemented with three multivariate regression methods, Principal Component Regression (PCR), Partial Least Squares Regression (PLS) and Support Vector Machines Regression (SVMsR), as well as with a classification method, Support Vector Machines Classification (SVMsC), for carrying out calibration. A very fast spectrophotometer provided instantaneously and simultaneously the total transmitted radiation spectrum (I^{λ}) and the cardiac-related pulsatile component (ΔI^{λ}) superimposed on I^{λ} in human fingertips over a wavelength range from 900 to 1700 nm with resolution of 8 nm in 100 Hz sampling. From a family of I^{λ}s including information relating to various BGL values, differential optical densities (ΔOD^{λ}s (=$\log(1+\Delta I^{\lambda}/I^{\lambda})$) were calculated and normalized by the ΔOD^{λ} values at 1100 nm. Finally, the 2nd derivatives of the normalized ΔOD^{λ}s served as regressors. Subsequently, calibration models from regressors and regressands (the corresponding measured BGL or classified BGL values) were constructed with PCR, PLS, SVMsR and SVMsC. Each regression model showed a relatively good result by evaluating a 5-fold cross validation using total 100 data-sets: Clarke error grid analysis indicated a good correlation in each model compared with the measured BGL values, and the SVMsR calibration provided the best plot distributions. Good regression models were obtained by these three methods. This study suggests that *"pulse glucometry"* can produce clinically acceptable results when implemented with regression and classification type calibrations, and, through rapid BGL assessment, promises to offer a more practical, easier and more convenient way for diabetes screening and health care in normal subjects than is possible with existing methods.

INTRODUCTION

Measurement of blood glucose concentration (BGL) has long been considered as an essential need for self-care in diabetes and for screening in pre-diabetes, as well as for health care in normal subjects. For diabetes management in particular, frequent measurements of BGL is inevitable [1], and thus many kinds of portable devices for self-monitoring of blood glucose (SMBG) have been commercialized worldwide. However, many of the current SMBG devices are based on the user puncturing their skin with a small needle or lancet and squeezing the surrounding tissue to withdraw a blood sample, and this is a severe limitation; frequent monitoring is necessary and the repeated procedure of skin puncturing becomes painful and troublesome and, furthermore, can cause an infection.

Although a non-puncturing type of BGL device, the GlucoWatch Biographer, which is based on reverse-iontophoresis to draw glucose molecules *via* inner skin [2], has been approved by the FDA, its measurement procedure can still cause skin irritation after repeated applications [3]. Truly non-invasive BGL measurement has therefore been most desired and has been explored worldwide particularly using optical methods such as surface plasmon resonance [4], photo-acoustic technique [5], optical coherence tomography [6], diffuse reflectance spectrometry [7], and so on [8, 9]. However, despite considerable efforts in these developments for more than four decades reliable and clinically acceptable measurement methods have not yet emerged. The major obstacle to success with *in vivo* optical BGL measurement is the very small *in vivo* optical signature of glucose. Furthermore, certain optical characteristic features of biological tissues create significant interference, for example: other absorbing species; multiple scatter in skin, muscle, and bone; and the strong absorption bands of water [10-12]. Consequently, a method to observe only blood has long been sought.

We have recently focused attention on *in vivo* spectrophotometric measurement in living tissues, with analysis to obtain parameters related to blood including blood glucose, and have subsequently proposed a novel technique named "*pulse glucometry*". This is a non-invasive approach for BGL monitoring based on very high speed near infrared spectroscopy, having a sampling rate significantly faster than the cardiac cycle [13-15].

In the main, methods of *in vivo* and *in vitro* optical spectroscopic analysis, including "*pulse glucometry*", have

*Address correspondence to this author at the Graduate School of Natural Science and Technology, Kanazawa University, Kakuma, Kanazawa 920-1192, Japan; Tel: +81-(0)76-234-6467; Fax: +81-(0)76-234-4737;
E-mail: mtsogawa@kenroku.kanazawa-u.ac.jp

1874-3285/09

been implemented with standard chemometric procedures. These have utilized multivariate calibration models constructed either by simple multiple linear regression (MLR) or multiple regression based schemes, such as partial least squares regression (PLS) and principal component regression (PCR) [16]. MLR, PLS and PCR are generally used for pure linear calibration models or linearly-transformable (nonlinear) calibration. Just recently, through developments in the field of multivariate statistical analysis, a kernel-based method has appeared with the emergence of the support vector machines (SVMs) including the kernel trick [17, 18]. This kernel trick is one of several useful methods for converting a linear classifier algorithm into a nonlinear one. The SVMs method is also currently regarded as one of the strongest methods of supervised learning applied to classification and regression.

This paper deals, firstly, with an attempt to apply the nonlinear method, SVMs, to "*pulse glucometry*" to obtain a multivariate calibration regression model. This is then compared with our prior use of the PLS method reported previously [13, 15] and the conventional PCR. Secondly, the classification of BGL levels using SVMs is attempted by application to "*pulse glucometry*."

MATERIALS AND METHODS

A. Basic Principle of Pulse Glucometry

Details of the previously proposed method of "*pulse glucometry*" are reported elsewhere [13, 15]. Briefly, we assume a simplified optical model of biological tissue, comprising of three compartments, as shown in Fig. (**1**): arterial; venous; bloodless tissue. The essence of this new method is to utilize a blood volume change in a tissue segment under optical interrogation, such as a finger, and then, with a subtraction process, to remove the influences of basal interfering elements such as bone, skin, muscle, water and non-pulsatile blood [13]. We perform very rapid spectrophotometry in a finger such that the changes in optical absorption and scattering properties of the tissue produced by cardiac related blood volume pulses can be determined accurately.

If we use incident radiation at various wavelengths (λ) with intensity (I^{λ}_0) to interrogate the tissue, the transmitted radiation intensity, I^{λ}, can be obtained. This transmitted intensity I^{λ} contains very small pulsatile changes in intensity (ΔI^{λ}). These observed pulsatile optical changes are due to cardiac related arterial volume changes. The overall optical signal obtained is the photoplethysmograpm (PPG). If the transmitted intensities at time t_i and t_{i+1} ($I^{\lambda}(t_i)$, $I^{\lambda}(t_{i+1})$) are detected during the same cardiac cycle, we can obtain the optical densities at t_i ($OD^{\lambda}(t_i)=\log(I^{\lambda}o/I^{\lambda}(t_i))$ and t_{i+1} ($OD^{\lambda}(t_{i+1})=\log(Io^{\lambda}/I^{\lambda}(t_{i+1}))$. Hence we can calculate the difference between $OD^{\lambda}(t_i)$ and $OD^{\lambda}(t_{i+1})$, ($\Delta OD^{\lambda}(t_i)=\log(I^{\lambda}(t_{i+1})/I^{\lambda}(t_i))=\log(1+\Delta I^{\lambda}(t_i)/I^{\lambda}(t_i))$, where $I^{\lambda}(t_{i+1})=I^{\lambda}(t_i)+\Delta I^{\lambda}(t_i)$, as shown in the upper-right part of Fig. (**2**). This has the effect of removing the interfering elements to acquire only the change in intensity due to the pulsating blood volume.

When we choose $\Delta I^{\lambda}(t_i)$ as a pulsatile component ΔI^{λ} for different wavelengths, λ_1, λ_2, --- λ_n, ΔOD^{λ} can be calculated as a function of λ. If such spectra are derived at different BGL levels (BGL_i), we can obtain a family of spectra, as schematically shown in the right part of Fig. (**2**), which may then be used for multivariate analyses. Calibration models constructed by several multivariate analyses are applied for spectrophotometric BGL measurements: Measured spectra of ΔOD^{λ} *vs* λ for the unknown BGL derived from *in vivo* measurements (see the left lower part of Fig. (**2**)) on a finger can be compared with the derived models and predicted BGL values thereby calculated.

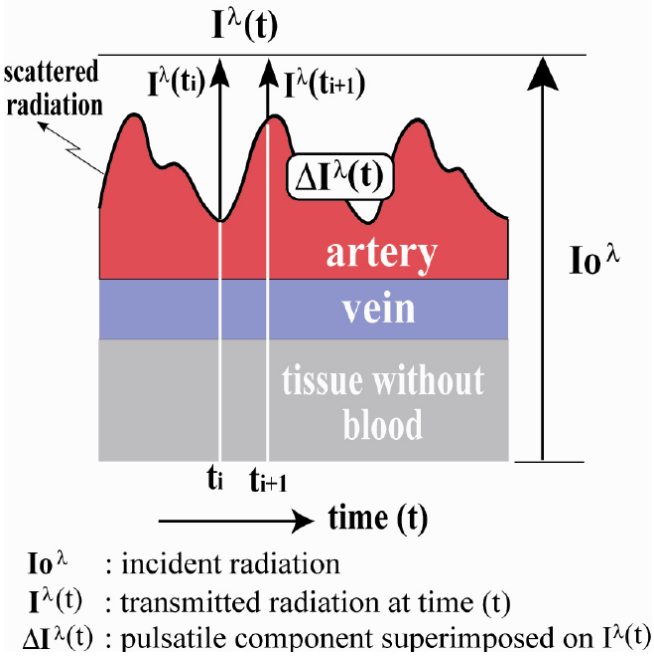

Fig. (1). Simplified optical model of living tissue.

B. Experimental Setup

For the present study the measurement system consisted of a light source (halogen lamp: maximum power 150 W), an optical fiber of 10-mm diameter for the incident radiation and a single low-OH optical fiber of 1.2-mm diameter for collecting the transmitted radiation, a spectrometer (polychromator, M25-TP; Bunkoh-Keiki Co. Ltd., Japan), a linear, liquid nitrogen cooled (−50 to −100 °C), InGaAs photodiode-array (multi-photodetector, OMA V: 512-1.7(LN); Princeton Instruments Co., USA), and a conventional personal computer with an appropriate interface. Using this system, the transmitted radiation intensity (I^{λ}) over the wavelength (λ) range from 900 to 1700 nm was measured with a resolution of better than 8 nm and with 16-bit digitization. The maximum spectrum sampling speed achievable with this system is 1800 spectra/s, but in the experiment described here we adopted a speed of 100 spectra/s, which was sufficient to obtain accurately the cardiac-related transmitted intensity change superimposed on the intensity (I^{λ}) (ΔI^{λ}: i.e., photoplethysmographic pulsation) over this wavelength range.

C. Subjects and Experimental Procedures

In order to carry out *in vivo* measurements, oral glucose tolerance tests (OGTT) were carried out to create varying BGL levels in 10 healthy volunteers (22 to 59 years old; 8 males and 2 females). Prior to the experiments, the local ethics committee approved the experimental procedures and informed consent was obtained from each subject. The

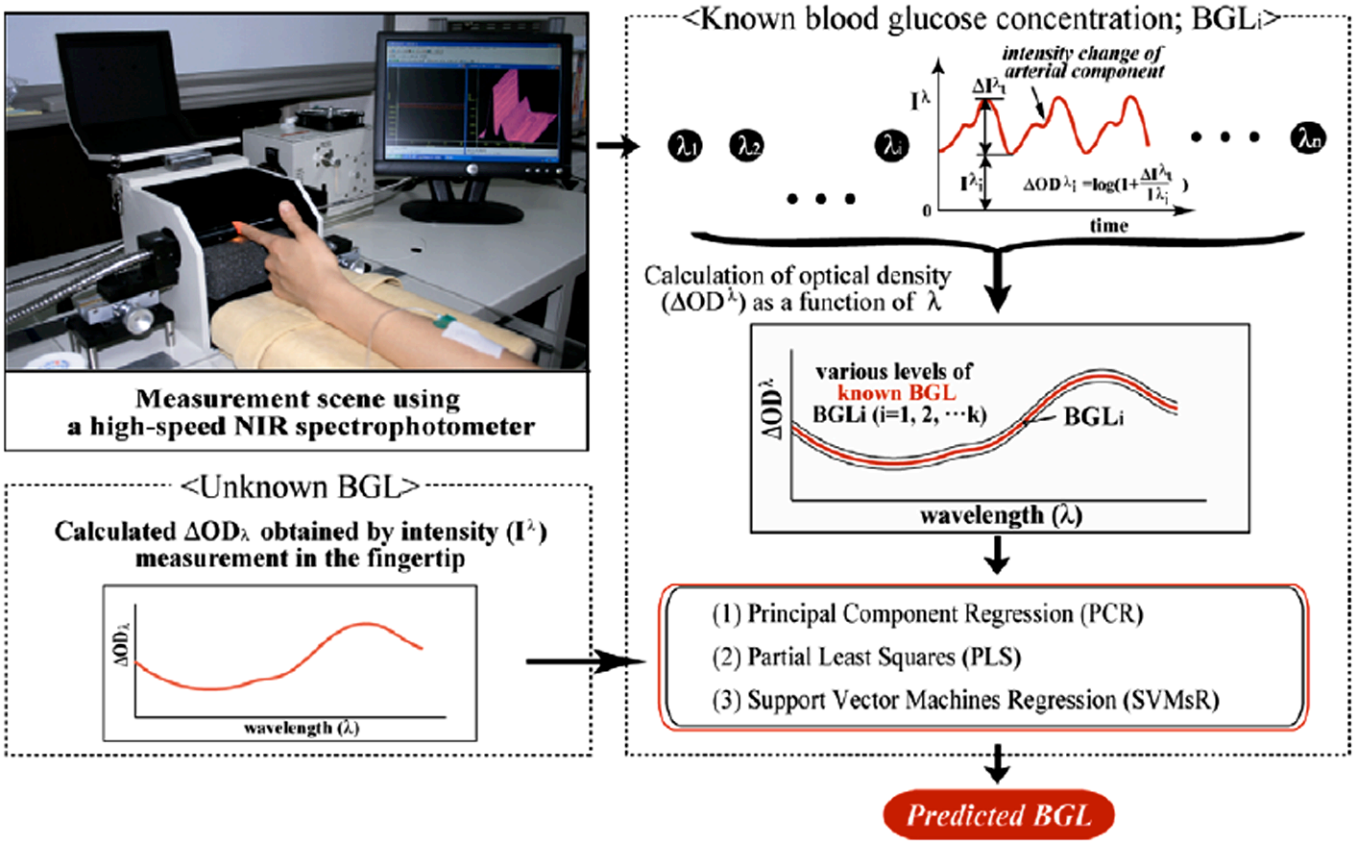

Fig. (2). Measurement setup (left upper part), procedures for constructing multivariate regression calibration models (right part), and prediction of BGL using optical measurement with unknown BGL (left lower part) based on "*pulse glucometry*".

subjects were asked to abstain from food and alcohol from 9 p.m. on the previous day until the end of the experiment on the next morning. The experiment began at 9 a.m., in a room maintained at a temperature of 25 ˚C. During the experiment, the subjects were also requested to sit quietly in a chair to undergo the test.

In this study, radiation intensity measurements were made in the left index fingertip at 10 min intervals before and after oral uptake of glucose solution (75 g/225 ml, Trelan-G75, Shimizu Seiyaku, Co. Ltd., Japan) for a study period of 100 or 120 min. During the optical measurement, the subject's left hand was held horizontally at heart level. Immediately after each optical measurement blood samples (about 3 ml) were collected from the cephalic vein of the left forearm and analyzed by an automatic blood analyzer (DRI-CHEM 7000, Fujifilm Medical Co. Ltd., Japan) to obtain the reference (measured) BGL values (regressand for calibration models). The BGL values obtained in the present study ranged from 100.7 to 246.3 mg/dl (5.59-13.7 mmol/l).

For the optical intensity measurements, the fingertip was placed carefully in an adjustable space between the ends of the transmitting and detecting fibers so as to ensure gentle contact with the skin, as shown in the left upper part of Fig. (**2**). The intensity measurements were made for about 20 to 30 s (corresponding 20 to 30 cardiac pulses), and stable and almost constant intensity signals (I^{λ} and ΔI^{λ}) for 3 to 5 successive cardiac beats were used as analytical data.

D. Signal Pre-Processing

To improve further the signal-to-noise ratio (S/N) of the pulsatile intensity change (ΔI^{λ}) superimposed on the I^{λ} signal over the wavelength range from 900 to 1700 nm, the Savitzky-Golay filtering method, which is used generally for signal pre-processing, was applied for both wavelength (λ) and time (t). Parameters of the Savitzky-Golay filterers were empirically selected; the polynomial order was 2 and the frame size was 3 for the wavelength direction and the polynomial order 2 and the frame size 15 for the time-sequential direction. With these parameters an S/N of more than 25 dB was obtained.

The mean ΔOD^{λ} spectrum calculated from the 3 to 5 successive cardiac beats was used for multivariate analyses, and a family of these spectra for different BGL levels (known BGL values) served as the data-set. The ΔOD^{λ} spectra thus obtained were then normalized ($\Delta OD^{\lambda}_{norm}$) using the ΔOD^{λ} value at 1100 nm as 1.0 together with its minimum value as 0 in each spectrum [13]. Finally, the second order difference spectra ($\delta^2\Delta OD^{\lambda}_{norm}/\delta\lambda^2$; $\delta\lambda$=8 nm) were derived to reject baseline fluctuations of spectra. This second order difference spectral method is usually applied in the field of chemometrics, in order to remove low frequency sources of variations that are not related to solution concerned [19]. In the calculation of the spectra $\delta\lambda$ was equivalent to the wavelength resolution of the measurement system (8 nm).

The processing of these data was carried out using MATLAB^{TM} version 7.x software and a conventional

personal computer (Dimension 9100 with a CPU Pentium D 830 and 2GB memory, DELL Inc., Japan).

E. Multivariate Calibration Regression and Classification Models

Using the data-sets of the difference spectra (regressor) and the corresponding known (measured) BGL values (regressand), two linear regression methods, PCR and PLS, and a non-linear regression method, SVMsR, were applied to create multivariate calibration models. Table **1** shows features of each multivariate regression method. 5-fold cross-validation was introduced to evaluate the calibration models. In order to obtain the resultant calibration model, parameters on each regression were searched repeatedly.

Then, the classification model constructed by SVMsC was attempted for the data-sets described above. Firstly, the known BGL levels were classified into three classes on the basis of criteria used for diabetes screening [20]. In this study, optical data $\delta^2 \Delta OD^{\lambda}_{norm}/\delta\lambda^2$ were classified by SVMsC in the case of fasting blood glucose (FBG) as "Normal class (BGL < 110 mg/dl)", "Impaired Fasting Glycaemia (IFG) class (110 ≤ BGL ≤ 126)" or "Diabetes Mellitus (DM) class (BGL > 126)". On the other hand, data were classified in the case of 2-hour postprandial blood glucose (PBG) as "Normal class (BGL < 140)", "Impaired Glucose Tolerance (IGT) class (140 ≤ BGL ≤ 200)" or "Diabetes Mellitus class (BGL > 200)". Then, 5-fold cross-validation was also applied to evaluate the calibration models. Parameters on each classification were also searched repeatedly.

To implement these calibration procedures, the software "R" version 2.3.1 and the kernlab module version 08-2 for the software "R" [21-23] were used on a conventional personal computer.

RESULTS

Obtained optical data containing noise and/or artifact were rejected manually and then 100 data-sets were selected. Fig. (**3**) shows an example of the differential optical density ((a): ΔOD^{λ}), normalized ΔOD^{λ} ((b): $\Delta OD^{\lambda}_{norm}$), and the 2nd derivatives of $\Delta OD^{\lambda}_{norm}$ spectrum ((c): $\delta^2 \Delta OD^{\lambda}_{norm}/\delta\lambda^2$) obtained in the fingertip of a healthy female subject. Using a family of these spectra determined in all subjects studied, multivariate calibration models were constructed and evaluated. Resultant parameters for each method were obtained as follows. For PCR, the 1st to 20th principal components were introduced into the regression model. For PLS, the number of latent variable was reduced to 15 against a data dimension (dimension of regressor) of 100. For PCR the parameter determination was done by evaluating models for the first n principle components with the parameter n from 1 to 100. Then, the best parameter n = 20 was selected by counting the number of plots falling within Region A of Clarke error grid. In the parameter determination of PLS, the models with latency variables from 1 to 100 were evaluated, and the variable of 15 was selected in the same way. For SVMsR, the ANOVA RBF (radial basis function) kernel with a degree of 0.01 and sigma of 1 was used in training and ε in Vapnik's insensitive-loss function was 0.123. For SVMsC, the ANOVA RBF kernel with degree of 0.01 and sigma of 1 was used in training and ε in Vapnik's insensitive-loss function was 0.15. Table **1** summarizes these multivariate analysis methods and the obtained parameters.

Fig. (**4**) includes scatter plots showing a comparison of the estimated and measured BGL values by Clarke error grid analysis. As shown in this diagram, almost all data points were within clinically acceptable regions [24], that is, the regions A and B in each calibration. Among them, the SVMsR calibration model provided the best plot distributions (Region A: 84/100, Region B: 16/100 and Region C-E: 0/100) compared with the PCR model (Region A: 78/100, Region B: 22/100 and Region C-E: 0/100) and the PLS model (Region A: 80/100, Region B: 19/100 and Region C-E: 1/100). Therefore, it might reasonably be suggested that SVMsR can be used for constructing a multivariate calibration model as part of the procedure of implementing "*pulse glucometry*". Table **2** shows the result using the nonlinear classification model (SVMsC). In the case of FBG (Table **2a**), the accuracy rate (and the probability of misclassification) was 2/4 (2/4) for Normal, 3/11 (8/11) for IFG, and 80/85 (5/85) for DM. In the case of

Table 1 Summary of Multivariate Regression and Classification Methods and Parameters Used in the Present Study

Methods	Aspects on multivariate analysis				
	Regression/Classification	**Linear/Non Linear**	**Calculation Time**	**Parameters**	**Values**
PCR	Regression	Linear	Short	Applied Principal Components	1st to 20th
PLS	Regression	Linear	Short	Number of Latency Variable	15
SVMsR	Regression	Non Linear	Intermediate	Kernel	ANOVA RBF
				Kernel Parameter Degree	1
				Kernel Parameter Sigma	0.01
				ε in Vapnik's Insensitive Loss Function	0.123
SVMsC	Classification	Non Linear	Intermediate	Kernel	ANOVA RBF
				Kernel Parameter Degree	1
				Kernel Parameter Sigma	0.35
				ε in Vapnik's Insensitive Loss Function	0.05

PBG (Table **2b**), these were 13/23 (10/23) for Normal, 53/69 (16/69) for IGT and, 4/8 (4/8) for DM. Additionally, the mean square errors of each calibration model were as follows: PCR; 28.15 [mg/dl], PLS; 31.54 [mg/dl] and SVMsR; 28.27 [mg/dl].

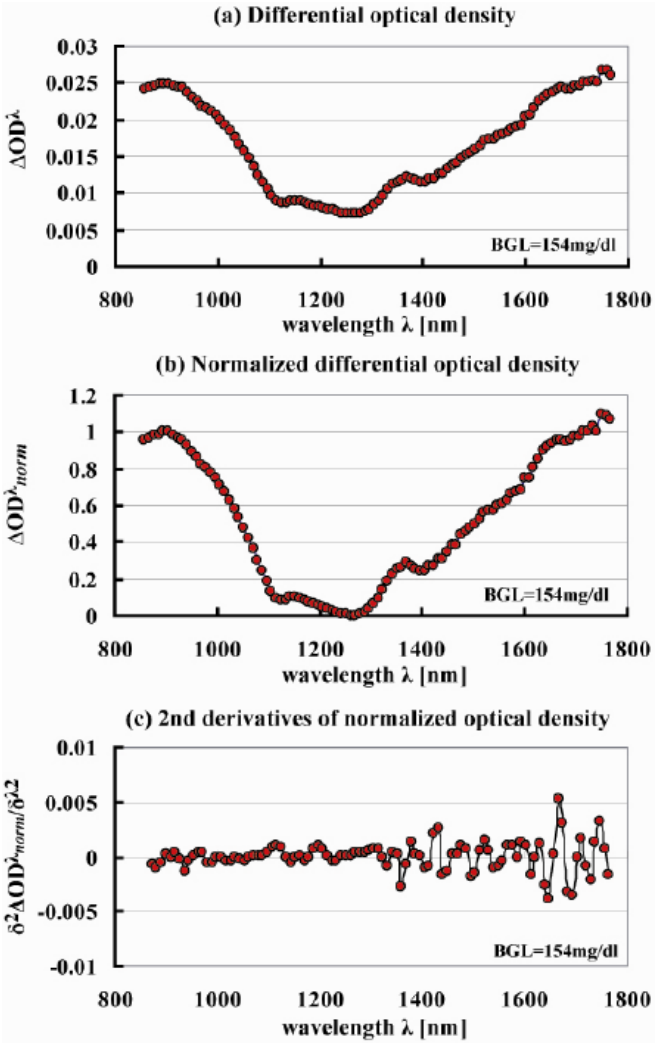

Fig. (3). Optical data obtained over the wavelength range from 900 nm to 1700 nm by the very-fast NIR spectrophotometric system obtained in the fingertip of a healthy female subject: (**a**) differential optical density (ΔOD^{λ}), (**b**) normalized differential optical density ($\Delta OD^{\lambda}_{norm}$), and (**c**) the 2nd derivatives of normalized optical density spectrum ($\delta^2 \Delta OD^{\lambda}_{norm}/\delta\lambda^2$).

DISCUSSION

In the present study, in order to predict blood glucose (BGL) levels using "*pulse glucometry*", we introduced two linear (PCR and PLS) and one nonlinear (SVMsR) calibration methods. Our results indicated that there were no significant differences in the performance of these three calibration methods, as shown in the Clarke error plots (see Fig. **4**), although the SVMsR model appears to produce a better regression than the others. However, it is not to say that the regression by SVMsR is superior to the other methods, since each has strengths and weaknesses. It is the case that the PLS appraoch is one of the most widely used regression techniques in the chemometrics field [16]. From a

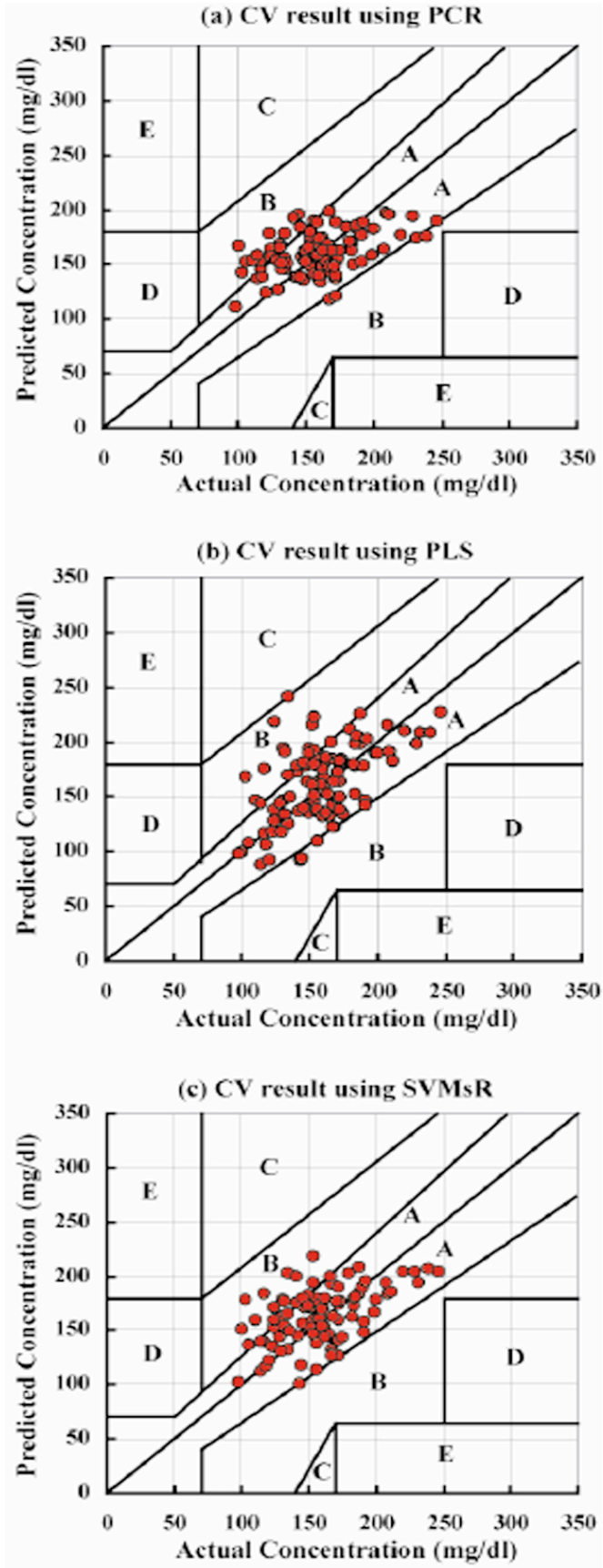

Fig. (4). Scatter diagrams evaluated by 5-fold cross validation showing comparison of measured and predicted BGL values by Clarke error grid analysis: (**a**) PCR, (**b**) PLS, and (**c**) SVMsR.

Table 2. Results of Classification Type Calibration by SVMsC

(a) In the Case of Fasting Blood Glucose (FBG)				
		Measured BGL Value (mg/dl)		
		～110(Normal)	110～126 (Impaired Fasting Glycaemia)	126～(Diabetes Mellitus)
Classified BGL (mg/dl)	～110(Normal)	2	0	0
	110～126 (Impaired Fasting Glycaemia)	0	3	5
	126～(Diabetes Mellitus)	2	8	80
(b) In the Case of 2-Hour Postprandial Blood Glucose (PBG)				
		Measured BGL value (mg/dl)		
		～140(Normal)	140～200 (Impaired Glucose Tolerance)	200～(Diabetes Mellitus)
Classified BGL (mg/dl)	～140(Normal)	13	12	1
	140～200 (Impaired Glucose Tolerance)	10	53	3
	200～(Diabetes Mellitus)	0	4	4

theoretical viewpoint, it can be said that the conventional PLS is only applicable to a linear model. If nonlinear elements are involved in a system then the PLS based calibration cannot avoid erroneous factors originating from the nonlinearity. At the same time, also from the theoretical viewpoint, the SVMsR can give solutions for nonlinear problems [17, 18]. In fact, there have been some studies attempting to compare the performance of the SVMsR with the PLS for spectral regression applications in the chemometrics field and these have, in fact, pointed out the superiority of the SVMsR over the PLS [25, 26]. In their work on powdered milk analysis using near infra-red spectrophotometry, Borin *et al*. [26] clearly showed that starch, whey protein and sucrose concentrations could be accurately predicted by the SVMsR method. Their model gave clearly superior results for calibration sets than those derived by the PLS. In contrast, taking the present consistent results of majority (around 80%) of measurements within region A of the Clarke error plot into considering, it is demonstrated that the optical technique *"pulse glucometry"* provides clinically promising BGL measurements.

It is relevant to consider the well-known method of 'pulse oximetry' which is used for *in vivo* measurement of blood oxygenation. This method also uses a photoplethysmographic technique to measure the optical intensity corresponding to arterial blood volume pulsations, in this case at two wavelengths [27]. A simple ratio calculated from the transmitted or reflected optical intensities and their pulsatile components at two wavelengths is used to determine the relative proportions of the two most significant chromophores, oxy- and deoxyhemoglobin. In this method, the processing to calculate the ratio can be considered as a linearization function. Similarly, in our "*pulse glucometry*" approach, the processing of ΔOD^{λ} normalized to $\Delta OD^{\lambda}_{norm}$ can also achieve such linearization. It is, in general, difficult to determine the most appropriate linearizing method and, in fact, we introduce a nonlinear problem solver, SVMsR, in this study.

It is perhaps unrealistic at present to expect that non-invasive BGL measurement could be a more accurate alternative to invasive methods. From the standpoint of practical use, the classification type calibration (qualitative in nature), attempted in the present study, might be valuable to consider in favour of the regression type calibration (providing quantitative BGL levels). At present, diabetes sufferers seem compelled to rely on frequent and long-term finger-pricking for the control of BGL levels using an SMBG device. The classification approach using a non-invasive method like "*pulse glucometry*" could therefore be a viable alternative available for those with established diabetes, for pre-diabetes screening, as well as for health care in normal subjects, offering an easier and more convenient way of achieving rapid self BGL monitoring during normal daily life. It is also notable that, in the field of urine glucose test strips, rough quantification of urine glucose levels has been successful and is widely utilized. Needless to say, regression type calibration is necessary for accurate management of diabetes, such as self-adjustment of insulin dosage. The present study therefore deals with both possible approaches of the quantitative and qualitative BGL assessment using multivariate calibration and classification models. The present classification result seems to be good for groups with a large number of subjects; DM group in the case of FBG and IGT group in the case of PBG. Since the numbers of data sets in this study were not sufficient for the groups of Normal and IFG in FBG and for those of Normal and DM in PBG, it is at present difficult to lead make firm conclusions on the adequacy of classification. Further investigations will be needed to obtain an appropriate number of data sets so as to allow a statistical analysis.

Since the present study is a first stage to validate the applicability of multivariate calibration and classification methods to the newly proposed "*pulse glucometry*", further work is still required to obtain comprehensive optical data in a large group of diabetic subjects and also to assess the full potential of this method for clinical and research purposes. Currently, OGTT is recommended for evaluation of patients in whom diabetes is still strongly suspected but who have normal FPG (fasting plasma glucose) or IFG (impaired fasting glucose) [28]. Therefore, diabetes mellitus patients were not selected as subjects in this preliminary phase study. In the near future, however, we will plan a safe experimental procedure for diabetic patients. In addition, the present experimental setup is laboratory-based and thus further development is needed to

design a small portable device using several LEDs as light sources which will be suitable for use by patients and normal subjects to achieve self-monitoring of BGL.

CONCLUSION

A recently proposed non-invasive *in vivo* BGL optical measurement technique named "*pulse glucometry*" was combined with three types of regression analyses (PCR, PLS and SVMsR) and a classification analysis (SVMsC) to construct multivariate calibration models. The present method provides clinically promising BGL measurements with multivariate regressions. Also, a proper classification model was obtained using the nonlinear method, SVMsC. These results provide preliminary evidence that *"pulse glucometry"* with SVMsR can be applied effectively to predict BGL levels non-invasively and a classification type calibration could be applicable for diabetes screening and health care in normal subjects in a more practical, easier and more convenient way through rapid BGL assessment. Further comprehensive studies will need to be carried out in order to investigate an optimal construction of multivariate regression and classification models including data from diabetic subjects as well as to design a compact BGL device based on "*pulse glucometry*" for practical use.

ACKNOWLEDGMENT

The authors wish to give their sincere thanks to Professor K. Yamakoshi, Kanazawa University, and Prof P. Rolfe, Harbin Institute of Technology, China, and OBH Ltd., UK, for their valuable suggestions and comments on this work and for their assistance in preparing the manuscript. This work was partly supported by Grant-in-Aid for Scientific Research (A) (21240047), The Ministry of Education, Culture, Sports, Science and Technology, Japan.

REFERENCES

[1] Lawton J, Peel E, Douglas M, Parry O. `Urine testing is a waste of time': newly diagnosed Type 2 diabetes patients' perceptions of self-monitoring. Diabetes Med 2004; 21(9): 1045-8.

[2] Tierney MJ, Tamada JA, Potts RO, *et al.* The GlucoWatch biographer: a frequent automatic and noninvasive glucose monitor. Ann Med 2000; 32(9): 632-41.

[3] Eastman RC, Chase HP, Buckingham B, *et al.* Use of the GlucoWatch biographer in children and adolescents with diabetes. Pediatric Diabetes 2002; 3(3): 127-34.

[4] Aslan K, Lakowicz JR, Geddes CD. Nanogold-plasmon-resonance-based glucose sensing. Anal Biochem 2004; 330(1): 145-55.

[5] MacKenzie HA, Ashton HS, Spiers S, *et al.* Advances in photoacoustic noninvasive glucose testing. Clin Chem 1999; 45(9): 1587-95.

[6] Maier JS, Walker SA, Fantini S, Franceschini MA, Gratton E. Possible correlation between blood glucose concentration and the reduced scattering coefficient of tissues in the near infrared. Opt Lett 1994; 19(24): 2062-4.

[7] Marbach R, Koschinsky T, Gries FA, Heise HM. Noninvasive blood glucose assay by near-infrared diffuse reflectance spectroscopy of the human inner lip. Appl Spectrosc 1993; 47(7): 875-81.

[8] Zeller H, Novak P, Landgraf R. Blood glucose measurement by infrared spectroscopy. Int J Artif Organs 1989; 12(2): 129-35.

[9] Berger AJ, Itzkan I, Feld MS. Feasibility of measuring blood glucose concentration by near-infrared Raman spectroscopy. Spectrochim Acta A Mol Biomol Spectrosc 1997; 53A(2): 287-92.

[10] Heise HM. In: Siesler HW, Ozaki Y, Kawata S, Heise HM, Eds. Near-Infrared Spectroscopy. Wiley-VCH : Weinheim 2002.

[11] Cote GL, Fox MD, Northrop RB. Noninvasive optical polarimetric glucose sensing using a true phase measurement technique. IEEE Trans Biomed Eng 1992; 39(7): 752-6.

[12] Guclu B, Engbretson GA, Bolanowski SJ. Constrained optimization of Drude's equations eliminates effects of confounding molecules for the polarimetric measurement of glucose. J Biomed Opt 2004; 9(5): 967-77.

[13] Yakmakoshi K, Yamakoshi Y. Pulse glucometry: a new approach for noninvasive blood glucose measurement using instantaneous differential near-infrared spectrophotometry. J Biomed Opt 2006; 11(5): 054028.

[14] Yamakoshi K. A Non-invasive Blood constituent Measuring Instrument and Measuring Method, International Patent PCT/JP03/03587, Mar 25 2003.

[15] Yamakoshi K, Yamakoshi Y, Nogawa M, Yamakoshi T, Tanaka S. A New Approach for Noninvasive Blood Glucose Measurement Using Differential Near Infrared Spectrophotometry. In: Proceedings of World Congress on Medical Physics and Biomedical Engineering 2006.

[16] Wold S, Sjöström M, Eriksson L. PLS-regression: a basic tool of chemometrics. Chemometrics Intell Lab Syst 2001; 58(2): 109-30.

[17] Cristianini N, Shawe-Taylor J. An Introduction to Support Vector Machines and other kernel-based learning methods. Cambridge University Press: Cambridge 2000.

[18] Trafalis TB, Gilbert RC. Robust classification and regression using support vector machines. Elsevier BV: Amsterdam 2006.

[19] Beebe KR, Pell RJ, Seasholts MB, Eds. Chemometrics: a practical guide. Wiley: New York 1998.

[20] Alberti KGMM, Zimmet PZ for the WHO Consultation Group. Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus Provisional report of a WHO consultation. Diabetes Med 1998; 15(7): 539-53.

[21] R Development Core Team, R: A language and environment for statistical computing. R Foundation for Statistical Computing: Vienna 2005.

[22] Ihaka R, Gentleman RR. A language for data analysis and graphics. J Comput Graph Stat 1996; 5(3): 299-314.

[23] Karatzoglou, A., Smola, A., hornik, K., Zeileis,A. kernlab-An S4 Package for Kernel Methods in R. J Stat Software 2004; 11, Issue 9.

[24] Clarke WL, Cox D, Gonder-Frederick LA, Carter W. Evaluating clinical accuracy of systems for self-monitoring of blood glucose. Diabetes Care 1987; 10(5): 622-8.

[25] Thissen U, Pepers M, Ustun B, Melssen WJ, Buydens LMC. Comparing support vector machines to PLS for spectral regression applications. Chemometrics Intell Lab Syst 2004; 73(2): 169-79.

[26] Borin A, Ferrão MF, Mello C, Maretto DA, Poppi RJ. Least-squares support vector machines and near infrared spectroscopy for quantification of common adulterants in powdered milk. Anal Chim Acta 2006; 579(1): 25-32.

[27] Aoyagi T, Miyasaka K. Pulse oximetry: its invention, contribution to medicine, and future tasks. Anesth Analg 2002, 94(Suppl 1): S1-3.

[28] American Diabetes Association. Standards of medical care in diabetes -- 2008. Diabetes Care 2008; 31(Suppl 1): S12-S54.

Received: June 8, 2009 Revised: July 29, 2009 Accepted: July 30, 2009