A New Method of Screening for Diabetic Neuropathy using Laser Doppler and Photoplethysmography

Sung Woo Kim

The Graduate School
Yonsei University
Graduate Program in Biomedical Engineering

A New Method of Screening for Diabetic Neuropathy using Laser Doppler and Photoplethysmography

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Sung Woo Kim

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This certifies that the dissertation of Sung Woo Kim is approved.

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Thesis Supervisor: Deok Won Kim

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Eun Seok Kang

_____

Hee Cheol Kang

_____

Taick Sang Nam

_____

Jong Gwan Yook

The Graduate School
Yonsei University
June 2015

감사의 글

이제 하나의 결실이 맺어지고 있습니다. 꿈같던 지난 날을 생각해보면 잠시 머무는 인생 길에서 새로운 소명과 비전을 보여주시고 오늘도 동행하시는 하나님께 모든 영광 올립니다.

늘 학자로서의 참된 소양과 지식, 바른 길로 지도해 주셨던 김덕원 교수님께 진심으로 감사 드립니다. 연세의대 본관을 지나 교수님을 찾아 뵈었던 그 길이 항상 마음 깊이 남을 듯 합니다. '晝耕夜讀'으로 논문 출판을 준비하던 제게 큰 격려와 조언을 아낌없이 주신 육종관 교수님께 감사 드립니다. 신촌 세브란스병원을 오랜만에 방문해서 인사 드려도 반갑게 맞아 주신 내분비내과 강은석 교수님, 가정의학과 강희철 교수님과 연세의대 생리학교실에 계신 남택상 교수님께 감사 드립니다.

지금까지 함께했던 연세대 동기들 선후배님, 학교 친구들과 선생님, 직장 동료들과 모든 친인척 분들께도 감사 드립니다. 사랑으로 낳아주시고 언제나 힘써 길러주신 아버지 어머니께, 누구보다 깊은 배려와 기도로 큰 힘이 되어주시는 장인어른 장모님께 항상 감사 드립니다. 나와 아내를 똑 닮은 사랑하는 아들 지후와 딸 하빈이에게 자랑스런 아빠로 있어주고 싶고,

끝으로 이세상 단 하나뿐인 내 아내에게 정말 사랑한다고 언제나 그 자리에 변함없는 소나무 같이 그대를 나의 소중한 평생 친구로 반려자로 아껴주고 삶의 그루터기가 되려한다고 말하고 싶습니다.

2015년 6월
김성우 드림

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ABSTRACT

A new method of screening for diabetic neuropathy using laser Doppler and photoplethysmography

Sung Woo Kim

Graduate Program in Biomedical Engineering
The Graduate School, Yonsei University

(Directed by Professor Deok Won Kim)

The purpose of this study is to suggest a simple, new method of screening for diabetic neuropathy. We measured blood volume changes by photoplethysmography (PPG) and blood perfusion by laser Doppler (LD) in the index fingers and big toes in 40 control subjects and in 50 (19 mild, 17 moderate, and 14 severe based on the nerve conduction velocity (NCV) test) and 35 diabetic patients with and without neuropathy, respectively. According to the results of PPG and LD measurements, the toe to finger ratios obtained from the neuropathic group were significantly higher than those from the control ($p<0.001$) and the non-neuropathic groups ($p<0.001$). Based on the NCV, the sensitivity of the LD method (92.0%) was higher than that of the PPG method (84.0%) for both left and right sides. Although specificity of the LD (92.8%) was also higher than the PPG

(84.3%) bilaterally, the PPG showed better reproducibility (5.5% versus 9.5%) and a significant ratio increase with severity, while the LD did not. Our suggested PPG method using the toe to finger ratio is reliable, simple, economical, and accurate, and could become a new effective screening tool for the early detection of diabetic neuropathy.

Key words: Blood volume change, Diabetic neuropathy, Laser Doppler, Photoplethysmography, Toe to finger ratio

I. INTRODUCTION

The incidence of diabetes (i.e. diabetes mellitus) is increasing dramatically due to economic development and lifestyle changes. Some chronic diseases such as diabetes and hypertension are also reaching epidemic proportions, because our eating habits and life-related behaviors have changed over the past decade [1].

The diabetes is known as a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. There is a pathogenic process which induce the development of diabetes; this range from autoimmune destruction of the pancreatic β-cells with consequent insulin deficiency to abnormalities that result in resistance to insulin action [2]. A glucose metabolism describes as the mechanism of the insulin action in Figure 1-2.

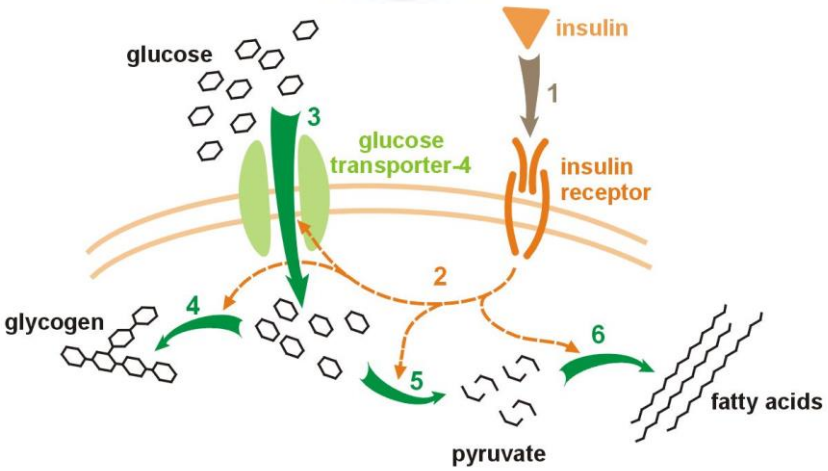

Fig. 1. Insulin glucose metabolism [3].

The basis of the abnormalities in carbohydrate, fat, and protein metabolism in diabetes is deficient action of insulin on target tissues. Deficient insulin action results from inadequate insulin secretion and/or diminished tissue responses to insulin at one or more points in the complex pathways of hormone action. The diabetes can be classified into the four categories; type I diabetes, type II diabetes, gestational diabetes mellitus, and other specific types (e.g. monogenic diabetes syndromes, diseases of the exocrine pancreas, drug- or chemical-induced diabetes). Among these types, main types are often defined as “type I” or “type II” diabetes. Type I diabetes occurs when these cells are destroyed by the body’s own immune system. It is also known as an insulin-dependent diabetes mellitus and a disease characterized by high levels of blood glucose resulting from defects in insulin secretion, insulin action, or both. Autoimmune, genetic, and environmental factors are involved in the development of type I diabetes. Type II diabetes is the most common type of diabetes. In type II diabetes, the body is able to produce insulin but it is either not sufficient or the body is not responding to its effects, leading to a build-up of glucose in the blood. In contrast to people with type I diabetes, the majority of those with type II diabetes do not usually require daily doses of insulin to survive. However, they may be prescribed insulin together with oral medication, a healthy diet and increased physical activity to manage their condition. Type II diabetes usually occurs in adults, but it is now increasingly seen in children and

adolescents. The development of type II diabetes is influenced by a number of risk factors such as an age, an overweight, a family history, an obstetric history, a physical inactivity and a poor diet [2].

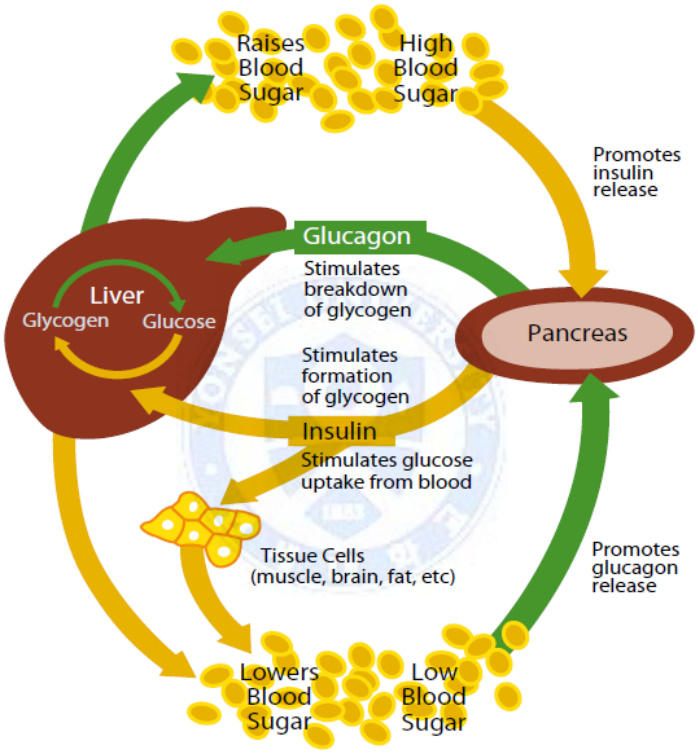

Fig. 2. Phenomenon of the insulin production and action [7].

The Korean Diabetes Association reported that the Korean diabetic population increased approximately 10-fold from 1970 to 2001 [4]. The Growth from Knowledge (GfK) market measures reported that the diabetic population in the United States had increased by approximately 86% over the decade. Figure 3 shows an overall increase of the U.S. diabetes prevalence from 1995 to 2005 [5].

As shown in Figure 4, the prevalence of diabetes was 12.4% in Korean adults aged 30 years and older. About 4.0 million Koreans (male: 2.2 million, female: 1.7 million) had diabetes, 20% of Korean adults were diagnosed as having impaired fasting glucose, and 28% of the subjects with diabetes were undiagnosed. Moreover, One-third of patients with diabetes have only reached their target blood glucose level. Even if a relatively generous recommendation of the American Diabetes Association (ADA) was applied, about 43% of patients were under adequate glycemic control. The other one-third of patients with diabetes had microvascular complications such as diabetic neuropathy, diabetic retinopathy, and peripheral artery disease. The Korean diabetic population expected to reach about 6 million in 2050. They were determined from data on the 2011 Korea National Health and Nutritional Examination Survey (KNHANES) conducted by the Korea Centers for Disease Control and Prevention (KCDC) and the Korean Ministry of Health and Welfare [6].

Recently, World Health Organization (WHO) and International Diabetes

Federation (IDF) reported an official document to measure the global diabetic population in 2014 [7]. It was estimated that 386.6 million people have diabetes for the 220 countries and territories (male: 199.2 million, female: 187.4 million).

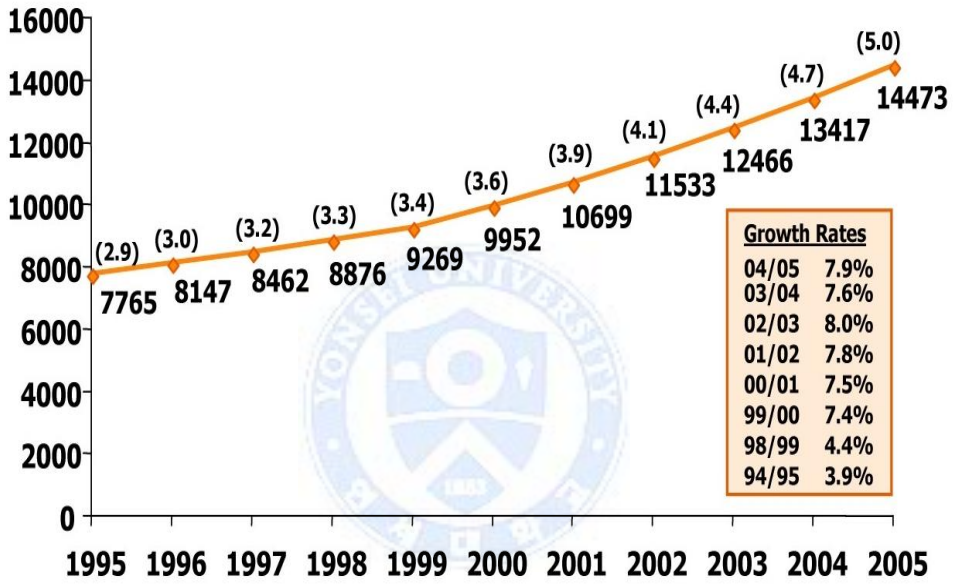

Fig. 3. An overall increase of the U.S. diabetes population over decade.

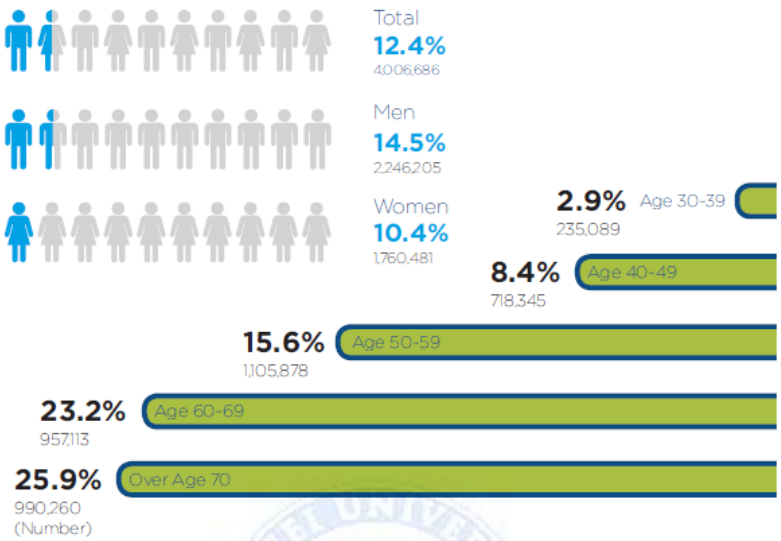

Fig. 4. The prevalence of diabetes in Korean adults aged 30 years and older.

This diabetes mellitus can cause damage to the nerves throughout the body when blood glucose and blood pressure are too high. It can lead to problems with digestion, erectile dysfunction, and many other functions. Among the most commonly affected areas are the extremities, in particular the feet. Nerve damage in these areas is called peripheral neuropathy, and can lead to pain, tingling, and loss of feeling as shown in Figure 5. Loss of feeling is particularly important because it can allow injuries to go unnoticed, leading to serious infections and possible amputations. People with diabetes carry a risk of amputation that may be more than 25 times greater than that of people without diabetes. However, with comprehensive management, a large proportion of amputations related to diabetes can be prevented. Even when amputation takes place, the remaining leg and the person's life can be saved by good follow-up care from a multidisciplinary foot team. People with diabetes should regularly examine their feet.

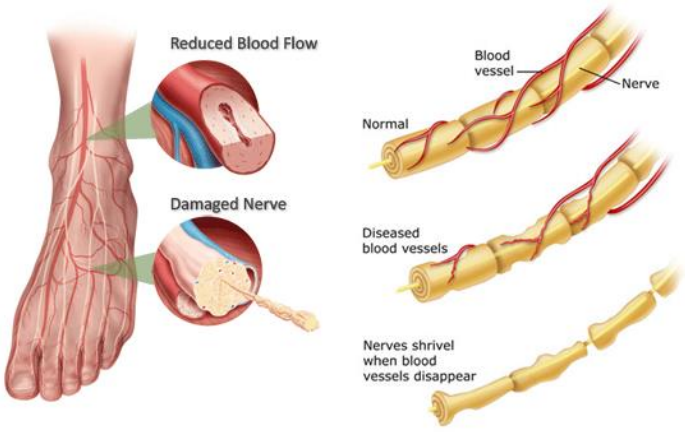

Fig. 5. Damaged nerves with the diabetes.

Among many other diabetic complications, diabetic foot disease is considered one of the most serious as it may cause ulceration and subsequent amputation of the legs as shown in Figure 6. It was reported that approximately 30,000 diabetic patients undergo foot surgery each year in the United States [8]. According to the national diabetes statistics report (Centers for Disease Control and Prevention, U.S. Department of Health & Human Services, 2014) which was recently based on scientific data and epidemiologic estimation of the U.S. diabetes population, it also mentioned that the amputation was increased about 73,000 non-traumatic lower-limb amputations of adults with diagnosed diabetes in 2010. About 60% of non-traumatic lower-limb amputations among people aged 20 years or older occurred in people with diagnosed diabetes. These, however, are preventable by early diagnosis of diabetic foot [9].

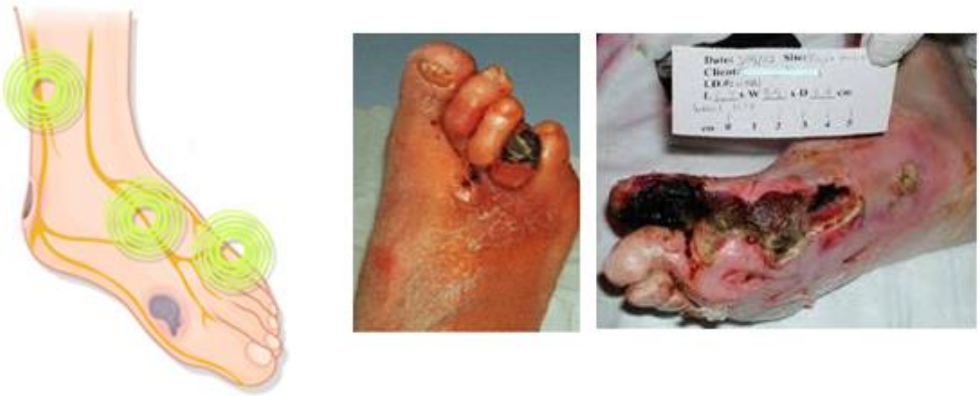

Fig. 6. The dangerous cases of the diabetic foot.

The causes of diabetic foot disease include neuropathy, neuro-ischemia, and ischemia. Of these etiologies, neuropathy is the most common and accounts for up to 80-90% of the diabetic foot population [10]. Diabetic neuropathy results from nerve damage and can lead to loss of sensation or to abnormal feelings in the feet. It may even increase the likelihood of foot injuries developing into ulcers [8].

As shown in Figure 7, the nerve conduction velocity (NCV) test has been considered the gold standard method for diagnosing diabetic neuropathy, but it requires the application of a strong electrical stimulus to nerves, causing discomfort and pain in patients [11].

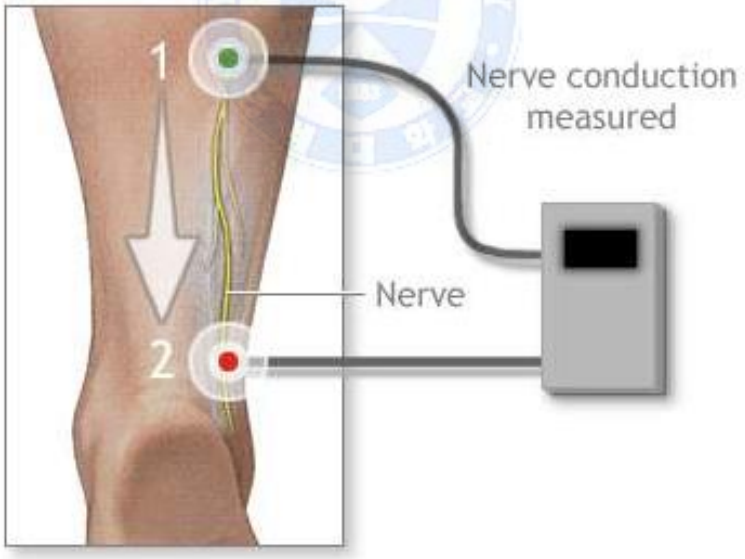

Fig. 7. Nerve conduction velocity test.

Many studies have been conducted by measuring blood volume changes in the fingers and toes because it was reported that patients with diabetic neuropathy have increased blood flow to their toes [12-17]. Decreased sympathetic tone in the feet of neuropathy patients results in an open arteriovenous shunt (AVS). This causes increased blood flow in the feet [18] and toes, but does not have an effect on blood flow in the fingers [17]. On the contrary, diabetics with neuropathy showed smaller finger pulp and larger toe pulp blood flows than non-diabetics using laser Doppler (LD) [19]. As shown in Figure 8, an arteriovenous shunt is increasingly opened by the disappearance of the peripheral sympathetic nerve.

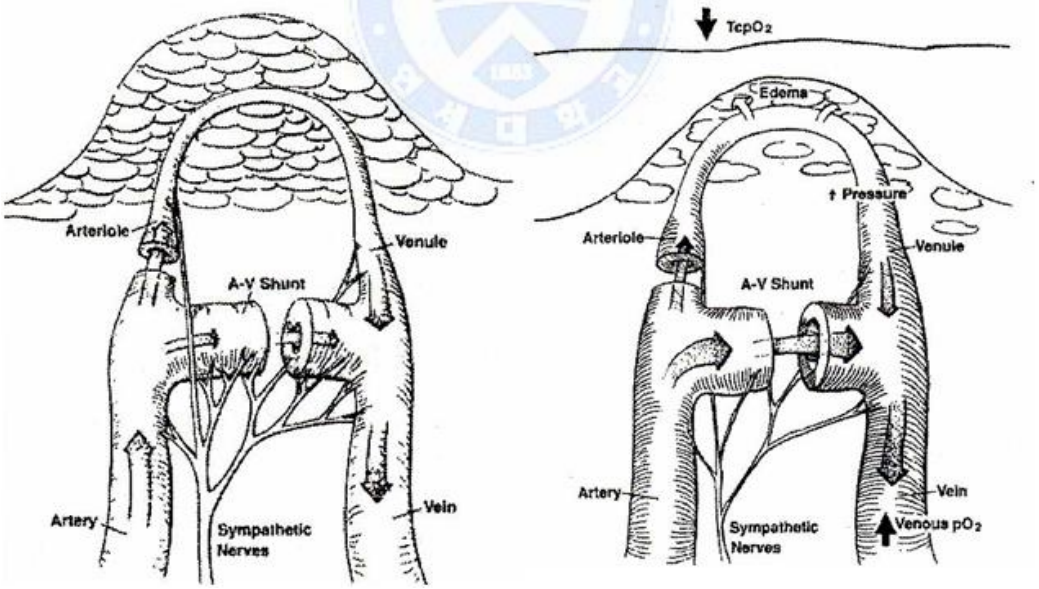

Fig. 8. An arteriovenous shunt in the diabetic neuropathy.

Therefore, the toe to finger blood flow ratio in diabetics with neuropathy is larger than that in diabetics without it due to the presence of constant or decreased finger blood flow coupled with an increased toe blood flow. Based on these findings, this ratio may be a promising new screening parameter for diabetic neuropathy. In addition, by using the ratio, we can minimize variation due to difference in absolute blood volume changes among subjects and in skin temperature during measurement.

LD has been used for measuring foot microcirculation [20-22] as shown in Figure 9. LD and photoplethysmography (PPG) have been used for monitoring blood perfusion in skin, venous reflux conditions, and skin flaps during plastic surgery [23]. In this study, the noninvasive techniques of PPG and LD were used to measure the blood volume changes and perfusions of the fingers and toes, respectively. We found optimal ratios for PPG and for LD that can distinguish diabetic patients with and without neuropathy. In addition, we determined the sensitivity and specificity using the NCV test, and the reproducibility of both PPG and LD.

Fig. 9. A measurement of foot microcirculation using laser Doppler.

II. MATERIALS AND METHODS

1. Participants

Three groups of subjects were studied. The first group included 40 healthy, non-diabetic subjects, the second group included 35 diabetic patients without neuropathy, and the third group included 50 diabetic patients with neuropathy. Of those 50 diabetic patients with neuropathy, there were 19 mild, 17 moderate, and 14 severe as determined by the NCV test. The diabetic patients with neuropathy had been diagnosed by the NCV test at the Yonsei University Medical Center, Seoul, Korea.

As shown in Table 1, all groups were matched for sex, age, and body mass index (BMI), and the two diabetic groups were also matched for type and duration of diabetes. The control subjects had a fasting glucose level between 3.9-5.8 mmol/L and less than 6.7 mmol/L two hours after breakfast. None of the control subjects reported current use of any medications. All subjects were informed about the purpose of and procedure for the study and subsequently gave their informed consent. The Yonsei Medical Research Ethics Committee reviewed the full protocol and approved this study.

Table 1. Characteristics of experimental groups.

	Control	Diabetic groups		p-value
		Non-neuropathy	Neuropathy	
Number of subjects	40	35	50	-
Sex [M/F]	18 / 22	15 / 20	21 / 29	-
Age [years]	65.8 ± 8.9	61.0 ± 8.0	65.1 ± 9.0	0.099
Body mass index [kg/m2]	22.6 ± 1.4	23.5 ± 2.7	22.9 ± 3.3	0.261
Systole [mmHg]	121.2 ± 8.5	126.5 ± 15.0	131.1 ± 18.9	0.236
Diastole [mmHg]	79.5 ± 3.2	78.8 ± 10.2	77.9 ± 10.4	0.680
Diabetes duration [years]	-	16.7 ± 6.7	13.7 ± 8.6	0.380
Type I	-	1	2	-
Type II	-	34	48	-
Smokers	9	7	4	-
HbA1c [%]	-	10.2 ± 5.6	9.8 ± 2.8	0.493
Fasting glucose [mmol/L]	< 5.8	8.3 ± 2.1	8.8 ± 4.0	0.429
HDL [mg/dL]	-	52.9 ± 15.1	50.2 ± 17.6	0.466
LDL [mg/dL]	-	122.1 ± 13.9	102.7 ± 35.2	0.053

2. Hardware for PPG measurement system

The wavelengths of the PPG sensors (DS0-100A Durasensor, Nellcor, USA) were 660 nm (red) and 940 nm (infrared) as shown in Figure 10. Signal amplification, filtering, and normalization were performed using the operational amplifiers (TL082, Texas Instruments, USA), 10 Hz low pass filter, and PIC microcontroller (PIC 16C711, Microchip, USA), respectively. The final signal was sent to a notebook computer (Sens V20, Samsung, Korea) through a DAQ-pad (PCI-6020E, National Instruments, USA) as shown in Figure 11-12. For accurate and precise measurement, the PPG signal measurement system was calibrated by connecting the output of the SpO_2 simulator (Index® 2_{XLFE}, Fluke, USA) to the input of the PPG system. Then, the output signals from both instruments were matched by adjusting the potentiometer of the PPG system for all four channels. Our constructed PPG measurement system utilized four channels to allow the simultaneous measurement of the PPG signals from the left and right fingers and toes.

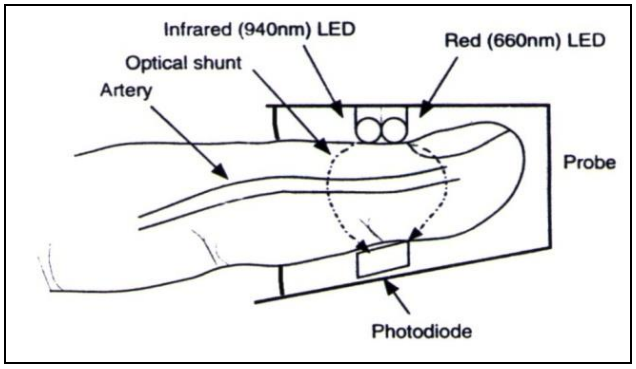

Fig. 10. A diagram of photoplethysmography sensor.

Fig. 11. A photo of 4-ch photoplethysmography measurement device.

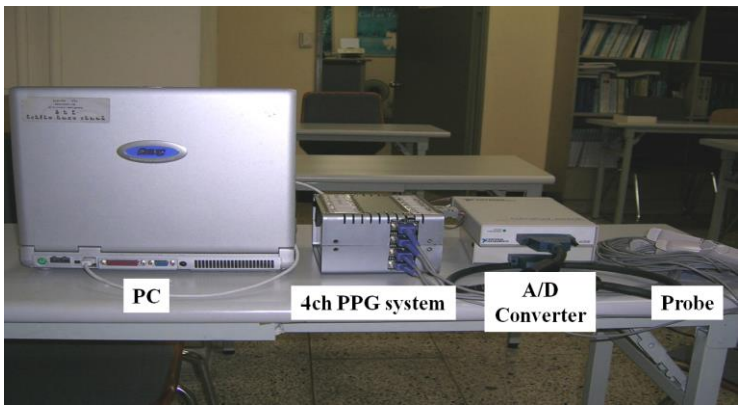

Fig. 12. A photo of the 4-ch PPG measurement system.

3. Software of PPG signal measurement system

As shown in Figure 13-14, LabVIEW 6.1 (National Instrument, USA) was used to develop the real time data acquisition and signal analysis program. The amplitudes representing the differences between the peaks and valleys of each red LED waveform were added and averaged within the selected window to obtain the mean blood volume changes of the fingers and toes. They were then used to obtain the left and right blood volume change ratios. Finally, the toe to finger ratio was used to minimize the difference in the absolute blood volume and skin temperature between each subject as shown in Figure 15.

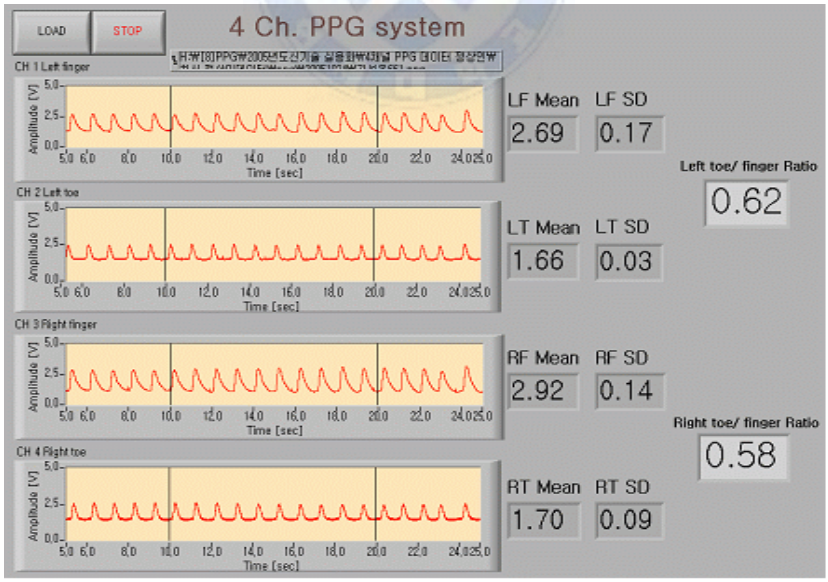

Fig. 13. An example of the 4-ch PPG measurement software.

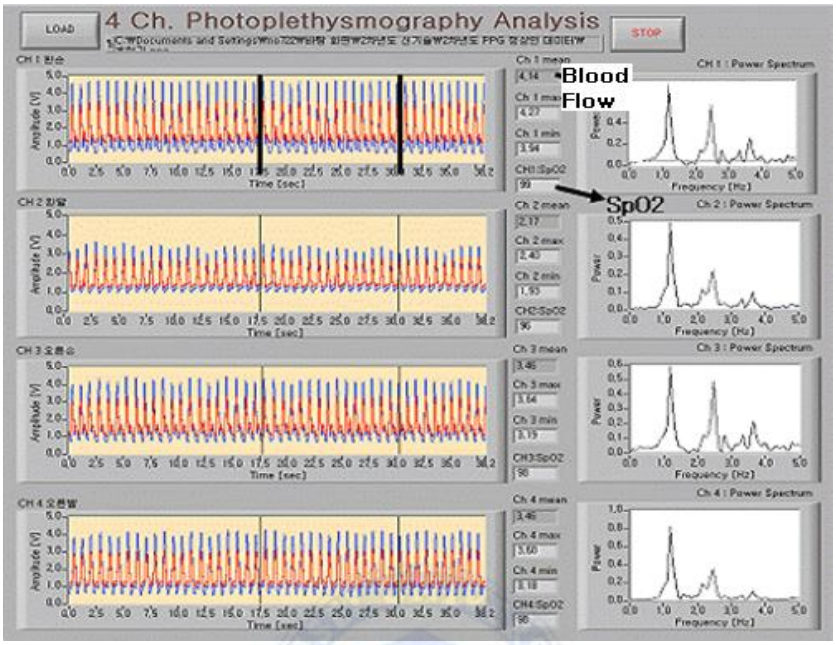

Fig. 14. Real-time data acquisition and analysis program.

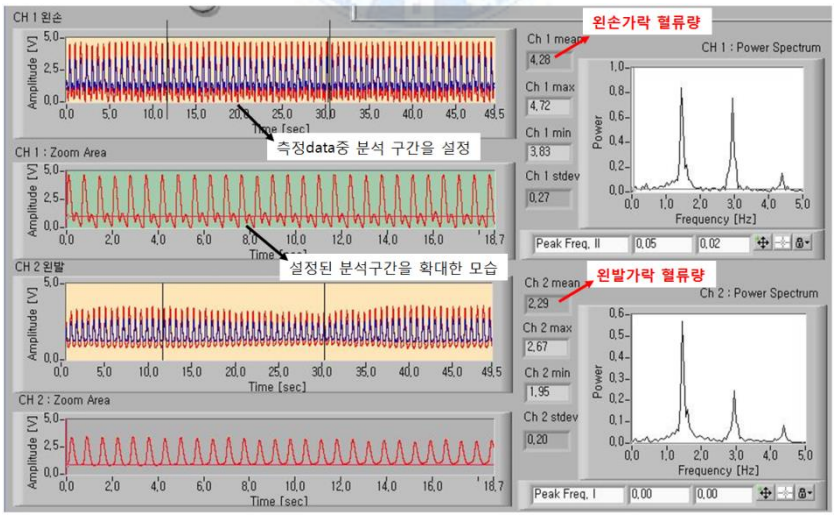

Fig. 15. Post-processing software of the photoplethysmogram.

4. Experimental procedure

4.1 Nerve Conduction Velocity Test

The NCV test (Neuroscreen, Jaeger & Toennies, Wuerzburg, Germany) was performed by a clinician at the Yonsei University Medical Center, Seoul, Korea. An active electrode was placed over the nerve segment being studied. The median and ulnar nerves were tested for the upper limb, and the peroneal, tibial, and sural nerves were tested for the lower limb. The motor nerves (median, ulnar, peroneal, and tibial nerves) and sensory nerves (median, ulnar, and sural nerves) were examined to determine the presence of neuropathy. Diabetic patients participating in this study were diagnosed with mild, moderate, or severe neuropathy if abnormality occurred on 1-2 nerves, 3-5 nerves, or 6-7 nerves, respectively. A total of 50 diabetic patients were diagnosed with neuropathy by the clinician.

4.2 Measurement procedure

The procedure for the measurements was as follows. Subjects rested in the supine position for a minimum of 10 minutes before beginning the experiment. The PPG signals from the index finger and first toe for both the left and right sides were simultaneously recorded in triplicate in the supine position by our constructed system. Bilateral blood perfusion and the temperatures of fingers and toes were also simultaneously recorded in triplicate using an LD perfusion monitoring and temperature unit (PF 5010 and 5020, Perimed, Sweden). The small angled thermostatic probes (457, Perimed, Sweden) were used to measure perfusion and temperature simultaneously with double-sided adhesive strips (PF 10-3, Perimed) as shown in Figure 16-17.

PeriSoft for Windows (ver 2.5, Perimed) software was used for data storage and analysis. Each measurement lasted 30 seconds and was recorded three times in order to verify the reproducibility of PPG and LD. The electrodes were replaced three times for each repeated PPG and LD measurement. For later analysis, stable ten-second intervals of PPG and LD signals were selected. The room temperature was maintained at 23 ℃ during the experiment.

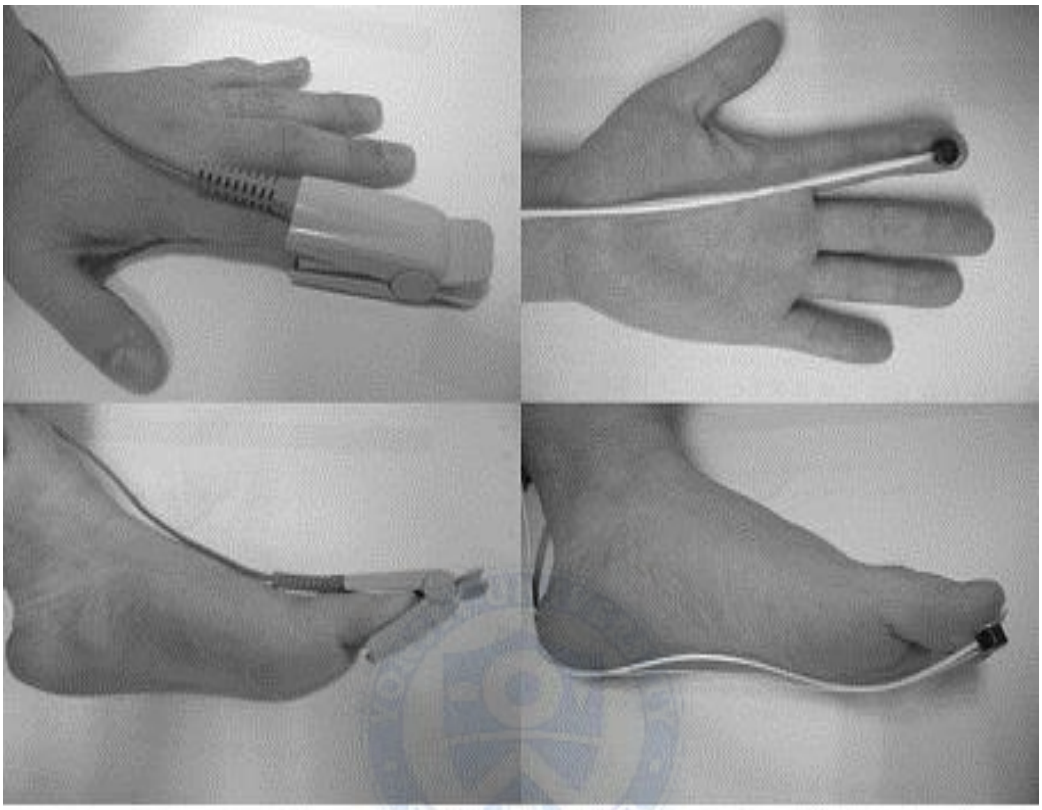

Fig. 16. The location of PPG sensors and LD probes during measurement.

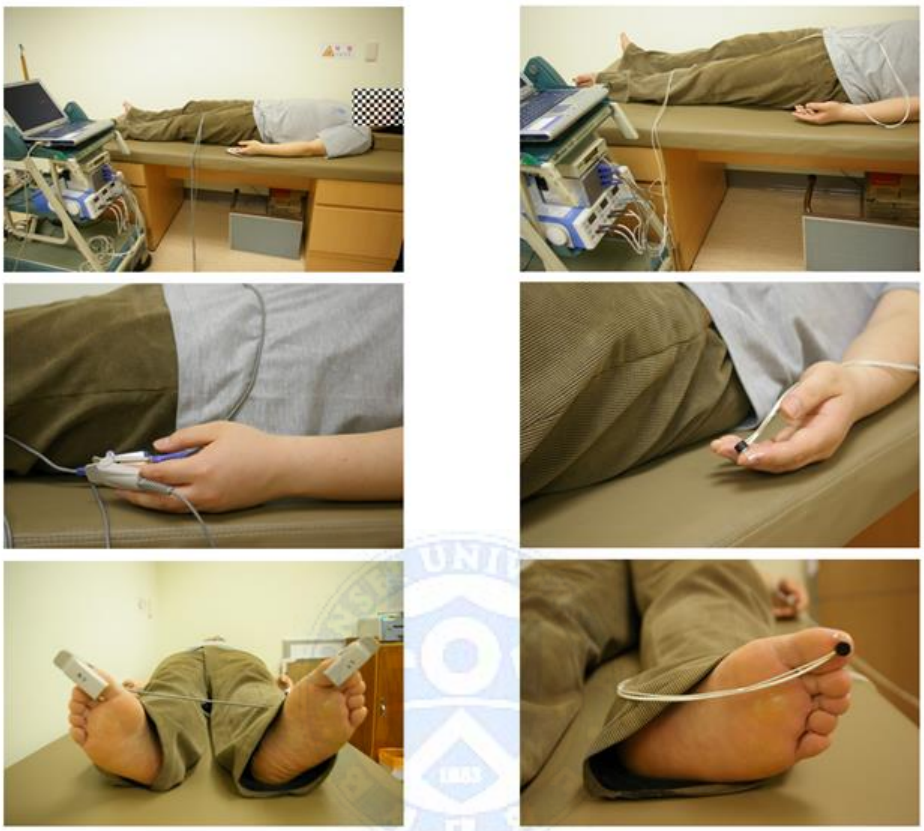

Fig. 17. A photo of measurement using the PPG and LD devices.

5. Statistical analysis

All data are shown as means and standard deviations. $p<0.05$ was considered statistically significant. The independent sample t-test and one-way ANOVA test were performed using SPSS 10.0 for Windows (SPSS Inc, Chicago, IL, USA). The Bonferroni multiple comparison method was performed for further analysis in the ANOVA test.

III. RESULTS

1. Clinical characteristics

There were no statistically significant differences among the three groups in age (p=0.099), BMI (p=0.261), systolic (p=0.236) or diastolic (p=0.680) blood pressures (Table 1). There were also no statistically significant differences between the two diabetic groups in diabetes duration (p=0.380), glycolysed hemoglobin A1c (HbA1c, p=0.493), fasting glucose (p=0.429), high density lipoprotein (HDL, p=0.466), or low density lipoprotein (LDL, p=0.053).

2. Blood volume change and skin temperature

Because there were no statistically significant differences between the left and right sides of fingers and toes in blood volume changes and perfusion, the left and right blood volume changes and perfusion were pooled. Blood volume changes and temperatures of fingers and toes were obtained by the PPG for the control subjects and patients with and without neuropathy. The toe temperature, ranging from 25 to 34 ℃, had a wider distribution than that of the finger which ranged from 29 to 35 ℃. There were no significant differences in the toe and finger temperatures among the three groups.

Figure 18 shows finger and toe blood volume changes measured by the PPG (a, b) and LD (c, d) for the five groups. The bold lines inside the box plots indicate median values. The upper and lower lines of the box are at the 25th and 75th percentiles and the top and bottom whiskers are the highest and lowest values, respectively. The significance p levels were shown among the control, non-neuropathic, and neuropathic groups pooling the three groups, and among the mild, moderate, and severe groups. As expected in Figs. 18a and 18b, the PPG method showed no significant differences in blood volume changes between the control and non-neuropathic groups in the fingers and toes, respectively. However, there was a significant decrease in the blood volume changes of the neuropathic group

compared with that of the non-neuropathic group in the fingers ($p<0.01$) and a significant increase in the toes ($p<0.001$). Furthermore, there was a significant decrease in the blood volume changes of the neuropathic group compared with that of the control group in the fingers ($p<0.001$) and a significant increase in the toes ($p<0.001$). The above finding is consistent with the study wherein 76% of diabetics with neuropathy showed smaller finger pulp and larger toe pulp blood flows than non-diabetics [19]. Therefore, this finding not only validated previous studies [12-16, 19, 24] but confirmed our assumption that the toe blood volume changes in the neuropathic group would be larger than those in the non-neuropathic group.

As shown in Figs. 18c and 18d, there was a significant difference in the finger blood perfusion measured by the LD between the control and non-neuropathic groups ($p<0.01$). This finding by LD is different from that by PPG, and while future studies are required to further investigate this discrepancy, it may be due to the different methodology and measured volume sizes of the PPG and LD methods. Though the toe blood perfusion of the neuropathic group measured by LD was larger than those of the control and non-neuropathic groups, it was not significant ($p>0.05$). This finding is consistent with a Nabuurs-Fransen study [22] wherein foot LD flux in patients with peripheral polyneuropathy was higher than that of the control without significance (7.4 vs. 5.9). While the measured position

(foot vs. toe) was different between their study and ours, both studies showed the same trend.

The above disparity between the PPG and LD was mitigated or eliminated by utilizing the ratio of toe to finger blood volume change (or perfusion) as discussed in the following section (Fig. 20).

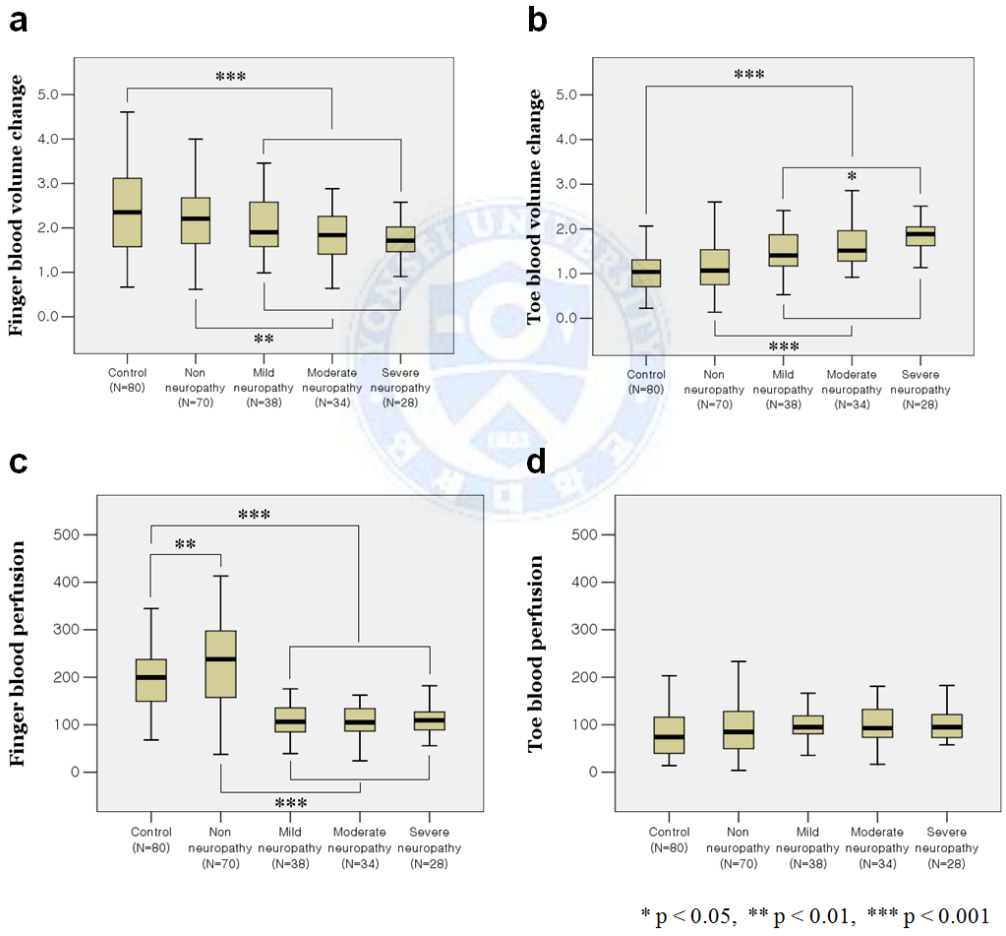

Fig. 18. Box plots for finger (a) and toe (b) blood volume changes by PPG, and for finger (c) and toe (d) blood perfusion for the five groups.

3. Blood volume change ratios and temperature differences

Figure 19 shows the temperature difference between fingers and toes and the ratios of blood volume change by PPG (a) and perfusion by LD (b) for the control (n=80), non-neuropathic (n=70), and neuropathic groups (n=100). The vertical lines are the optimal ratios found applying one of the three methods, which will be discussed on the next section. The temperature differences of the control and non-neuropathic groups were widely distributed in the range of 0 to 8 ℃, while those of the neuropathy group were distributed from 0 to 4.5 ℃. There were no neuropathy patients with a temperature difference greater than 4.5 ℃. This finding suggests that a subject is not considered neuropathic if his or her temperature difference is higher than 4.5 ℃. The two vertical lines in Figs. 19a and 19b are the optimal boundaries differentiating neuropathic from non-neuropathic and control groups, and will be explained later in this paper.

Figure 20 shows the box plots for the calculated ratios for the five groups by the PPG and LD methods. There were significant differences in the blood volume change ratios between the control and neuropathic groups ($p<0.001$) as well as between the non-neuropathic and neuropathic groups ($p<0.001$), but not between the control and non-neuropathic groups ($p>0.05$) by both methods. The difference between the PPG and LD methods is that there are ratio increases with severity for

PPG but not for LD for the neuropathic group. In this sense, PPG is a superior method.

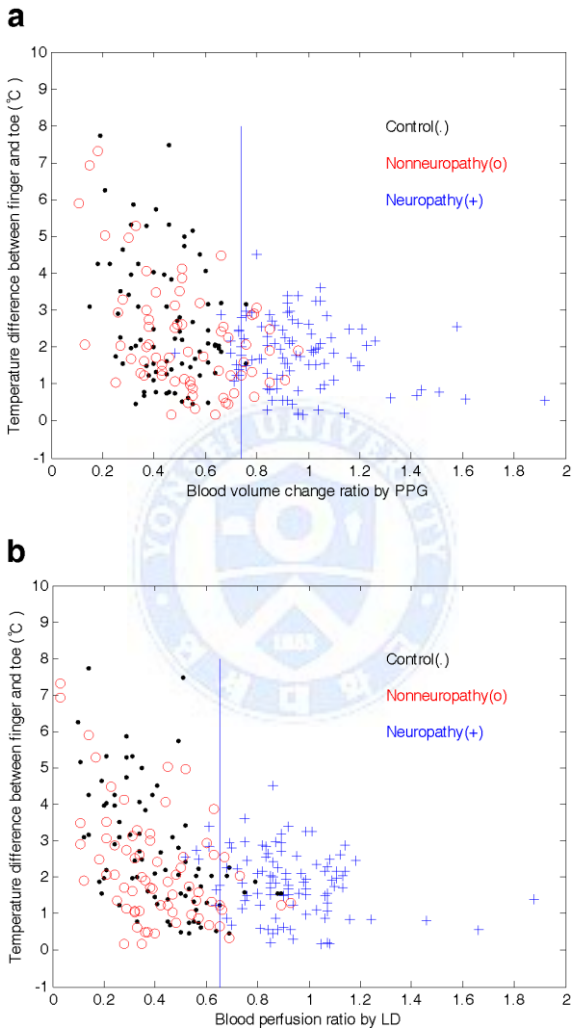

Fig. 19. PPG blood volume change ratio (a) and LD perfusion ratio (b) with temperature differences between fingers and toes by the LMS method (n = 80 for control, n = 70 for non-neuropathy, n = 100 for neuropathy).

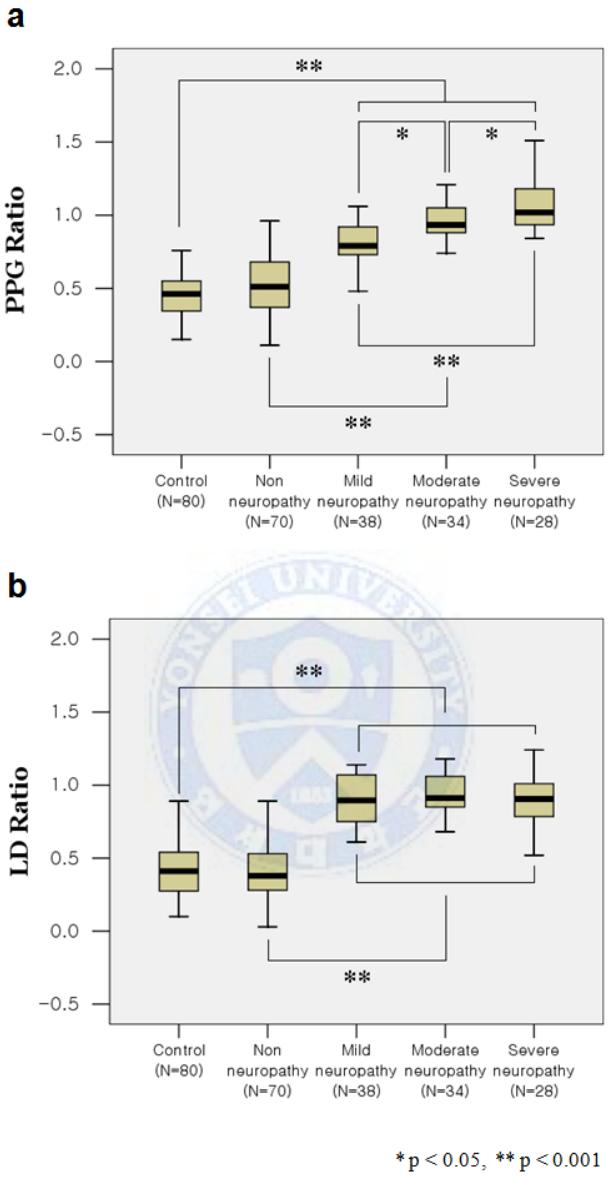

Fig. 20. Box plots for PPG blood volume change ratio (a) and LD perfusion ratio (b) for the five groups.

The reproducibility ((standard deviation/mean) x 100%) of the PPG method for the control, non-neuropathic, and neuropathic groups was 8.0 ± 5.9%, 2.8 ± 1.9%, and 5.4 ± 4.9%, respectively. The total mean reproducibility was 5.5 ± 5.1%. Those of the LD were 13.8 ± 9.1%, 5.3 ± 3.7%, and 9.0 ± 6.1%, respectively. The total mean reproducibility was 9.5 ± 7.5%. Reproducibility is one of the most important factors for reliable diagnosis in medicine, and in this regard PPG is the superior method as shown in Table 2.

Table 2. Comparison of reproducibility between PPG and LD.

Method	Group	Reproducibility (%)	Total mean reproducibility (%)
PPG	Control	8.0 ± 5.9	5.5 ± 5.1
	Non-neuropathy	2.8 ± 1.9	
	Neuropathy	5.4 ± 4.9	
LD	Control	13.8 ± 9.1	9.5 ± 7.5
	Non-neuropathy	5.3 ± 3.7	
	Neuropathy	9.0 ± 6.1	

4. Sensitivity and specificity of PPG and LD

Sensitivity and specificity were calculated as (1) and (2) where TP = true positive, TN = true negative, FP = false positive, and FN = false negative.

$$Sensitivity(\%) = \frac{TP}{TP + FN} \times 100 \quad (1)$$

$$Specificity(\%) = \frac{TN}{TN + FP} \times 100 \quad (2)$$

We applied Bayesian, least mean square (LMS), and Receiver Operating Characteristic (ROC) curve methods to assess the sensitivities and specificities of PPG and LD, which account for the inherent trade-off between sensitivity and specificity of the tests by determining the appropriate boundary value for each method [25-27]. The vertical lines in Figs. 19a and 19b represent the optimal PPG and LD ratios of 0.74 and 0.65, respectively, for distinguishing neuropathic from non-neuropathic diabetes using LMS as shown in Table 2. If the vertical line in Fig. 19a moves to the right, the boundary value of the blood volume change ratio increases. At the same time, the sensitivity decreases and the specificity increases

since the blood volume change ratio of neuropathic diabetes is larger than that of non-neuropathic diabetes.

Table 3 shows the calculated sensitivities and specificities with the corresponding optimal ratios of PPG and LD methods for the Bayesian, LMS, and ROC curve methods. The LD method showed better sensitivity and specificity than the PPG for all three methods. In clinical applications, the higher the sensitivity of a test, the better the diagnosis. Therefore, the lowest boundary value of 0.65 is considered optimal for LD, and thus the corresponding sensitivity and specificity are 93.0 and 91.4%. The PPG method by LMS has a sensitivity of 86.0% and specificity of 82.8%. While different tests for neuropathy have low correlations among themselves [24, 28], both of our proposed methods demonstrated satisfactory sensitivities and specificities. This may be due to normalization with the toe to finger ratio, which minimizes the variation of absolute blood flow between subjects and the influence of skin temperature.

Table 3. Sensitivity, specificity, and boundary values calculated by the Bayesian, LMS, and ROC methods for both PPG and LD.

Method		Sensitivity (%)	Specificity (%)	Boundary value
PPG	Bayesian	79.0	89.0	0.79
	LMS	86.0	82.8	0.74
	ROC	84.0	84.3	0.75
LD	Bayesian	81.0	97.1	0.75
	LMS	93.0	91.4	0.65
	ROC	92.0	92.8	0.66

IV. DISCUSSION

The toe to finger ratios of the neuropathic group by LD was increased by a decrease in finger blood perfusion but not by an increase in toe blood perfusion as shown in Figs 18c and 18d. When we compared our results to those from a previous study (http://www.your-feet.com/pages/diabetic.aspx), the toe blood flow by LD in diabetic neuropathic patients did not differ significantly from blood flow in controls and non-neuropathic diabetic patients. This result is in accordance with our measured toe blood flow using LD. However, other studies have shown conflicting results. In these studies, the feet of diabetic patients with neuropathy showed increased skin blood flow when compared with those of diabetic patients without neuropathy and control subjects [18]. These results are in accordance with our results from measuring toe blood flow using PPG.

Wigington showed that finger blood flow in patients with diabetes was lower than in those without it [19], a result similar to ours using PPG. Though our proposed ratio method cannot discriminate between finger and toe neuropathy, it has the potential to become an effective, new screening tool for the detection of diabetic neuropathy as it most commonly affects feet before hands [19, 29].

Another advantage to our proposed method is that toe to finger ratios with a diabetic neuropathic foot would be increased either by decreased finger blood

volume coupled with increased toe blood volume as seen with our PPG method, or by decreased finger blood perfusion with unchanged toe blood perfusion like that seen with our LD.

The LD and PPG methods are different both in principle and in region of measurement. The LD used in this study is a reflected mode and the distance between the transmitting and receiving fibers is only 0.25 mm, and thus the measuring volume or depth (0.5-1 mm) is very small. Conversely, PPG is a transmitted mode and the distance between the transmitting and receiving transducers is the depth of the finger or toe being measured, and thus its measuring volume is considerably larger than that of LD. Therefore, both values cannot and should not be the same, although they may show the same trend in part, as evidenced by our results. In this study, we used LD in order to indirectly support the validity of the PPG measurements because LD has previously been used in many studies.

V. CONCLUSION

One of the most important findings in this study is that the blood volume change ratio of toe to finger may distinguish neuropathic diabetes from non-neuropathic diabetes with a high sensitivity and specificity. The suggested PPG method using this ratio provided a sensitivity of 86.0%, a specificity of 82.8%, and a mean reproducibility of 5.5%, while the LD showed a higher sensitivity of 93.0% and a higher specificity of 91.4%, but a lower mean reproducibility of 9.5%. While the LD method is superior in its sensitivity and specificity, it is expensive, complex, and has relatively poor reproducibility compared with that of PPG. The PPG method also showed proportionally increased ratios with neuropathic severity while the LD did not. The suggested PPG system has proven to be highly reproducible, simple, economical, and accurate and has opened up the possibility for its use as a new effective screening tool for the early detection of diabetic neuropathy.

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ABSTRACT (IN KOREAN)

레이저 도플러와 광혈류 측정법을 이용한
당뇨병성 신경병증의 새로운 진단법 개발

<지도교수 김 덕 원>

연세대학교 대학원 생체공학협동과정

김 성 우

본 연구의 목적은 당뇨병성 신경병증을 비침습적인 방법으로 사전에 간편하게 스크리닝하는 새로운 진단 방법을 제안하는 것이다. 임상에서 혈중 산소포화도 실시간 모니터링, 심혈관계 특성 진단 및 분석에 널리 사용되고 있는 Photoplethysmography(PPG)를 기반으로 인체의 말초 부위인 검지 손가락과 엄지 발가락의 혈류 및 혈류량 변화를 측정하였다. PPG 측정은 비침습적인 혈류 계측 방법으로서 인체에 전혀 고통을 주지 않으며, 기존의 여러 검사법 중 신경 전도 검사에 비해 안전하고 간략한 측정방법과 절차로부터 신속하게 당뇨병성 신경병증을 사전 진단할 수 있다는 장점이 있다. 객관적인 실험데이터 분석을 위해

PPG방식과 Laser Doppler방식을 동시에 이용하여 피험자 총 125명의 좌/우측 검지 손가락과 엄지 발가락 혈류데이터(N=250)를 수집하였고 첫 번째 피험자 그룹은 당뇨병 진단을 받지 않은 정상인 40명, 두 번째 피험자 그룹은 당뇨병 진단을 받았으나 신경병증이 없는 환자 35명, 마지막 피험자 그룹은 신경병증이 확진 된 당뇨병 환자 50명이었다. 이 당뇨병성 신경병증 환자그룹 50명은 신촌세브란스병원 당뇨병센터에서 신경전도검사를 통해 각 Mild 19명, Moderate 17명, Severe 14명으로 신경병증 중증도가 구분되었고 모든 피험자 모집과 연구 프로토콜은 해당병원 산하의 연세의료원 연구윤리위원회 IRB승인 아래 이루어졌다.

본 연구에서 진행된 분석 결과로부터 당뇨병성 신경병증 조기진단을 위한 최적의 손가락과 발가락 혈류비를 도출할 수 있었으며, 당뇨병성 신경병증의 선별검사로서 기존 검사의 단점을 보완할 수 있는 새로운 스크리닝 방법의 가능성을 확인하였다. 개발된 PPG 측정 시스템 및 상용화된 Laser Doppler 측정 방법에서 당뇨병성 신경병증 환자그룹의 혈류비가 각각 정상인 그룹, 일반 당뇨병 환자그룹 보다 통계적으로 유의하게 높았고($p<0.001$), 이는 선행 연구에서도 기 검증된 결과로서 유사하게 나타났다. 당뇨병성 신경병증 판별을 위한 최적의 혈류비는

PPG 방법에서 0.74, Laser Doppler 방법에서 0.65로 결정되었으며, 추가적으로 측정된 손가락-발가락 온도 차이가 4.5℃ 이하인 경우에 이것은 정상인 보다 신경병증 환자의 발가락 혈류량이 상대적으로 증가되어 나타나는 현상으로서 온도 차이 분포의 경계 값은 4.5℃로 결정되었다. PPG와 Laser Doppler 방법에서 결정된 최적의 혈류비를 기반으로 당뇨병성 신경병증의 새로운 스크리닝 지표에 대해 민감도, 특이도, 재현성을 검증하였고 최소평균제곱법(Least Mean Square)으로부터 각각 Laser Doppler방법은 민감도 및 특이도 93.0%, 91.4% PPG방법은 민감도 및 특이도 86.0%, 82.8%이고 수신자동작특성법(Receiver Operating Characteristic)으로부터 Laser Doppler방법은 민감도 및 특이도 92.0%, 92.8% PPG방법은 민감도 및 특이도 84.0%, 84.3%로 나타났다. 전체적인 성능 측면에서 기존 연구와 비교했을 때 각 성능이 우수하게 나타났으며 Laser Doppler방법이 PPG방법에 비해 전반적으로 민감도와 특이도는 높지만, 재현성은 PPG방법이 5.5%로서 약 1.7배 우수하고 진단에 필요한 신뢰도, 시간절약, 경제성측면에서는 PPG방법이 유리하다. 뿐만 아니라 당뇨병성 신경병증의 중증도에 따라 PPG방법에서는 통계적으로 유의한 구분이 가능함을 확인하였다.

이처럼 비침습적인 혈류 측정방법을 이용해 최적의 손가락과 발가락 혈류비를 도출하여 당뇨병성 신경병증을 선별 할 수 있었고, 신경병증 확진을 위한 신경전도검사 이전에 간편한 사전 검사로서 당뇨병성 신경병증의 조기진단을 위한 새로운 진단법을 제시하면서 임상에서의 진단 비용절감 및 시간절약 효과를 기대하며 국내외 당뇨병성 신경병증으로 발생하는 족부병변에 의한 절단을 예방하는데 기여하기를 바란다.

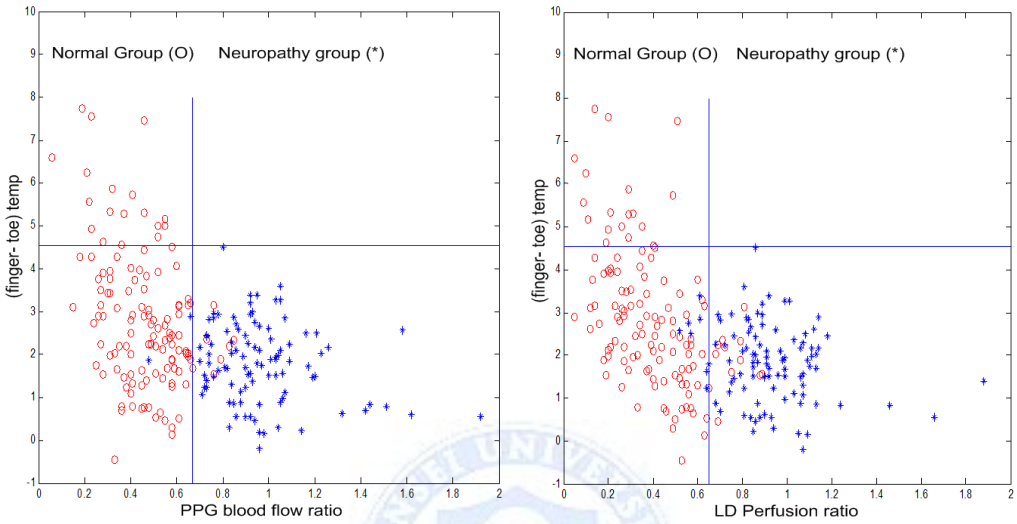
핵심되는 말 : 당뇨병성 신경병증, 광혈류 측정법, 레이저 도플러, 혈류량 변화, 발가락/손가락 혈류비

APPENDIX

APPENDIX I. ABBREVIATIONS

ADA :	American Diabetes Association
AVS :	Arteriovenous shunt
BMI :	Body mass index
GfK :	Growth from Knowledge
IDF :	International Diabetes Federation
KCDC :	Korea Centers for Disease Control and Prevention
KNHANES :	Korea National Health and Nutritional Examination Survey
LD :	Laser Doppler
LMS :	Least mean square
NCV :	Nerve conduction velocity
PPG :	Photoplethysmography
ROC :	Receiver Operating Characteristic
WHO :	World Health Organization

APPENDIX II. THE RESULTS OF THE PRELIMINARY STUDY



(a) (b)

Fig.A1. PPG blood flow ratio (a) and LD perfusion ratio (b) with temperature differences between fingers and toes by the ROC method (n = 128 for control, n = 100 for neuropathy).

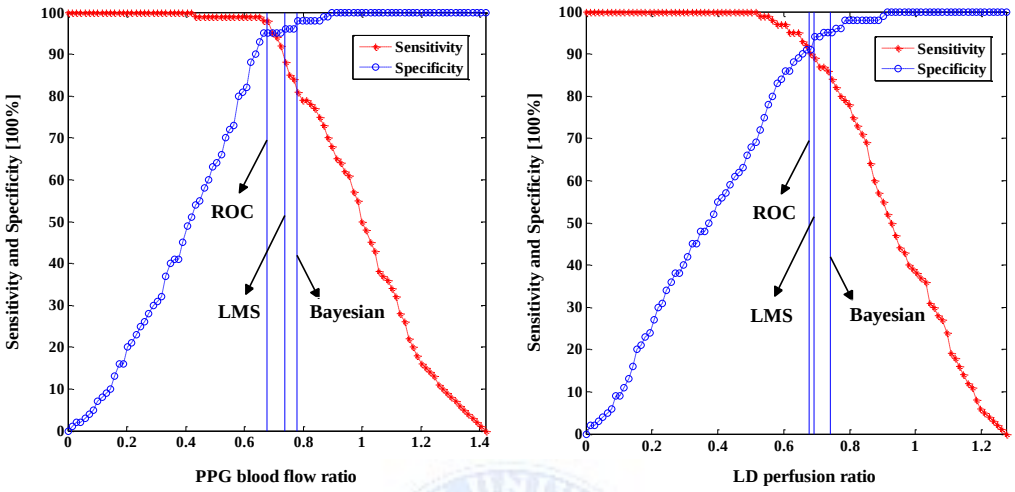


Fig.A2. The optimal ratio of PPG blood flow (a) and LD perfusion (b). These are explained by selecting a superior performance among the Bayesian, LMS, and ROC method (n = 128 for control, n = 100 for neuropathy).

Table.A1. Sensitivity, specificity, and boundary values calculated by the Bayesian, LMS, and ROC methods for both PPG and LD in a preliminary study.

Method		Sensitivity (%)	Specificity (%)	Boundary value
PPG	Bayesian	82.0	97.7	0.78
	LMS	88.0	96.1	0.74
	ROC	95.3	95.3	0.68
LD	Bayesian	84.0	95.3	0.74
	LMS	89.0	93.8	0.69
	ROC	92.0	91.4	0.68

APPENDIX III. ARTERIOVENOUS SHUNT AND SYMPATHETIC NERVE

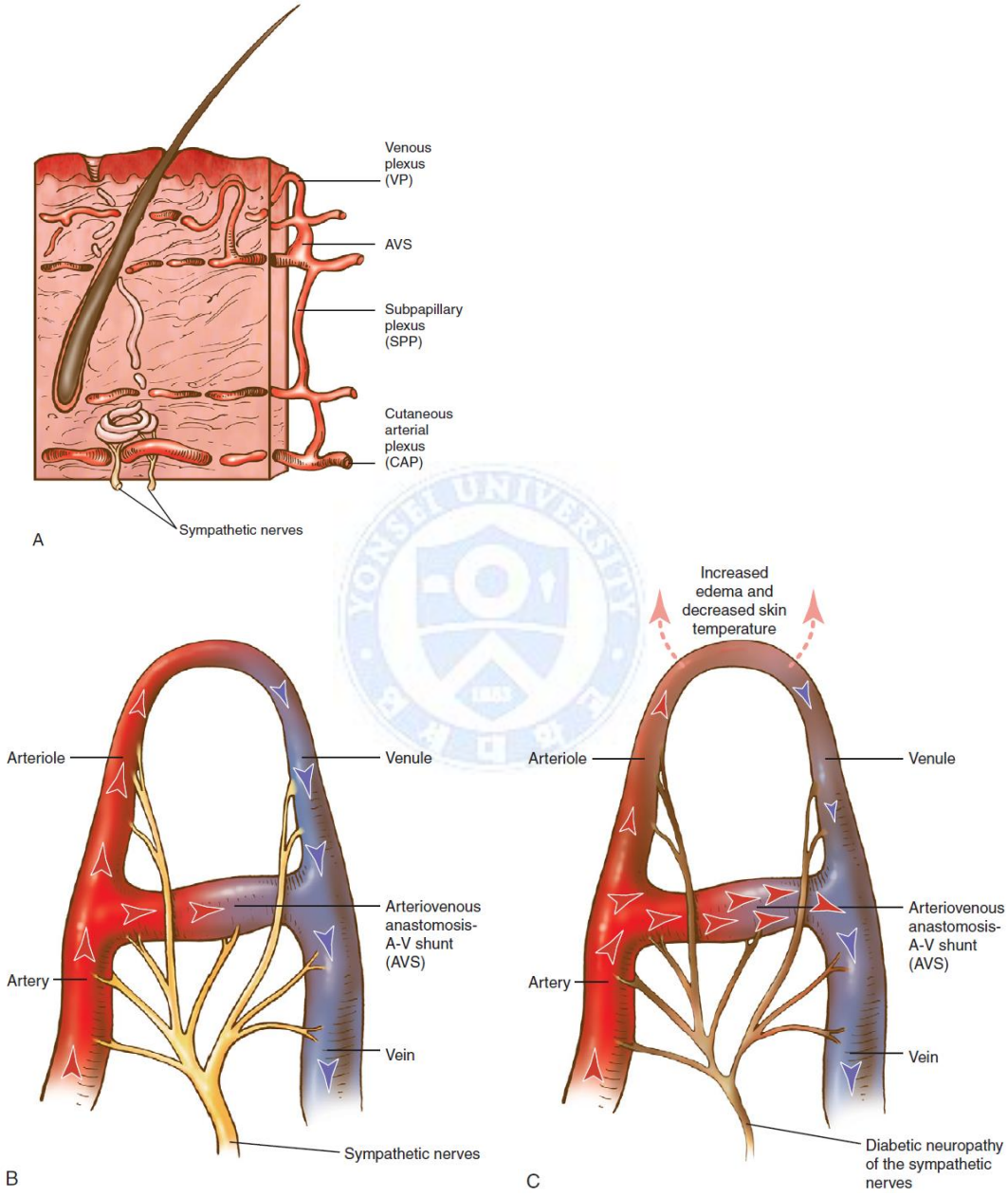


Fig.A3. Plantar AV shunts in normal and neuropathic diabetic individuals.