Pulp Blood Flow Velocity Analysis Using Ultrasound Doppler Flowmetry in Anterior Teeth And Intra- and Inter-examiner Reliability

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Pulp Blood Flow Velocity Analysis Using Ultrasound Doppler Flowmetry in Anterior Teeth And Intra- and Inter-examiner Reliability

Directed by Professor Sung-Ho Park

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감사의 글

2012년 3월, 두근거리는 마음으로 내디뎠던 보존과로의 첫 발걸음과 함께한 대학원 과정이, 2016년 6월, 어느덧 학위논문과 함께 막바지에 다다랐습니다. 그 동안 도움을 주신 모든 분들께 짧으나마 이 감사의 글로 마음을 전하고자 합니다.

먼저, 가장 가까운 곳에서 항상 든든하게 저를 응원해주시고 지켜봐 주신 아버지, 어머니께 감사 드립니다. 수련 받는 동안, 대학원 과정을 밟아 나가는 동안, 그리고 지금까지도 항상 걱정해주시는 부모님의 마음이 느껴질 때마다 마음 한 켠이 편안해지고 따듯해집니다. 말로는 잘 표현하지 못하는 아들이지만 항상 감사하고 사랑합니다.

대학원 과정 4년간 그리고 연세대학교 치과대학병원 보존과 3년의 수련 과정 동안 지도와 조언을 아끼지 않으신, 이 논문이 완성되기까지 부족한 제자의 시시콜콜한 질문에도 항상 웃으며 해결의 길을 알려주신 지도교수 박성호 교수님께 감사 드립니다. 논문 심사를 위해 여행 일정 전에 논문을 읽고 미리 조언해주신 이승종 교수님, 무심한 듯하지만 부족한 점을 정확히 찍어 알려주시고 도와주신 정일영 교수님, 해외 학회 참석에도 불구하고 조언과 격려를 아끼지 않으신 통합진료과 김기덕 교수님, 종료 심사 날까지도 세세하고 꼼꼼하게 관심 가져주신 치과생체재료공학교실 김광만 교수님께도 감사 드립니다.

4년의 대학원 과정 내에 수련 과정이 모두 겹쳐있기에, 수련 과정이 곧 대학원 과정이었습니다. 보존과 수련 과정 동안 때로는 엄하게, 때로는 인자하게 이끌어 주신 이찬영 교수님, 노병덕 교수님, 김의성 교수님, 신유석 교수님, 이 모든 분께 감사 드립니다.

마지막으로 함께 의국 생활하며 힘든 일, 즐거운 일 함께 겪고 지금은 모두 행복한 미래를 향해 열심히 살고 있는 내 수련 동기들 - 강민지, 김건태, 장선미, 정건석에게, 그리고 보존과 선후배들에게 그 동안 미처 말하지 못한 감사의 마음을 전합니다.

2016년 6월

조 용 욱

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Abstract

Pulp Blood Flow Velocity Analysis Using Ultrasound Doppler Flowmetry in Anterior Teeth And Intra- and Inter-examiner Reliability

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1. Objectives

Clinical diagnosis of pulpal status is difficult and occasionally confusing. A thermal (cold or hot) test and electric pulp test (EPT) are commonly used to evaluate pulpal nerve sensibility. However, these tests say nothing about pulp blood flow, which means true pulp vitality. The aims of this study were as follows: 1) to measure pulp blood flow velocities of clinically normal maxillary anterior teeth of healthy adults using ultrasound Doppler flowmetry (UDF), 2) and to evaluate intra- and inter-examiner reliability of UDF.

2. Materials and Methods

1) Part 1: Measurement of pulp blood flow velocities in maxillary anterior teeth

A total of 359 anterior teeth from 62 patients (mean age, 29.8 years; range, 22 to 52 years; 26 females and 36 males) were included. The data were collected according to tooth type (three groups: central incisors, lateral incisors, and canines). An MM-D-K (Minimax, Moscow, Russia) ultrasound Doppler imaging instrument with a 20-MHz probe was used to measure pulp blood flow velocity. Differences between the tooth types were analysed with one-way ANOVA and a Bonferroni correction at the 95% confidence level.

2) Part 2: Intra-examiner and inter-examiner reliability

A total of 222 anterior teeth from 37 patients (mean age, 25.4 years; range 21 to 32 years; 13 females and 24 males) were included. An MM-D-K (Minimax, Moscow, Russia) ultrasound Doppler imaging instrument with a 20-MHz probe was used to measure pulp blood flow velocity. For the intra-examiner reliability, one examiner measured the patients at about 1 week intervals for three weeks. For the inter-examiner reliability, the blood flows of patients were measured by five examiners using the UDF. Data were analysed with a repeated measure ANOVA (RM_ANOVA) and Tukey as a post hoc test at the 95% confidence level. Intraclass correlation coefficients (ICCs) were calculated from the data for intra- and inter-examiner reliability.

3. Results

1) Part 1: Measurement of pulp blood flow velocities in maxillary anterior teeth

The mean average linear velocities during the systolic period (Vams) of the central incisors, lateral incisors, and canines were 0.58 cm/s, 0.58 cm/s, and 0.52 cm/s, respectively. There were no significant differences in the mean Vams between the tooth types (*P* >0.05).

2) Part 2: Intra-examiner and inter-examiner reliability

Intra-examiner reliability: There were no significant differences in the Vam measurements among three time points (*P*>0.05), and ICC was 0.56.

Inter-examiner reliability: There were significant differences in the average linear velocities during the systolic period (Vam) measurements among the five examiners (*P*=0.044), and ICC was 0.41.

4. Conclusions

Within the limitations of this study, the pulp blood flow velocities of clinically normal, maxillary anterior teeth of healthy adults were 0.56 ± 0.46 cm/s. There were no significant differences in mean blood flow velocities between maxillary central incisors, lateral incisors, and canines.

Intra-examiner consistency demonstrated no significant differences among the measured time points and moderate to low reproducibility. Inter-examiner consistency demonstrated statistically significant differences among examiners and a low reliability. The methods which can reduce the variations between different examiners should be investigated in the near future.

Key words: pulp blood flow velocity; pulp vitality; ultrasound Doppler; intra-examiner reliability; inter-examiner reliability

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I. INTRODUCTION

Several methods are used to assess the status of the pulp, including thermal (cold or hot) test (Gopikrishna et al. 2007), electric pulp test (EPT) (Gopikrishna et al. 2007, Karayilmaz & Kirzioglu 2011), laser Doppler flowmetry (LDF) (Kimura et al. 2000, Karayilmaz & Kirzioglu 2011), ultrasound Doppler flowmetry (UDF) (Thierfelder et al. 1978, Chandler 1988, Yoon et al. 2010, 2012), and pulse oximetry (Radhakrishnan et al.

2002, Jafarzadeh & Rosenberg 2009, Karayilmaz & Kirzioglu 2011). Although thermal test and EPT are commonly used, these two methods are indirect methods of assessing the sensibility of the pulp nerve. These tests rely on subjective responses of patients and have low reliability when sensibility is affected by extrinsic factors (Gopikrishna et al. 2007, Yoon et al. 2010). Evaluation of pulp blood supply is more reliable for direct pulp vitality testing (Fratkin et al. 1999, Radhakrishnan et al. 2002, Jafarzadeh & Rosenberg 2009), particularly in teeth with immature root apices or following temporary or permanent loss of sensory function due to trauma or orthognathic surgery (Radhakrishnan et al. 2002, Jafarzadeh & Rosenberg 2009, Yoon et al. 2010).

Experimental trials to evaluate pulp blood flow have been conducted. Methods such as radioisotope clearance tests (Hock et al. 1980, Kim et al. 1983) and hydrogen gas desaturation tests (Aukland et al. 1964, Tonder & Aukland 1975) are clinically unacceptable due to their invasiveness. LDF and pulse oximetry are non-invasive, quantitative, and objective because they are not painful, easily accepted by sensitive patients after trauma, and they do not rely on patient responses. (Radhakrishnan et al. 2002, Emshoff et al. 2004, Strobl et al. 2004).

Among the various methods above mentioned, LDF has been widely researched and is considered reliable, but it may be affected by the optical properties of pulp chamber filling material and clinical technique due to the sensitivity of the laser beam used (Odor et al. 1996a, 1996b). For instance, in situations in which the pulp chamber is filled with necrotic debris (untreated) or is filled with gutta-percha or composite resin (root canal treated), the same negative recordings must be obtained if an instrument accurately detects pulp blood flow because there is no vital tissue in either case. However, the results of laser Doppler flowmetry will be different due to differences in the optical properties of the crown (Jafarzadeh 2009). LDF is also sensitive to the position, holding method of the probe (Cotti et al. 2003, Rajendran & Sundaresan 2007) and noise and vibration from the external environment (Vongsavan & Matthews 1993).

UDF has been used in the medical field as a non-invasive and radiation-free technique for the assessment of blood flow in micro-vascular systems. UDF shares the same Doppler principle with LDF. The difference between UDF and LDF is the source; LDF is an optical measurement method that uses a laser beam, whereas UDF is a physical measurement method that uses ultrasound. For all waves, e.g. sound, light, laser, ultrasound, there is change in the frequency of wave when it is reflected from a moving object. This effect is known as the 'Doppler effect' or 'Doppler shift'(White 1982). This change in frequency (f_d, known as Doppler frequency) is proportional to the velocity of the moving object:

$$f_d = (2\ f_0\ v \cos\quad)\ c^{-1} \qquad \cdots\cdots\cdots\cdots\cdots\cdots \text{Equation 1}$$

, where f_0 is the transmitted frequency, v is the velocity of the moving object,   is the angle between the movement vector and the transmitted wave, and c is the velocity of the wave in the matrix.

In the UDF, when the ultrasound probe emits the wave to a moving red blood cell, the cell reflects the incident wave. Therefore, the frequency of the reflected wave is altered according to the Doppler principle. This frequency shift is detected and analysed by the flowmetry machine and the flow velocity of RBC—blood flow velocity—can be calculated from above equation. The monitor of UDF machine shows real-time wave patterns within given time periods, and the machine calculates the blood flow velocity, pulsation index, and circulation index, and, simultaneously, the examiner can listen to pulsation sounds in real time.

Several studies have shown that ultrasound Doppler can be successfully used in the differential diagnosis of periapical granulomas and cysts based on the detection of the micro-vascularity of the lesion (Cotti et al. 2003) and can be used in follow-up evaluations of the healing of periapical lesions after root canal treatment (Rajendran & Sundaresan 2007). The authors of these studies concluded that UDF offers sufficient information about micro-vascularity. Recently, Yoon et al. (2010) evaluated blood flow velocity in root-filled teeth and contralateral teeth with healthy pulp using UDF and illustrated the potential of examining pulp blood flow velocity with UDF (Yoon et al. 2010). Additionally, Yoon et al. (2012) reported that blood flow velocity changes before and after infiltration anaesthesia can be measured using UDF (Yoon et al. 2012).

There has been relatively little research about the reproducibility of ultrasound Doppler in the medical field. In a study of the ultrasound Doppler velocimeter in the examination of vertebral artery blood flow velocity (Thomas et al. 2009), the authors concluded that the inter-examiner reliability was poor (kappa ranged from 0.15 to 0.26). Also, in another study of intra- and inter-observer variability of basal flow velocity using transcranial Doppler ultrasonography, there was a reproducibility when the measurements were repeated by the same examiner (high intra-examiner reliability), but this was more variable when detected by different investigators (low inter-examiner reliability) (Ceravolo et al. 1992).

For the clinical application of UDF, the pulp blood flow velocities of teeth with healthy pulp measured by UDF is required. And the intra- and inter-examiner consistencies are important indicators for evaluating the reliability of newly developed medical devices such as UDF. Therefore, the purpose of this article were as follows: 1) to measure the pulp blood flow velocities of clinically normal, maxillary anterior teeth of healthy adults using UDF, 2) to evaluate intra- and inter-examiner reliability of UDF.

II. MATERIALS AND METHODS

Part 1: Measurement of pulp blood flow velocities in maxillary anterior teeth

1. Subjects

A total of 359 anterior teeth from 62 patients (mean age, 29.8 years; range 22 to 52 years; 26 females and 36 males) were included. The data were collected according to tooth type (three groups: central incisors, lateral incisors, canines). Patients with systemic cardiovascular disease were excluded. The inclusion criteria for the teeth were as follows: 1) no caries or restoration, 2) no history of orthodontic treatment or trauma, 3) clinically normal responses to thermal and EPT (Gentle Pulse Analog Pulp Vitality Tester, Parkell Inc., NY, USA), no sensitive response to percussion on, and tooth mobility within the normal range, and 4) normal periradicular state on a periapical radiograph. This study was approved by the Ethics Committee of the dental hospital at Yonsei University (2-2010-0002).

2. Apparatus

An MM-D-K (Minimax Ltd., St. Petersburg, Russia) ultrasound Doppler imaging instrument (Figure 1) with a 20-MHz probe was used to measure pulp blood flow velocities. The tip diameter of the ultrasound probe was approximately 2.5 mm, and the entire length of the probe was approximately 20 cm (Figure 2). An ultrasound gel (Pro-gel II: Dayo Medical, Seoul, Korea) was applied as a coupling agent. When the measurement was carried out, Doppler spectrums were recorded and flow parameters were calculated using Minimax Doppler 1.71 (Minimax Ltd., St. Petersburg, Russia) software program (Figure 3).

Figure 1. Doppler ultrasound system (MM-D-K: Minimax Ltd., St. Petersburg, Russia).

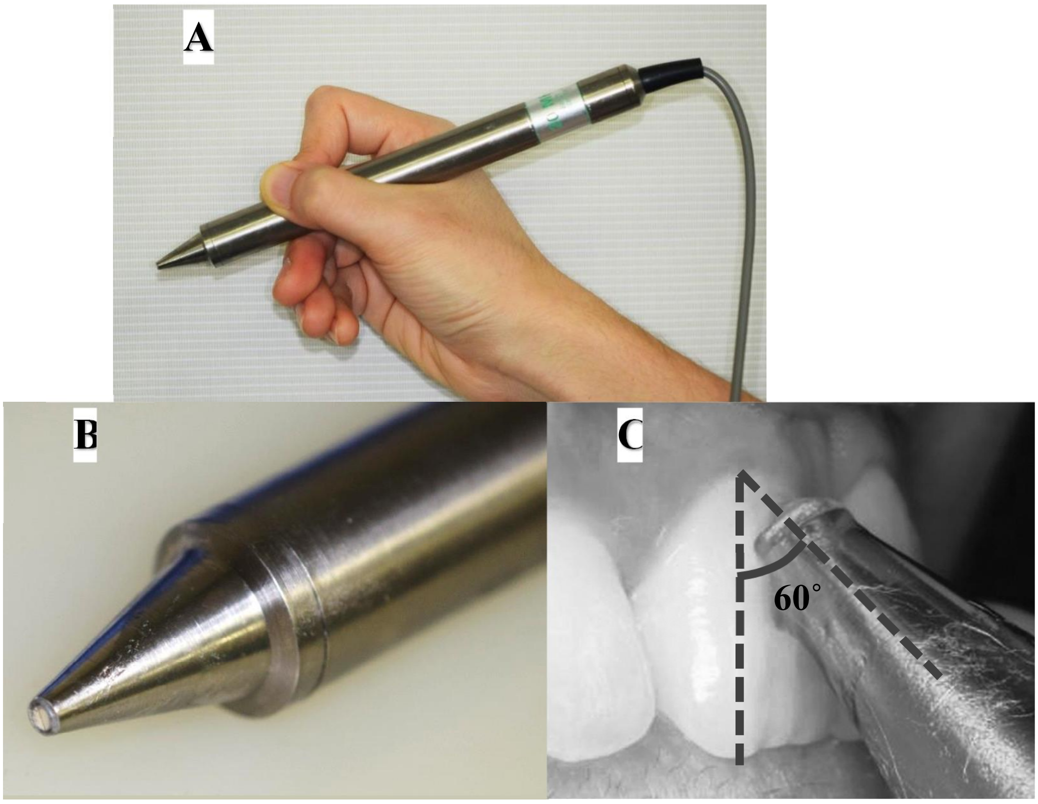

Figure. 2. Ultrasonic probe of the Minimax Doppler machine

A. Ultrasonic probe held in hand

B. Tip of the ultrasonic probe

C. Measurement position

3. Procedures

All procedures were performed by a single examiner who was well educated and skilled for measuring the pulp blood flow velocity using UDF to reduce inter-examiner errors. Each patient was placed in a semi-supine position and allowed to rest for more than 10 minutes prior to the measurements to ensure relaxation. After drying the tooth surface, the ultrasound gel was applied to the cervical area of the tooth, and a probe was placed at a 60-degree angle to the labial surface (Fig. 2C). Because the examiner could hear the pulsating sounds in real time, the probe was hand-held at the site of maximal pulsation. The probe position was fixed at the site of the most clear pulsating sound for 8 seconds so that the wave pattern was recorded clearly.

4. UDF parameters for pulp blood flow measurement

The measurement parameters are listed in Table 1. Under these parameters, the linear velocities (Vs, Vam, and Vd) are more meaningful than the volume velocities (Qs and Qam). Volume velocities, which are determined by the cardiac output volume, are similar regardless of vessel diameter or type (e.g., these velocities are similar between the aorta, the arterioles and the capillaries) (Riva et al. 1985, Jamieson et al. 2014).

By definition, the maximum linear velocity during the systolic period (Vs) and the minimum linear velocity during the diastolic period (Vd) are more strongly affected by blood pressure than are the average velocity values. Thus, in the present study, the average linear velocities during the systolic period (Vam) were analysed. All these velocity paratmeters were calculated from the spectrum of Minimax Doppler 1.71(Minimax Ltd., St. Petersburg, Russia) software.

The pulsation index (PI) reflects the elastic properties of blood vessels; PI = (Vs – Vd) / Vam, and resistance index (RI) reflects the state of resistance to blood flow distal to the measure site; RI = (Vs – Vd) / Vs.

Table 1. Ultrasound Doppler flowmetry parameters

Parameter	Definition
Vs (cm/s)	Maximum linear velocity during the systolic period
Vam (cm/s)	Average linear velocity during the systolic period
Vd (cm/s)	Minimum linear velocity during the diastolic period
Qs (ml/min)	Maximum volume velocity during the systolic period
Qam (ml/min)	Average volume velocity during the systolic period
PI	Pulsation index: resilience of the blood vessel
RI	Resistance index: circulation resistance

5. Statistical Analyses

The raw data were statistically analysed using SPSS v23.0 software (IBM Corp., Somers, NY, USA). The Vam values of the central incisors, lateral incisors, and canines were compared with a one-way ANOVA and a Bonferroni correction at the 95% confidence level.

Part 2: Intra-examiner and inter-examiner reliability

1. Subjects

A total of 222 anterior teeth from 37 patients (mean age, 25.4 years; range 21 to 32 years; 13 females and 24 males) were included. The data were collected regardless of tooth type (central incisors, lateral incisors, or canines). Patients with systemic cardiovascular disease were excluded. The inclusion criteria for the tooth were as follows: 1) no caries or restoration; 2) no history of orthodontic treatment or trauma; 3) clinically normal responses to thermal test and EPT (Gentle Pulse Analog Pulp Vitality Tester, Parkell Inc., NY, USA); 4) no sensitive response to percussion on the teeth, 5) teeth mobility within the normal range; and 6) normal periradicular state on a periapical radiograph. This study was approved by the Ethics Committee of the dental hospital at Yonsei University (2-2010-0002).

2. Apparatus

As in Part 1 study, the same apparatus (MM-D-K: Minimax Ltd., St. Petersburg, Russia) and ultrasound gel (Pro-gel II: Dayo Medical, Seoul, Korea), same software (Minimax Doppler 1.71: Minimax Ltd., St. Petersburg, Russia) were used.

3. Procedures

As in Part 1 study, the same measuring protocol was followed.

For the intra-examiner reliability, one examiner (YW) measured the blood flow velocities at about 1 week intervals for three weeks. For the inter-examiner reliability, the blood flow velocities of patients were measured by five examiners (SM, JH, KH, WY, SH) using the UDF. The sequence of examiners was randomly assigned, and all examiners were blinded from the recorded data which was measured by other examiners. At least a 10 minute interval was given between each measurement so the patient could relax. At each measuring, the above inclusion criteria were confirmed and the examiner was blinded from the previously measured data. All the above six examiners were in the resident course at the department of Conservative Dentistry in the dental hospital of Yonsei University and well educated and skilled for measuring the pulp blood flow velocity using UDF.

4. Statistical Analyses

The raw data were statistically analysed using SPSS v23.0 software (IBM Corp., Somers, NY, USA). The measured blood flow velocities were analysed with a repeated measure ANOVA (RM_ANOVA) and Tukey as a post hoc test at the 95% confidence level. Additionally, intraclass correlation coefficients (ICCs) were calculated from the data for intra- and inter-examiner reliability.

III. RESULTS

Part 1: Measurement of pulp blood flow velocities in maxillary anterior teeth

The mean average linear velocity during the systolic period (Vams) of the central incisor, lateral incisor, and canine were 0.58 cm/s, 0.58 cm/s, and 0.52 cm/s, respectively (Table 2). All teeth in this study exhibited pulsed waveforms and pulsating sounds that were characteristic of arterioles in teeth with healthy pulp (Figure 3). There were no significant differences in the mean Vams between the central incisors, lateral incisors, and canines (*P* >0.05).

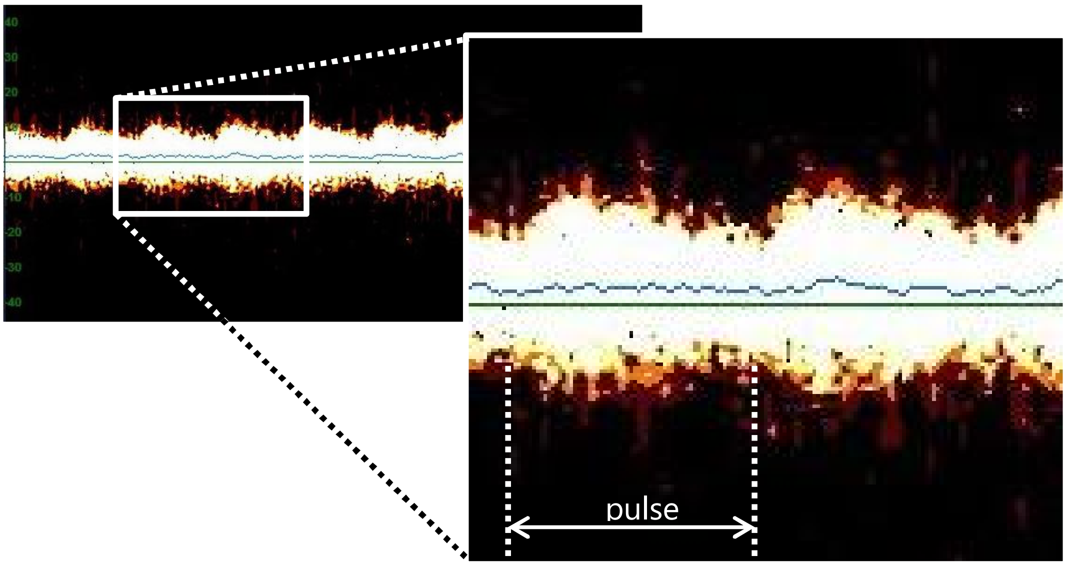

Figure 3. A Characteristic pulsed waveform of an arteriole in a vital tooth

Table 2. Pulp blood flow velocity measurements in maxillary anterior teeth

Tooth type	Sample size	Mean Vam ± SD (cm/s)
Central incisor	119	0.58 ± 0.49
Lateral incisor	120	0.58 ± 0.47
Canine	120	0.52 ± 0.41
Total	359	**0.56 ± 0.46**

Part 2: Intra-examiner and inter-examiner reliability

1. Intra-examiner reliability

There were no significant differences in the Vam measurements among three time points (P>0.05), and ICC was 0.56 (Table 4).

2. Inter-examiner reliability

There were significant differences in the average linear velocities during the systolic period (Vam) measurements among the five examiners (P=0.044) (Table 3), and ICC was 0.41. The measured data from examiner 3 (KH) were significantly higher than examiner 1 (SM, P=0.045) and examiner 2 (JH, P=0.003) (Table 3).

Table 3. The result of intra-examiner reliability

Measurement	Sample size	Mean Vam ± SD (cm/s)	P value of RM_ANOVA	ICC
1st		0.547 ± 0.363		
2nd	222	0.590 ± 0.242	0.603	0.56
3rd		0.611 ± 0.210		

Table 4. The result of inter-examiner reliability

Examiner	Sample size	Mean Vam ± SD (cm/s)	P value of RM_ANOVA	ICC
SM		0.495 ± 0.330 A		
JH		0.468 ± 0.306 A		
KH	222	0.561 ± 0.381 B	0.044	0.41
WY		0.525 ± 0.401 AB		
SH		0.509 ± 0.388 AB		

Means followed by different letters have significant difference according to Tukey's test (*P*<0.05)

IV. DISCUSSION

Laser Doppler flowmetry (LDF) requires a zero point setting and calibration using a Brownian motion medium prior to each measurement and demonstrates the relative blood flow velocity in perfusion units or flux units (Chen & Abbott 2011, Chen et al. 2012). In contrast, ultrasound Doppler flowmetry (UDF) does not require a zero point setting or calibration procedure; thus, UDF can reveal the absolute velocity value in cm/s or mm/s.

The MM-D-K (Minimax Ltd., St. Petersburg, Russia) is the only Doppler ultrasound machine that is clinically available for a dental use. In UDF, the quality of the acquired image and the penetration depth of the ultrasonic wave are influenced by the frequency of the probe (Berson et al. 1999).

Low-frequency probes (i.e., those that utilize frequencies below 10 MHz) provide sufficient penetration to depths exceeding 2 cm. However, low frequency probes create low quality images and cannot detect small vessels with low flow velocities (Berson et al. 1999). In contrast, high-frequency probes (i.e., probes that utilized frequencies above 50 MHz) provide high quality images and sensitive detection of small vessels with low flow velocities but only provide shallow penetration depths of less than 1 mm.

The penetration depth of ultrasonic wave are also affected by the acoustic impedance of the matrix which the wave passes through. The acoustic impedances of soft tissues are about 1.63 (10^6 kg / m^2·s) (Mast 2000), however, those of enamel and dentin are 18.8 and

7.6 (10^6 kg / m^2·s) each (Blodgett 2003). Thus, the versatility of high-frequency probes which utilized frequency above 40~50 MHz is much limited, and technical difficulties are associated with their use (Christopher et al. 1996, 1997).

A 20-MHz probe was used in the present studies. Although this frequency is known to produce images of limited resolution (80 ㎛ axially and 250 ㎛ laterally) (Berson et al. 1999), present studies aimed to measure pulp blood flow velocities and not to study images; thus, image resolution was not relevant to these studies. Twenty-megahertz probes can detect blood flows as deep as 0.8 cm and low flow velocities below 0.05 cm/s (Berson et al. 1999); thus, the 20-MHz probe was useful for the evaluation of small arterioles and venules in the oral-maxillofacial region. This frequency is also recommended for detecting vessels at 0.1 – 8mm depth, according to the manufacturer's instruction.

Especially the MM-D-K machine was fabricated not to fit only on a soft tissue, but also on dental hard tissues; enamel and dentin. The matrix of measuring target (soft tissue, or dental hard tissue) was selected prior to the measuring. The acoustic impedance of the matrix was considered and calculated in the Minimax Doppler 1.71 software automatically.

Generally, blood flow velocities were greater in vessels with larger diameters. In a study of blood flow velocities that utilized triplex ultrasonography, the mean blood flow velocity of the common carotid artery, which has a diameter of 5.4 – 7.5 mm, was found to be 48.2 cm/s (minimum 29.0 cm/s, maximum 67.0 cm/s), while the mean blood flow velocity of the vertebral artery, which has a diameter of 2.3 – 3.8 mm, was found to be 31.4 cm/s (minimum 17.4 cm/s, maximum 45.4 cm/s) (Owolabi et al. 2014). Moreover, in another study that used a duplex colour Doppler imager, the authors concluded that there was a positive and linear correlation between vessel diameter and blood flow velocity (Stoner et al. 2004).

Studies of the blood flow velocities in relatively small–diameter arterioles have been performed in ophthalmology. As measured using laser blood flowmeter, the mean blood flow velocity of retinal arterioles, which has a diameter of 85 – 129 ㎛, has been found to be 2.2 – 4.2 cm/s (Gilmore et al. 2005), and the mean blood flow velocity of conjunctival arterioles, which has a diameter of 6 – 12 ㎛, has been found to be 0.052 – 0.326 cm/s based on high-speed video microcinematography (Koutsiaris et al. 2010).

The targets of the UDF measurements in the present studies were the pulp arterioles. The diameters of pulp arterioles have been reported to be 20 – 40 ㎛ (Okamura et al. 1994) and 14 – 51 ㎛ (Tomaszewska et al. 2013). Given the positive correlation between vessel diameter and flow velocity, it can be deduced that the blood flow velocities of the pulp arterioles, which are 10 – 50 ㎛ in diameter, were approximately 0.1 – 2.0 cm/s. In the present studies, the blood flow velocities ranged from 0.44 to 0.67 cm/s (95% confidence interval). These findings are consistent with results reported in previous studies of 0.27 – 0.79 cm/s (Yoon et al. 2010, 2012) and 0.18 – 0.56 cm/s (Qu et al. 2014).

In the part 1 study, there were no significant differences in the mean blood flow velocities of the maxillary central incisors, lateral incisors, and canines; however, the measured data had a relatively large standard deviation, which is consistent with other studies (Norer et al. 1999, Roy et al. 2008). Several predisposing factors, including vessel diameter and baseline blood pressure, might affect these values.

Vessel diameter and blood pressure might be the most critical factors that affect pulp blood flow velocity (Stoner et al. 2004, Owolabi et al. 2014). Vessel diameters are controlled by systemic and/or local factors of each patient and cannot be controlled by the examiner. Vessel diameters are also related to patient age, and the samples of these studies consisted of young patients with a mean age 29.8 years and 25.4 years, part 1 and 2 each. Blood pressure cannot be strictly controlled by the examiner, but the use of relaxation procedures prior to measurements can reduce blood pressure variation (Norer et al. 1999, Akpınar et al. 2004). In these studies, the patients were provided with a sufficient amount of rest that exceeded 10 minutes prior to each measurement and were placed in the semi-supine position during the measurement. Further studies of the relationship between blood pressure and blood flow velocity measured by UDF are required.

Probe angle, probe position, probe design, and the characteristics of the probe holder might also affect these results (Jafarzadeh 2009). These factors should be controlled and standardized in each study. In the part 1 study, one experienced examiner measured all of the subjects using the same probe and techniques to eliminate variations between examiners. Also, all six examiners (SM, JS, KH, WY, SH and YW) in the part 2 study were well trained to measure the pulp blood flow velocity using UDF.

However, the part 2 study showed that the inter-examiner consistency among five examiners (SM, JS, KH, WY and SH) had a significant difference (P=0.044) and the inter-examiner ICC value was low (0.41). There could be several explanations. The ultrasound probe always requires the coupling gel to be between the probe tip and the tooth surface for conduction of ultrasound. If the coupling gel came in contact with the soft tissue, the ultrasonic wave could have spread out and the reflecting wave from the soft tissue was recorded by the probe. Careless or excessive use of the coupling gel could make the result inaccurate.

In this situation, tooth isolation using a dental dam could be considered (Polat et al. 2005, Kijsamanmith et al. 2011). Another possible cause was the contact site of the probe tip on tooth surface. During the repeated measuring by several examiners, the contact site on tooth surface might be vary. In such situations, the use of an individualized repositioning resin stent with a probe tip-sized hole at the labial surface of the stent (Yoon et al. 2012) for each patient might help to reduce positioning variation due to the repetition of measurement by several examiners.

The probe angle is one of the crucial factors which affects to the result. According to the Equation 1, the calculated velocity ‘v’ is inversely proportional to the cosine value of α, where α is the probe angle. In this study, the probe angle was set to 60˚ between examiners. Due to the characteristic of cosine value, the calculated velocity might be exponentially increased if the probe angle was increased above 60˚

From a statistical perspective, examiners 1 (SM) and 2 (JH) were significantly different from examiners 3 (KH), 4 (WY), and 5 (SH) (Table 1). When the data from examiner 1 were excluded, calculated inter-examiner ICC could be increased to 0.43. In the same way, when the data from both examiners 1 and 2 were excluded, ICC could be increased up to 0.51, which is similar to intra-examiner ICC (0.56) (Table 5).

Table 5. Inter-examiner ICC changes by exclusion of significantly different examiner(s)

	Inter-examiner ICC
All examiners included	0.41
Examiner 1 (SM) excluded	0.43
Both examiner 1 (SM) and 2 (JH) excluded	0.51

As a clinician, the most useful application of UDF might be the longitudinal pulp vitality evaluation of traumatized or immature teeth. In these situations, a long-term monitoring is generally required for the diagnosis of pulp necrosis. According to the part 2 study, the intra-examiner consistency showed no significant difference (*P*>0.05) and the intra-examiner ICC value was moderate (0.56). A single clinician could observe the pulp vitality for a long period of time using UDF, but a cautious interpretation and comprehensive clinical examination must be followed. And also the use of an individualized repositioning resin stent with a probe tip-sized hole at the labial surface of the stent (Yoon et al. 2012) for each patient could be considered to reduce positioning variation due to the repetition of measurement during a longitudinal monitoring.

V. CONCLUSION

The pulp blood velocities of clinically normal, maxillary anterior teeth of healthy adults were 0.56 ± 0.46 cm/s. There were no significant differences in mean blood flow velocities between maxillary central incisors, lateral incisors, and canines.

Intra-examiner consistency demonstrated no significant differences among the measured time points and moderate reproducibility. Inter-examiner consistency demonstrated statistically significant differences among examiners and a low reliability.

The methods which can reduce the variations between different examiners — e.g. usage of individualized teeth splint with probe tip-sized hole, standardized measuring protocol training between examiners — should be investigated in the near future. Within the limitations of this study, it is expected that a single clinician could observed the pulp vitality for a longitudinal monitoring, however, a cautious interpretation and comprehensive clinical examination must be accompanied.

References

Akpınar KE, Er K, Polat S, Polat NT (2004) Effect of Gingiva on Laser Doppler Pulpal Blood Flow Measurements. Journal of Endodontics 30, 138-40.

Aukland K, Bower BF, Berliner RW (1964) Measurement of Local Blood Flow with Hydrogen Gas. Circulation Research 14, 164-87.

Berson M, Grégoire JM, Gens F et al. (1999) High frequency (20 MHz) ultrasonic devices: advantages and applications. European Journal of Ultrasound 10, 53-63.

Blodgett DW (2003) Applications of laser-based ultrasonics to the characterization of the internal structure of teeth. The Journal of the Acoustical Society of America 114(1), 542-9.

Ceravolo MG, Minciotti P, Orlandini M, Provinciali L (1992) Intra- and inter-observer variability of basal flow velocity and vascular reactivity measurements using transcranial Doppler sonography. Neurological research 14(Suppl. 2), 122-4.

Chandler NP (1988) Vitality testing of teeth using Doppler ultrasound. In: Price R, Evans JA, eds. Blood Flow Measurements in Clinical Diagnosis. pp. 190-2. London, UK: Biological Engineering Society.

Chen E, Abbott PV (2011) Evaluation of Accuracy, Reliability, and Repeatability of Five Dental Pulp Tests. Journal of Endodontics 37, 1619-23.

Chen E, Goonewardene M, Abbott P (2012) Monitoring dental pulp sensibility and blood flow in patients receiving mandibular orthognathic surgery. International Endodontic Journal 45, 215-23.

Christopher DA, Burns PN, Armstrong J, Foster FS (1996) A high-frequency continuous-wave Doppler ultrasound system for the detection of blood flow in the microcirculation. Ultrasound in Medicine & Biology 22, 1191-203.

Christopher DA, Burns PN, Starkoski BG, Foster FS (1997) A high-frequency pulsed-wave Doppler ultrasound system for the detection and imaging of blood flow in the microcirculation. Ultrasound in Medicine & Biology 23, 997-1015.

Cotti E, Campisi G, Ambu R, Dettori C (2003) Ultrasound real-time imaging in the differential diagnosis of periapical lesions. International Endodontic Journal 36, 556-63.

Emshoff R, Emshoff I, Moschen I, Strobl H (2004) Laser Doppler flow measurements of pulpal blood flow and severity of dental injury. International Endodontic Journal 37, 463-7.

Fratkin RD, Kenny DJ, Johnston DH (1999) Evaluation of a laser Doppler flowmeter to assess blood flow in human primary incisor teeth. Pediatric Dentistry 21, 53-6.

Gilmore ED, Hudson C, Preiss D, Fisher J (2005) Retinal arteriolar diameter, blood velocity, and blood flow response to an isocapnic hyperoxic provocation. American Journal of Physiology. Heart and Circulatory Physiology 288, H2912-17.

Gopikrishna V, Tinagupta K, Kandaswamy D (2007) Comparison of Electrical, Thermal, and Pulse Oximetry Methods for Assessing Pulp Vitality in Recently Traumatized Teeth. Journal of Endodontics 33, 531-5.

Hock J, Nuki K, Schlenker R, Hawks A (1980) Clearance rates of xenon-113 in non-inflamed and inflamed gingiva of dogs. Archives of Oral Biology 25, 445-9.

Jafarzadeh H (2009) Laser Doppler flowmetry in endodontics: a review. International Endodontic Journal 42, 476-90.

Jafarzadeh H, Rosenberg PA (2009) Pulse Oximetry: Review of a Potential Aid in Endodontic Diagnosis. Journal of Endodontics 35, 329-33.

Jamieson LH, Arys B, Low G, Bhargava R, Kumbla S, Jaremko JL (2014) Doppler Ultrasound Velocities and Resistive Indexes Immediately After Pediatric Liver Transplantation: Normal Ranges and Predictors of Failure. American Journal of Roentgenology 203, W110-16.

Karayilmaz H, Kirzioglu Z (2011) Comparison of the reliability of laser Doppler flowmetry, pulse oximetry and electric pulp tester in assessing the pulp vitality of human teeth. Journal of Oral Rehabilitation 38, 340-7.

Kijsamanmith K, Timpawat S, Vongsavan N, Matthews B (2011) A comparison between red and infrared light for recording pulpal blood flow from human anterior teeth with a laser Doppler flow meter. Archives of oral biology 56(6), 614-8.

Kim S, Schuessler G, Chien S (1983) Measurement of blood flow in the dental pulp of dogs with the 133xenon washout method. Archives of Oral Biology 28, 501-5.

Kimura Y, Wilder-Smith P, Matsumoto K (2000) Lasers in endodontics: a review. International Endodontic Journal 33, 173-85.

Koutsiaris AG, Tachmitzi SV, Papavasileiou P et al. (2010) Blood velocity pulse quantification in the human conjunctival pre-capillary arterioles. Microvascular Research 80, 202-8.

Mast TD (2000) Empirical relationships between acoustic parameters in human soft tissues. 1(2), 37-42.

Nissan R, Trope M, Zhang CD, Chance B (1992) Dual wavelength spectrophotometry as a diagnostic test of the pulp chamber contents. Oral surgery, oral medicine, and oral pathology 74(4), 508-14.

Norer B, Kranewitter R, Emshoff R (1999) Pulpal blood-flow characteristics of maxillary tooth morphotypes as assessed with laser Doppler flowmetry. Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology 87, 88-92.

Odor TM, Ford TR, McDonald F (1996a) Effect of probe design and bandwidth on laser Doppler readings from vital and root-filled teeth. Medical Engineering & Physics 18, 359-64.

Odor TM, Pitt Ford TR, McDonald F (1996b) Effect of wavelength and bandwidth on the clinical reliability of laser Doppler recordings. Endodontics & Dental Traumatology 12, 9-15.

Okamura K, Kobayashi I, Matsuo K et al. (1994) Ultrastructure of the neuromuscular junction of vasomotor nerves in the microvasculature of human dental pulp. Archives of Oral Biology 39, 171-6.

Owolabi MO, Agunloye AM, Ogunniyi A (2014) The relationship of flow velocities to vessel diameters differs between extracranial carotid and vertebral arteries of stroke patients. Journal of Clinical Ultrasound 42, 16-23.

Polat S, Er K, Polat NT (2005) The lamp effect of laser Doppler flowmetry on teeth. Journal of oral rehabilitation 32(11), 844-8.

Qu X, Ikawa M, Shimauchi H (2014) Improvement of the detection of human pulpal blood flow using a laser Doppler flowmeter modified for low flow velocity. Archives of Oral Biology 59, 199-206.

Radhakrishnan S, Munshi AK, Hegde AM (2002) Pulse oximetry: a diagnostic instrument in pulpal vitality testing. The Journal of Clinical Pediatric Dentistry 26, 141-5.

Rajendran N, Sundaresan B (2007) Efficacy of ultrasound and color power Doppler as a monitoring tool in the healing of endodontic periapical lesions. Journal of Endodontics 33, 181-6.

Riva CE, Grunwald JE, Sinclair SH, Petrig BL (1985) Blood velocity and volumetric flow rate in human retinal vessels. Investigative Ophthalmology & Visual Science 26, 1124-32.

Roy E, Alliot-Licht B, Dajean-Trutaud S, Fraysse C, Jean A, Armengol V (2008) Evaluation of the ability of laser Doppler flowmetry for the assessment of pulp vitality in general dental practice. Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology 106, 615-20.

Schnettler JM, Wallace JA (1991) Pulse oximetry as a diagnostic tool of pulpal vitality. Journal of endodontics 17(10), 488-90.

Stoner L, Sabatier M, Edge K, McCully K (2004) Relationship between blood velocity and conduit artery diameter and the effects of smoking on vascular responsiveness. Journal of Applied Physiology 96, 2139-45.

Strobl H, Emshoff I, Bertram S, Emshoff R (2004) Laser Doppler flow investigation of fractured permanent maxillary incisors. Journal of Oral Rehabilitation 31, 23-8.

Thierfelder C, Magnus S, Pardemann G, Vogel S (1978) [Functional vascular diagnostics of the dental pulp, a new application field for the ultrasonic Doppler procedure]. Das Deutsche Gesundheitswesen 33, 1105-9.

Thomas LC, Rivett DA, Bolton PS (2009) Validity of the Doppler velocimeter in examination of vertebral artery blood flow and its use in pre-manipulative screening of the neck. Manual therapy 14(5), 544-9.

Tomaszewska JM, Miskowiak B, Matthews-Brzozowska T, Wierzbicki P (2013) Characteristics of dental pulp in human upper first premolar teeth based on immunohistochemical and morphometric examinations. Folia Histochemica et Cytobiologica 51, 149-55.

Tonder KH, Aukland K (1975) Blood flow in the dental pulp in dogs measured by local

H2 gas desaturation technique. Archives of Oral Biology 20, 73-9.

Vongsavan N, Matthews B (1993) Some aspects of the use of laser Doppler flow meters for recording tissue blood flow. Experimental Physiology 78, 1-14.

White DN (1982) Johann Christian Doppler and his effect--a brief history. Ultrasound in medicine & biology 8(6), 583-91.

Yoon MJ, Kim E, Lee SJ, Bae YM, Kim S, Park SH (2010) Pulpal blood flow measurement with ultrasound Doppler imaging. Journal of Endodontics 36, 419-22.

Yoon MJ, Lee SJ, Kim E, Park SH (2012) Doppler ultrasound to detect pulpal blood flow changes during local anaesthesia. International Endodontic Journal 45, 83-7.

Abstract (IN KOREAN)

초음파 도플러를 이용한 치수 혈류 측정과 검사자내 및 검사자간 신뢰도 분석

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I. 목적

치수 상태에 대한 진단은 임상가들에게 때때로 어렵고도 모호하다. 일반적으로 치수 상태를 알아보기 위하여 한랭/온열 검사와 전기 치수 검사(EPT)가 흔히 사용되지만, 이 검사들은 치수내 신경 반응 여부를 알아보는 것으로, 진정한 의미의 치수 생활력을 의미하는 치수 혈류를 측정할 수는 없다. 이에 본 연구의 목적은 다음과 같다: 1) 초음파 도플러를 이용하여 임상적으로 정상인 상악 전치의 치수 혈류를 측정하고, 2) 초음파 도플러의 검사자내 신뢰도와 검사자간 신뢰도를 분석한다.

II. 방법 및 재료

1) 연구 1: 상악 전치의 치수 혈류 측정

62명의 환자 (평균 연령 29.8세, 최소 22세 ~ 최대 52세, 여성 26명과 남성 36명)로부터 총 359개 치아의 치수 혈류를 MM-D-K 초음파 도플러를 이용하여 측정하였다. 수집된 데이터를 치아 종류 별로 상악 중절치, 측절치, 견치로 나누어 일원배치 분산분석 및 Bonferroni 사후검정을 시행하여 95% 신뢰수준에서 통계처리하였다.

2) 연구 2: 검사자내 및 검사자간 신뢰도 분석

37명의 환자 (평균 연령 25.4세, 최소 21세 ~ 최대 32세, 여성 13명과 남성 24명)로부터 총 222개 치아의 치수 혈류를 MM-D-K 초음파 도플러를 이용하여 측정하였다. 검사자내 신뢰도 분석을 위하여 1명의 검사자가 약 1주의 간격으로 3회 측정하였다. 환자는 무작위 순서로 측정하였으며, 이전 측정된 데이터를 알 수 없게 하였다. 검사자간 신뢰도 분석을 위하여 총 5명의 검사자가 무작위 순서로 측정하였으며, 이전 측정된 데이터를 알 수 없게 하였다. 수집된 데이터는 반복측정 분산분석 및 Tukey 사후 검정을 시행하여 95% 신뢰수준에서 통계처리하였으며, 검사자내 및 검사자간 측정값 전체의 급내상관계수(Intraclass correlation coefficient)를 계산하였다.

III. 결과

1) 연구 1: 상악 전치의 치수 혈류 측정

상악 중절치, 측절치, 견치의 치수 혈류는 각각 0.58 cm/s, 0.58 cm/s, 0.52 cm/s 로 측정되였으며, 치아 종류에 따른 통계적 차이는 유의성을 보이지 않았다(P>0.05)

2) 연구 2: 검사자내 및 검사자간 신뢰도 분석

검사자내 신뢰도: 3회의 측정 사이에 반복측정 분산분석시 유의한 차이를 보이지 않았다(P>0.05). 급내상관계수는 0.56으로 중간수준의 일치도를 보였다.

검사자간 신뢰도: 5명의 검사자 사이에 반복측정 분산분석시 유의한 차이를 보였다(P=0.044). 급내상관계수는 0.41로 낮은 일치도를 보였다.

IV. 결론

본 연구에서 성인의, 임상적으로 정상인 상악 전치는 0.56 ± 0.46 cm/s의 치수 혈류를 보이는 것으로 측정되었다. 또한 상악 중절치, 측절치, 견치에 따른 통계적 차이는 보이지 않았다. 검사자내 신뢰도 분석에서 측정간 유의한 차이는 나타나지 않았으나, 일치도는 중간수준으로 계산되었다. 검사자간 신뢰도 분석에서는 검사자 사이의 통계적으로 유의한 차이를 보였으며, 낮

은 일치도가 나타났다. 초음파 도플러 측정 결과의 신뢰도를 높이고 변화를 줄이기 위하여 검사자의 측정 방법 동일화 교육 및 개인 맞춤형 스플린트 사용 등 추가적인 방법에 대한 연구가 필요하다.

핵심되는 말: 치수 혈류 속도, 치수 생활력, 초음파 도플러, 검사자내 신뢰도, 검사자간 신뢰도