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Improvement of the detection of human pulpal blood flow using a laser Doppler flowmeter modified for low flow velocity

Xiaofu Qu [a,b], Motohide Ikawa [a,*], Hidetoshi Shimauchi [a]

[a] Division of Periodontology and Endodontology, Department of Oral Biology, Tohoku University Graduate School of Dentistry, 4-1 Seiryo-machi, Aoba-ku, Sendai 980-8575, Japan

[b] Dalian Stomatological Hospital, Changjiang Road 935, Shahekou District, Dalian 116021, China

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ABSTRACT

Objective: Human pulpal blood flow (PBF) signals as measured by laser Doppler flowmeter (LDF) decrease with age. Although this decrease is considered to be due in part to slow blood flow, information regarding this velocity in humans has been lacking. The aims of the present study were to estimate the blood flow velocity in human dental pulp and to evaluate the validity of LDF modified for the measurement of slow blood flow.

Design: Mean blood flow velocities at the upper central incisor, gingiva, fingertip and forearm of 28 volunteers (mean age: 38.6 years old) were estimated using LDF with a frequency analyser. Blood flow signals at these measurement areas were recorded using two different LDFs: (a) one with a standard blood flow range; and (b) one modified for low blood flow velocity.

Results: The frequency range of the Doppler shift measured at the teeth with an opaque rubber dam was the narrowest (median: 4.3 kHz) among all of the measurement areas. The estimated mean blood flow velocity was the slowest at the teeth with a dam (median: 0.18 mm/s). LDF for low blood flow velocity detected larger and clearer pulsatile blood flow signals from the teeth with dams than did standard LDF.

Conclusions: The present results indicate that the velocity of PBF in humans is very low and that LDF modified for the measurement of slow blood flow is appropriate for PBF measurement in humans.

1. Introduction

Laser Doppler flowmeter (LDF) has been applied to measure pulpal blood flow (PBF) in both humans[1–3] and experimental animals.[4–7] Recently, LDF has become one of the most popular techniques applied for pulp vitality testing in traumatised teeth,[8,9] because of its greater sensitivity and specificity compared to other dental pulp tests.[10] However, LDF for measuring PBF seems to have limitations and difficulty when applied in elderly people. We previously reported age-related decreases in PBF, with the magnitude of PBF signal becoming very small in elderly subjects.[11] The vascular structures and blood vessels supplying the coronal portion of the pulp have been reported to decline with age.[12,13] As with other changes in the dentin-pulp complex involved in decreasing PBF, the size and the volume of the pulp decreases with the increase in calcified tissue with aging.[14,15]

* Corresponding author. Tel.: +81 22 717 8336; fax: +81 22 717 8339.
E-mail address: ikawa@dent.tohoku.ac.jp (M. Ikawa).

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Age-related changes in PBF could be also due to the decrease in velocity at the peripheral area of the dental pulp. However, only limited information is available regarding the blood flow velocity in the pulp. Kim[16] reported that the PBF velocity in rat incisors ranged between 0.11 mm/s and 2.1 mm/s and was slower than that in cat and rabbit omentum.[17] To our knowledge, measurement of the blood flow velocity of human pulp using LDF has been lacking. In the measurement of PBF using LDF, many technical and environmental conditions have been known to interfere the results.[18,19] Among these conditions, nonpulpal signals, principally obtained from nearby gingival tissue, could significantly contaminate the PBF signal.[20–22] For the elimination of the contamination, opaque, heavy-gauge rubber dams have been reported as sufficient.[3,23] An appropriate tooth isolation device should be applied to measure human PBF velocities.

LDF was generally designed for the measurement of organs that have abundant blood flow, such as the brain,[24,25] skin and[26,27] muscle,[28] but not for the dental pulp. Thus, it might not be ideal for a standard LDF to measure PBF with low blood volume and/or low blood flow velocity, particularly in elderly subjects. We speculated that the blood flow in human dental pulp would be very slow. The ability of LDF to measure slow PBF would be increased if the specifications of the flowmeter were modified to detect low blood flow velocity.

Therefore, the aims of the present study were: (1) to estimate human PBF velocity with dam application; and (2) to evaluate the validity of the PBF measured by the modified LDF for low blood flow velocity.

2. Materials and methods

2.1. Subjects

The experimental protocol of this study was approved (ref. no. 21-2) by the Research Ethics Committee of Tohoku University Graduate School of Dentistry, based on the guidelines set forth in the Declaration of Helsinki. A total of 28 healthy staff members (age range: 22–55, mean ± *SD*: 38.6 ± 10.4 years old) of Tohoku University Graduate School of Dentistry (20 men, 8 women), who had healthy vital upper central incisors, participated in this study. All of the volunteers were healthy with no systemic or cardiovascular disease, no evidence of hypertension and no current usage of cardiovascular medication. The upper central incisors of the subjects were diagnosed as healthy if the teeth were free of caries, defects, attrition, recession and discoloration. Prior to data recording, the purpose and methods of this study were explained to each subject, and written informed consent was obtained from all of them.

2.2. Estimation of the blood flow velocity in the tissue

Blood flow velocities of the tooth pulp, gingiva, skin of the fingertip and forearm of the subjects were estimated by the following procedures. The measurement areas were illuminated with a laser light (2 mW, 780 nm) of the standard LDF (FLO-C1HP Omegawave Inc., Tokyo), and the reflection light was guided to an opt-electric conversion amplifier of the LDF. The opt-electrically amplified signals were then conveyed to a frequency analyser (EZ Analyzer, Omegawave Inc., Tokyo) to analyse the power spectrum of the FFT of the Doppler shift. In order to set up the detection frequency range for the PBF measurement, it was important to examine the frequency characteristic of the power spectrum of the raw signals by using a frequency analyzer. We determined the detection frequency range (the cut-off frequency: COF) of each power spectrum to exclude signal noise at frequencies higher than the COF because those frequencies produced only opt-electric conversion amplifier noise. After determining the COF, the mean value of the frequency (*F*) was calculated using the signals lower than the COF. As the mean frequency (*F* kHz) is related to the mean velocity (*V* mm/s) of red blood cells,[29] *V* in the detected volume of the tissue was estimated as: $V = (2.9 \times F)/1000$ (mm/s).

2.3. Blood flow measurement using two different laser Doppler flowmeters

The blood flow signals of the tooth pulp, gingiva, fingertip and forearm of the subjects were measured by two LDFs. A standard LDF having the detected frequency range of 24 Hz–24 kHz was modified to have that of 24 Hz–5 kHz by the preliminary test of the FFT analysis. The blood flow range of the standard LDF was 0–100 (ml/min/100 g equivalent), and that of the modified LDF was 0–10 (ml/min/100 g equivalent). The analog outputs of the both LDFs were 0–10 V, and the 10 V were converted to 100 (ml/min/100 g equivalent) for the standard LDF and 10 (ml/min/100 g equivalent) for the modified LDF.

The gain resistor of the opt-electric amplifier of the modified flowmeter was 100 MΩ: 2.5 times of the resistor for the standard flowmeter used in this study (40 MΩ) (Fig. 1). The signal to noise ratio of the amplifier becomes higher by using a higher resistor, but the detectable frequency range becomes lower. We also added a 5 kHz low-pass filter in the operation circuit.

To compare the blood flow signals measured by the two flowmeters, a specially designed measurement probe (Fig. 2) was used to record the blood flow signals simultaneously, using the two flowmeters. This process was enabled by embedding a prism in a probe and dividing the reflection light from the tissues into to two optical fibres for receiving. The diameter of the fibres for light incidence and receiving the reflection light were both 0.1 mm, with a separation distance of 0.5 mm from the centre of each fibre.

2.4. Recording procedures

Prior to the measurement, impressions of the upper incisors were obtained to prepare an individual resin cap that covered the labial and palatal surfaces of one of the upper central incisors of the subjects, leaving a gap of approximately 2 mm between them. The cap was extended to cover the labial surface of the attached gingiva. Two stainless-steel tubes were embedded in the cap: one was in the labial tooth surface approximately 2 mm from the gingival margin, and the other was in the portion facing the gingiva. A small acrylic plate was prepared in advance for the measurements at the fingertip and