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ORIGINAL ARTICLE

Microcirculatory Characteristics in Neck/Shoulder of the Adults with Sedentary and Exercise Lifestyles

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Abstract High risk of musculoskeletal diseases had been demonstrated in many people with a sedentary lifestyle. As microcirculation provides primary information on tissue health, this paper aims to compare the perfusion characteristics in neck/shoulder of individuals at different physical activity levels. High power laser Doppler flowmetry (LDF) system and averaging algorithm were used to obtain the microcirculatory characteristics. Thirty-two participants with different exercise habit were recruited, which were divided into sedentary group ($n = 16$) and exercise group ($n = 16$). The participants in both groups were matched in age, gender, and body mass index. Peripheral blood perfusion signals on the neck-shoulder region pre- and post- upper trapezius stretching were acquired using LDF with a noninvasive wide separation probe. A modified beat-to-beat algorithm was then applied for the analysis of the microcirculatory signals, including pulsatile and non-pulsatile components. The Mann–Whitney U test was used to compare the differences of perfusion characteristics between these two groups. The pulsatile component of LDF signals in the exercise group was greater than that of the sedentary counterparts after the upper trapezius stretching ($P < 0.05$). Furthermore, the index of perfusion pulsatility (ratio of pulsatile component to mean LDF signal) of the exercise group was significantly higher than that of the sedentary group ($P < 0.01$). This index could differentiate these two groups both at the baseline and post-stretching. Even with low exercise volume, exercise group with regular physical activity appear noticeably different in microcirculatory characteristics in this study. The subjects who exercised had higher values of microcirculatory pulsatility. These findings may encourage people to exercise more often based on the benefit in microcirculation even with small increases in physical activity volume.

Keywords Microcirculation · Physical activity · Exercise · Laser-Doppler flowmetry · Pulsatility

1 Introduction

Regular physical activity (PA) not only benefits physical and mental health, it also plays a critical role in the prevention of many chronic diseases [1, 2]. Generally, the effect of PA on physical health may reflect both improved cardiorespiratory and muscular functions [3]. Furthermore, PA will also benefit on microcirculatory functions. For example, PA resulted in enhanced endothelium-dependent vasodilation [4], and therefore reduced cerebrovascular and cardiovascular events [5].

The improvement of the microcirculatory function as an effect of PA could be evaluated by several techniques. Laser-Doppler Flowmetry (LDF), using the Doppler effect,

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is one of the most convenient techniques for routine tissue perfusion assessments. Due to the limitation of the sampling depth, blood flux was measured using a intramuscular LDF technique by directly inserting the optical fiber into the deeper region in some early studies. The results showed that the blood flux increased during muscle working, and a marked reactive hyperemia was observed for a period after cessation [6, 7]. Generally, the sampling depth of the noninvasive LDF is determined by the corresponding laser power and the separation between the light emitting and the receiving optical fibers. Using a low power laser, the typical sampling depth is less than 1 mm if the fiber separation is 0.5 mm. On the other hand, Clough reported that the proportion of photons reaching depths greater than 1.4 mm was 65.0% using a non-invasive LDF technique, if a laser power of 20 mW and a skin probe with wider fiber separation of 4.0 mm were applied [8].

Further, recent studies using noninvasive LDF techniques and applying algorithms on signal processing have shown promising results in microcirculation investigation [9–11]. Hsiu processed the low power LDF perfusion signal with the aid of the synchronous ECG measurement by using the beat-to-beat algorithm to investigate microcirculatory characteristics in time domain. They had successfully differentiated the characteristics of microcirculatory perfusion and the microcirculatory regulatory activities at local vascular beds between subjects with polycystic ovary syndrome from healthy subjects [12]. A similar technique was also applied in our previous study, in which the variations of microcirculatory characteristics in healthy young subjects with different calf flexibility was found [13]. Therefore, a modified beat-to-beat algorithm and LDF with wider fiber separation were applied in this study.

According to the recommendation from the World Health Organization (WHO) for maintenance and promotion of health, all adults should accumulate at least 150 min of moderate-intensity physical activity throughout the week [14]. However, there are about 70.1% of workers in Taiwan exercised less than one time each week [15]. As neck and shoulder symptoms are common in sedentary occupations [16–18], the aim of this preliminary study is to assess the microcirculatory difference on neck-shoulder region of the workers with and without regular low-volume physical activity. By using a noninvasive LDF technique with the wider fiber separation, three microcirculatory characteristics were obtained from signal processing via a beat-by-beat algorithm. Besides, upper trapezoid muscle stretching is an efficacious and convenient exercises usually recommended as an appropriate therapeutic intervention to relieve stiffness and pain [19, 20]. The microcirculatory characteristics of pre- and post- stretching between the two groups were also compared in this study.

2 Materials and Methods

2.1 Subjects

The goal of our study is to explore the effect of low-volume physical activities on microcirculatory function. According to previous studies, it was not easy to detect the minimal clinical difference by only measuring resting mean blood flux. Therefore, the pulsatile microcirculatory blood flux was used as an indicator in this study. Furthermore, according to our experience, the coefficient of variation of pulsatile microcirculatory blood flux is about 0.4. If the level of significance $\alpha = 0.05$, power of test $1\text{-}\beta = 0.8$, then the minimum sample size is calculated as 16. Finally, a sample of convenience of 32 female participants was recruited in this study, sixteen of them with regular low-volume physical activities (exercise group) and the other sixteen participants were in a sedentary lifestyle (sedentary group). In the exercise group, eight subjects participated in a 1-h per week, supervised aerobic training session in a community health club. The other eight subjects engaged in biking or hiking regularly in each weekend. Six participants in the exercise group met the WHO recommended 150 min of moderate-intensity aerobic physical activity each week, but the other 10 subjects exercised less than the WHO standard for the past three months. The mean exercise time was 106 ± 63 min each week. The participants of the sedentary group had no exercise habit for the past three months. This study was approved by the ethics committee of Kuangtien General Hospital in Taichung city (approval number 10132). Informed consent of all the volunteers was obtained prior to the study.

2.2 Protocol

All trials were conducted in participants' personal break time during workdays. The participants were supine on a comfortable couch in a quiet, comfortable room with the temperature maintained at 24 ± 1 °C for at least 20 min before the measurements. There were two 10-min measurements to monitor the skin microcirculation around the neck-shoulder region for each subject. After the first baseline (BL) measurement was taken, participants practiced upper trapezius stretching fifteen times, with holding at stretched position for 10 s at a time. Then the other 10-min post-stretching (PS) measurement was conducted. All subjects did not take any medication for 3 days before the experiments. They were also asked not to consume food or drink for at least 1 h before test, and also to refrain from exercise and drinks containing alcohol or caffeine during the day of the trial. To avoid the interference from fasting, postprandial effects, or fatigue from work, all of the trials were performed only during 8:30 AM—11:00 AM or 2:00 PM—4:30 PM. The blood pressure and heart rate (HR) were monitored by a sphygmomanometer (Omron, Taiwan), and the questionnaires were administered to collect the information about personal exercise habits including exercise type, frequency, and period before trial.

The Chinese version of the Standardized Nordic Musculoskeletal Questionnaire (NMQ) were also administered to collect the information of perceived neck/shoulder symptoms. The level of pain intensity was rated on a 10-level visual analog scale indicated as 0 (no pain) to 10 (unbearable pain). More than half participants of the sedentary group reported neck/shoulder pain.

2.3 Measurement system

The microcirculatory perfusion on neck-shoulder region was measured using a high power LDF (VMS-LDF1-HP, Moor Instruments, UK) with a 785 nm, 20 mW laser (Class 3R per IEC 60825-1:2007) and a wide separation (4 mm) non-invasive skin probe (VP1-V2-HP). The LDF skin probe was attached on the 45% of the line from the seventh cervical vertebrae to the acromion on neck-shoulder, which is in the middle of the more-horizontal fibers of the upper trapezius. This is also the location of trigger point of myofascial pain syndrome [21]. All of the measurements were conducted according to the safety requirements of LDF. Calibration of the probes was performed using aqueous suspension of polystyrene latex particles before measurements. Brownian motion in the suspension provided a standard value. In this investigation, both microcirculatory flux and electrocardiograph (ECG) signals of participants were simultaneously and synchronously detected and sampled via an analog-to-digital converter (ADLINK, PCI-9111DG, Taiwan) with a sampling rate of 1024 Hz. The digital signals were sent to a personal computer for further analysis. ECG signals were measured with lead II using surface electrodes and the bio-impedance amplifier (EBI100C, BIOPAC system, USA). A high sampling rate (1024 Hz) was used to ensure that the locations of the ECG R-peaks of each pulse could be determined accurately.

2.4 Signal Processing

As the pulsatile blood pressure of peripheral arteries has been used as a diagnostic method, known as “pulse feeling”, for more than 3000 years in traditional Chinese medicine, previous studies have indicated that the pulsatile blood pressure wave plays an important role in regulating blood transportation in large arteries [22]. We follow this rationale to compare the pulsatile perfusion, which is driven by pulsatile pressure wave, between people with different exercise habits. Although the original LDF signal did not reveal a visually apparent regular waveform, we can use the periodicity of ECG signal as a reference to determine the mean waveform of the microcirculatory flux in one heartbeat period, in which the R peaks exhibited the periodicity of the heartbeats. This is because the driving force of the blood flow comes from the pressure pulse produced by the regular contractions of heart. The beat-to-beat algorithm, which derives the mean waveform of the 10-min LDF signal, are described as follows:

(1) Locate ECG R peaks and segment LDF signal according to ECG period:
Prior to locating R peak, the ECG signals were filtered with a digital high-pass filter with a cut-off frequency of 0.1 Hz to eliminate the baseline drift [23]. After locating the R peaks, we used the x-coordinate of the peaks as breaking points to divide the LDF signal into segments. Every segment can be treated as the signal of the blood perfusion within one period of heartbeat. Therefore, if there are N + 1 peaks found in ECG, it means that we can divide both ECG and LDF signals into N periods, giving us N segments of the LDF waveform.

(2) Normalize the N segments:
The problem of heartbeat periods of different lengths needs to be dealt with carefully. Though the heart beat pattern is regular most of the time, the period of heartbeat of a person is not a constant. Even for the same person, the period length varies slightly over time. Therefore, the segments which were divided according to the R peaks of ECG would not have the same length exactly, and should be normalized into the same length without distorting the overall waveform pattern. To achieve this goal, we use the interpolation and down sampling algorithm to either lengthen or shorten the divided LDF signal segments to make all of them of equal length.

(3) Derive the mean of the N waveform segments:
The mean of the N waveform segments was derived and the mean LDF signal waveform was obtained in this study, in which the parameters of perfusion characteristics were defined.

2.5 Statistical Analyses

The statistical analyses were performed using SPSS (release 19.0, SPSS Inc., Chicago, IL, USA). LDF data were not normally distributed, according to the results of the Kolomogoro-Smirnov goodness of fit test. Therefore, the changes of perfusion characteristics between the sedentary group and the exercise group were compared using the Mann–Whitney U test for non-parametric data. The

Wilcoxon signed-rank test, which is a test applied to paired samples, was used to evaluate the difference of perfusion characteristics before and after stretching. A value of $P < 0.05$ was considered statistically significant. The data are expressed as median, quartiles and range.

3 Results

Figure 1 illustrates two synchronous signals: original microcirculatory flux (LDF signal) and ECG obtained via an analog-to-digital converter synchronously. The periodic signal is ECG, in which the R peaks exhibited the periodicity of the heartbeats, while the LDF signal did not reveal a regular waveform. The mean microcirculatory waveform of more than 600 waves in the 10-min LDF measurement is shown in Fig. 2. The time zero of the x-axis was the time that the ECG R peak located. The peak of the mean LDF signal is located at about 0.25 s after the R peak. By analyzing the pulsatile and the nonpulsatile (static) components of LDF signals, we defined three indices as shown in Fig. 2: the mean value of LDF signals indicates the mean microcirculatory blood flux (MMBF) in the peripheral tissue; the pulsatile microcirculatory blood flux (PMBF) is defined as the mean height of the pulsatile component of the LDF signal. The perfusion pulsatility (PP) is defined as the ratio of the PMBF to MMBF as shown in Eq. (1).

$$PP = \frac{PMBF}{MMBF} \qquad (1)$$

The blood perfusion of upper trapezius as monitored by LDF was compared between the sedentary and the exercise groups. Table 1 shows the physical data and the pain level of the two groups. Both groups were matched in age, gender, body mass index and blood pressure, except heart rate. The mean pain level was slightly higher in the sedentary subjects than in the exercise group.

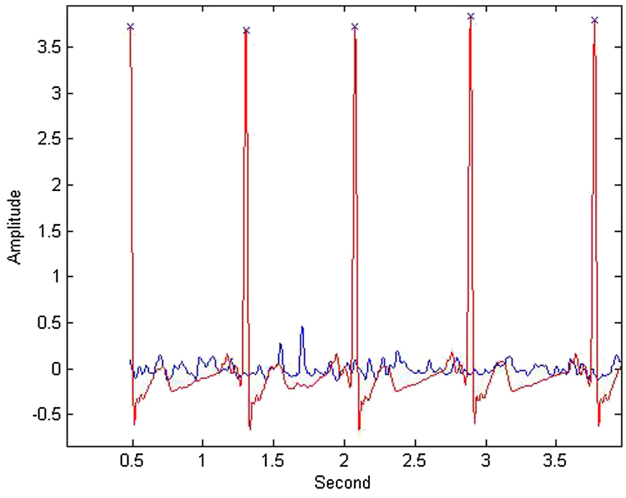

Fig. 1 LDF signal (*blue*), plotted together with ECG signal (*red*)

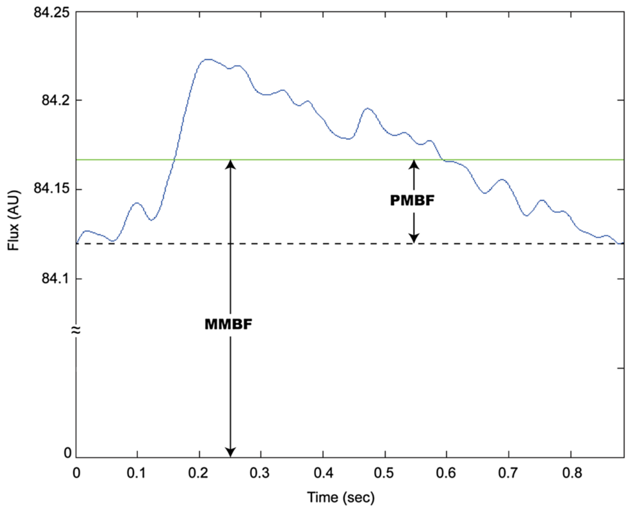

Fig. 2 The MMBF and PMBF of the mean LDF waveform derived from the segments of microcirculatory flux signal

Measurements were conducted pre- and post- stretching for each participant. Figure 3a–c show the MMBF (nonpulsatile component), PMBF (pulsatile component) and PP values (PMBF to MMBF ratio) of the skin perfusion on the upper trapezius of the two groups during pre-stretching (BL) and post-stretching (PS). The box represents the 25–75th percentiles. The line within the box is the median value; the whiskers are the minimum and maximum values. The perfusion values were all represented in arbitrary units.

In Fig. 3(a), the median (first quartile-third quartile) of the MMBF values of the sedentary group at BL and PS were 52.5 (44.3–143.0) and 52.5 (40.3–70.5), respectively. Similar to the level at PS of the sedentary group, the MMBF values of the exercise group were 51.0 (41.5–68.0) and 55.5 (37.0–73.8) at BL and PS, respectively. There were 62.5% (10/16) participants in the sedentary group showed a decrease in MMBF flux after stretching, and the difference is near significant ($P = 0.056$, shown in Table 2). In contrast, upper trapezius stretching only caused a slight change in the MMBF value of the exercise group. In the sedentary group, the behavior of the PMBF component pre- and post-stretching was similar to that of the MMBF. In addition, the medians of PMBF component at PS (5.5) in the sedentary group was significantly lower than that of the exercise group (8.0) ($P = 0.017$). Furthermore, the medians of the PP value of the exercise group, 0.140 and 0.140 (BL and PS) were significantly higher than those of the sedentary group, 0.100 and 0.105 ($P < 0.001$ and $p = 0.005$).

Table 1 Physical characteristics of the subjects in two groups

	Sedentary group (n = 16)	Exercise group (n = 16)	P
Age (years)	33 ± 7	31 ± 6	ns
Body mass (kg)	58 ± 12	55 ± 8	ns
Body height (cm)	161 ± 3	158 ± 6	ns
BMI	22 ± 5	22 ± 4	ns
Systolic pressure (mmHg)	113 ± 9	113 ± 16	ns
Diastolic pressure (mmHg)	74 ± 8	72 ± 14	ns
Heart rate (min^{-1})	77 ± 7	71 ± 9	0.045
Pain level	2.8 ± 2.4	1.5 ± 2.2	ns

Mean ± SD

Exercise group: the mean exercise time = 106 ± 63 min per week; sedentary group: without exercise habit for the past three months

4 Discussion

Although the microcirculatory pulsatility could not be observed in the original LDF signals monitored noninvasively, the mean pulsatile waveforms can be retrieved successfully via the beat-to-beat algorithm with the aid of the ECG signals measured synchronously. This result is not surprising considering the fact that the microcirculatory perfusion is driven by the pulsatile arterial pressure which is produced by the periodicity of heartbeat signal. The different characteristics of the mean microcirculatory waveform (MMBF, PMBF and PP) were observed pre- and post-stretching in this study for the participants with different exercise habits, even the exercise level did not meet the minimum requirements. High-blood-flux, which disappeared after stretching, was found on part of the sedentary participants, but this phenomenon was not observed in the exercise group. Compared with the sedentary group, the PP value is capable of distinguishing the perfusion characteristics of the exercise group, which remains a significantly higher value even after stretching.

It is known that the peripheral vascular beds are the principal sites of regulating the blood supply to tissues. The blood pressure remains almost constant in aorta and arteries, but drops in the regions of arterioles. Precapillary sphincters, following arterioles, control the entrance to microvascular beds, and the potential energy of blood pressure is transformed into the kinetic energy of blood flow when blood is extruded into microvascular beds. Meanwhile, the blood-flow driving force breaks up the red blood cells interlink as well as disassociate the cell aggregation and therefore accelerate the blood flow in the microcirculation [24]. The pulsatile pressure peak can drive blood flux more effectively in microvascular beds because the blood viscosity is reduced and the precapillary opening could be vastly dilated [11]. The higher PP, representing a larger pulsatile peak, in the individuals with exercise habits seems to imply the better perfusion efficiency of blood supply in the peripheral microvascular beds. Compared with the studies using the similar signal processing algorithm to analyze the LDF signals, the pulsatile microcirculation was significantly reduced in subjects with the polycystic ovary syndrome when compared with the healthy controls [12]. Also in our previous study of ankle flexibility, a higher perfusion pulsatility was found on the calf skin of the young subjects with better flexibility when compared with that of lower flexibility [25]. Further, in Van den Brande's study [26], he found the ratio of pulsatile to mean microcirculatory flux (similar to our PP index) was reduced in elderly. As the lower PP value implies the degradation of the microcirculatory function, our study indicates that even engaging in low volume exercise regularly should help to counterbalance the degradation.

High-blood-flux is another important characteristic, which was not found in the individuals with regular low-volume exercise habits, but was observed in part (62.5%) of the sedentary group. This phenomenon has also been observed in Røe's study in shoulder area, which the LDF with an invasive single-fiber technique was used [7]. Their results revealed that almost half the pain-afflicted subjects showed a higher blood flux signals during the tasks related to low level, repetitive and static activity patterns, and thus had a larger inter-individual variation than the healthy individuals. In the other study for the chronic pain, the marked high-blood-flux was also observed not only during the low level tasks, but also continued after cessation of the computer work, even as the EMG of the muscle had already declined to the baseline values [27]. We suggest the high-blood-flux phenomenon in this study should be similar to Røe's findings, because our measurements of the individuals with pain symptoms were also taken in the break time during the working day. It is worth mentioning that both the MMBF and the PMBF showed high-blood-flux only before stretching, but not after stretching. Although the shoulder pain levels of the sedentary workers were not significantly higher (Table 1), we suggest the

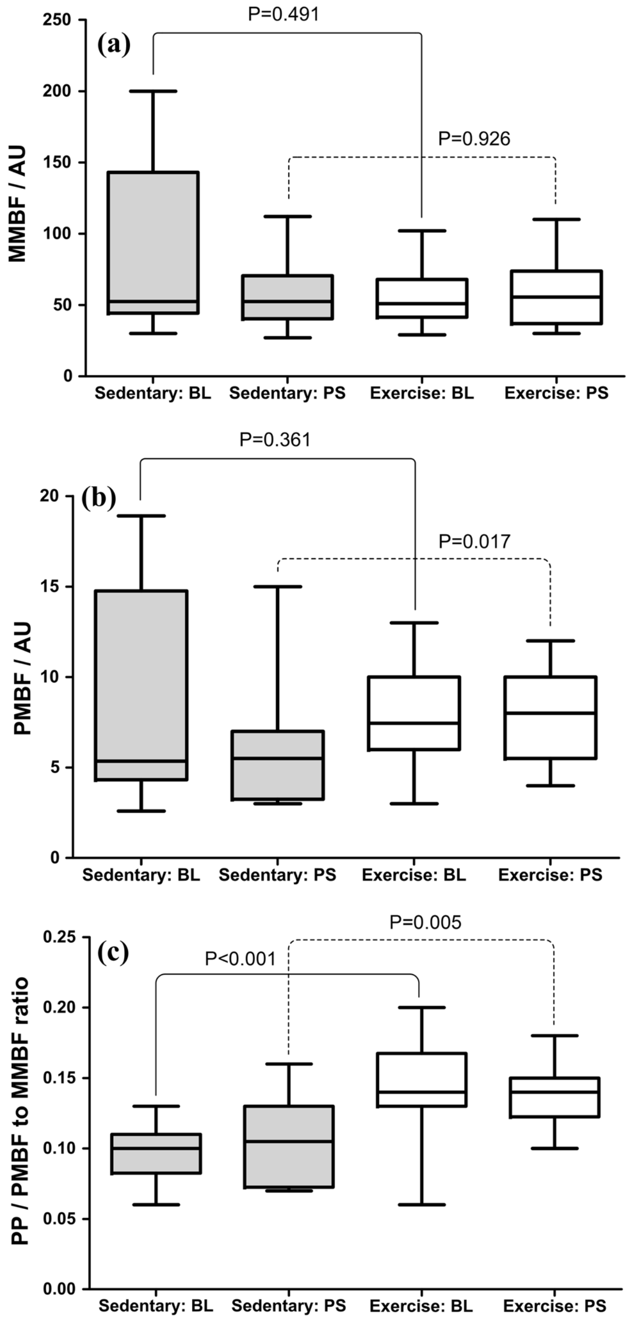

Fig. 3 **a** The mean microcirculatory blood flow (MMBF) of the mean LDF waveform in the sedentary group and the exercise group at baseline (BL) and post-stretching (PS). **b** The pulsatile microcirculatory blood flow (PMBF) of the mean LDF waveform in the sedentary group and the exercise group at baseline (BL) and post-stretching (PS). **c** The perfusion pulsatility (PP) of the blood perfusion of upper trapezius in the sedentary group and the exercise group at baseline (BL) and post-stretching (PS)

high-blood-flux could be the physiological response associated with muscle tightness, neck-shoulder pain or chronic inflammation induced by sedentary lifestyle. The mechanisms of the disappearance of the high blood flux after upper trapezius stretching needs further investigation.

Table 2 P-values of the difference between pre-stretching (BL) and post-stretching (PS) of the MMBF, PMBF and PP values in two groups

	Sedentary group (n = 16)	Exercise group (n = 16)
MMBF	0.056k	0.378
PMBF	0.201	1.000
PP	0.202	0.216

After the high-blood-flux was relieved, sedentary group exhibited 31% lower PMBF than the exercise group (Fig. 3(b)). In Van den Brande's study [26], in which the LDF measurement was performed on the dorsum of the foot, the mean resting flux (similar to MMBF) and the amplitude of the pulsatile flow (similar to PMBF) were significantly reduced in elderly people. It is known that aging leads to multitude of changes in the cardio-vascular system like increasing arterial stiffness, decreasing muscle blood supply, lower maximum oxygen consumption and decreasing pulse pressure wave amplification, etc. [28–30]. Davies suggested that it may be more likely that differences in lifestyle factors consecutively lead to a decrease in maximum oxygen consumption [28]. It is reasonably demonstrated that the reduction of PMBF in the sedentary group also reveal the degradation of the dynamic activities of microcirculation. Moreover, the lower PP (PMBF/MMBF) in sedentary people due to a greater decrease in PMBF (Fig. 3(b)) than in MMBF (Fig. 3(a)) further implies that for people in sedentary lifestyle, their PMBF might be degraded earlier than MMBF. Our finding reveals regular exercise will ease the degradation of the microcirculatory functions.

Skin thickness varies between different regions on the body surface. The thickness of the skin (epidermis plus dermis) ranges from 0.5 to 2 mm [31]. The cutaneous microcirculation is organized as two horizontal plexuses, including upper horizontal plexus (~1–1.5 mm below the skin surface) and the other is at the dermal-subcutaneous junction [32]. The measuring site in this study was located on the skin of upper back, around the middle of the horizontal fibers of the upper trapezius. The measuring depth of the non-invasive LDF applied in this study was greater than 1.4 mm [8], so the characteristics of microvascular perfusion should be mainly contributed by red cell flux localized to the sites of arterioles in dermis. Within the measured depth, we are unable to measure the vasomotion of the lower horizontal plexus of dermis. Instead, the vasomotion of upper horizontal plexus was measured, so the pulsatility of the perfusion signals was not obvious. However, with the aid of the synchronous ECG signals, the mean microvascular perfusion wave in one heartbeat period can be determined. Further, we proposed that the characteristics of skin microcirculation may reflect the microcirculation of muscle tissues below it. For example, the higher muscle microcirculatory flux of the pain-afflicted subjects in Røe's and Strøm's findings was known as the hyperemia [7, 27]. The high-blood-flux phenomenon was also demonstrated in the sedentary group with higher pain level in our study. Higher blood flux in these subjects was referred to pain symptoms.

The limitations of this study include the timing of the measurements of microcirculatory perfusions which could not be controlled precisely, because the most participants worked in a tight schedule. The high-blood-flux could be relieved just due to the relaxation during break time which cannot be attributed only by the neck-shoulder stretching. Besides, the precise physiological mechanisms leading to the difference of the microcirculatory characteristics between the two groups still need further study. With larger sample size, investigating the influence of the dynamical intervention on the responses of the pulsatile parameters is suggested to validate the contribution of stretching in future study.

5 Conclusion

Microcirculation is responsible for the blood supply to tissues, and therefore provides primary information on tissue health. This study indicated that regularly low-volume physical activities could affect the microcirculatory function. By using the high power LDF system with a wide fiber separation and the averaging algorithm, the significant effect of regular exercise habit on the shoulder microcirculation of the adults was observed. These findings may encourage people to exercise more often even with small increases in physical activity volume. More studies are needed to confirm that if the perfusion pulsatility with a reduced inter-individual variation and high-blood-flux could provide the information associated with chronic pain. Further, this objective, quantitative and non-invasive technique warrants further investigation for the applications in sport performance or rehabilitation.

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