Influence of exercise intensity on dynamics of blood volume caused by muscle pump action

Lu Chen*, Kei Hatano*, Takahiro Yunoki*, Tokuo Yano*

Abstract: The aim of this study was to determine whether total hemoglobin concentration (cHb) in the quadriceps muscle measured by near-infrared spectroscopy (NIRS) varies depending on the muscle pump action accompanying the muscle contraction during incremental exercise and whether this variation is enhanced by exercise intensity. The subjects of this study were 6 healthy adult males. After 5 minutes of rest, exercise intensity was gradually increased at a rate of 50 w per 5 min from 50 w to 200 w. From 200 w, the work rate was increased by 20 w per one min. The subjects continued to exercise until a pedal rotation speed of 60 rpm could not be maintained (exhaustion). cHb in the quadriceps muscle was measured continuously at 20 Hz during exercise. A decrease in cHb was observed in the first pedaling after starting the exercise. The dynamics of cHb during one pedaling was divided into three phases: the rapid decline (phase I), the rapid rise (phase II), and the transition phase from phase II to the next phase I (phase III). The height between the lowest value and the highest value of cHb during one cycle was defined as cHb amplitude. There was no significant change in amplitude of cHb with increasing exercise intensity. Also, each duration of cHb phases did not significantly change with increasing exercise intensity. Therefore, it was concluded that there is a muscle pumping effect on cHb dynamics during incremental exercise. However, it seemed that the defined amplitude for the cHb dynamics did not increase with increasing exercise intensity.

Key Words: total hemoglobin concentration, incremental exercise, exercise intensity, muscle blood volume

Introduction

When the muscles contract, blood vessels in the muscles are compressed, and the blood in the blood vessels is squeezed out. A valve in the vein prevents the blood from flowing backwards. Then, when the muscle relaxes, blood flows from the artery into the muscle (1, 2, 3). This muscle contraction with the action of the venous valve is called a muscle pump. During exercise, the amount of blood in the muscles fluctuates with this muscle pump action. In reports on conventional muscle pumping, the relationship between muscle pumping action and exercise intensity during dynamic motion has been discussed, but no consensus has been reached.

* Laboratory of Exercise Physiology, Faculty of Education, Hokkaido University
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Sheriff et al. (4) reported that muscular pumping action in treadmill exercise of medium intensity (6.4 km/h) caused a rapid increase in muscle blood flow at the start of exercise. However, Hamann et al. (5) reported that the muscle pump action in treadmill exercise of medium intensity (4.8 km/h) did not affect muscle blood flow at the start of exercise in a state in which the blood vessels were maximally expanded with adenosine. In addition, Lutjemeier et al. (6) reported that muscle pump action at a low load (4%-5% (0.07 w) of maximal voluntary contraction) caused an increase in active muscle blood flow but that there was no significant increase in active muscle blood flow with increase in exercise intensity. Therefore, it is not known whether muscle pump action affects blood flow of the active muscle and whether a difference in exercise intensity affects blood flow in the active muscle.

Foster et al. (7) reported that real-time changes in muscle blood volume could be determined by measuring the total hemoglobin concentration (cHb) in muscle tissue using near-infrared spectroscopy (NIRS). Takaishi et al. (8) used NIRS to examine changes in blood volume of active muscles during an incremental exercise and they found that blood volume decreased due to muscle contraction but that the dynamics of blood volume was not affected by exercise intensity. However, their study had the following two limitations. First, in their experiments, the frequency of cHb measured by NIRS was set to 2 Hz and pedal rotation speed was set to 50 revolutions per minute (50 rpm). As a result, it takes 1.2 seconds for one rotation of the pedal, but at a measurement frequency of 2 Hz, the measurement is performed at 0.5-second intervals, and the measurement timing is therefore shifted by 30 degrees at the crank angle. For example, the first sampling is performed from 0 degrees to 150 degrees (0.5 seconds) and from 150 degrees to 300 degrees (1.0 seconds). The second time is sampled from the first 300 degrees (1.0 seconds) to 90 degrees (1.5 seconds) and from 90 degrees to 240 degrees (2.0 seconds). In other words, by utilizing the fact that the crank angle at which cHb is measured deviates with every pedaling, they estimated muscle blood volume at each crank angle and obtained the dynamics of muscle blood volume with change in the crank angle (muscle contraction) during one pedal revolution at each exercise intensity. In order to obtain blood volume dynamics in a time series in this way, it is assumed that at each exercise intensity, the blood volume dynamics is the same for all pedaling. However, it is not clear whether this assumption is correct or not. Another limitation of their study is the possibility that the blood volume dynamics was smoothed because the measurement frequency of 2 Hz (cHb can only be measured about twice with one rotation of the pedal.).

The purpose of this study was to investigate whether fluctuation of cHb measured by NIRS is related to the muscle pumping effect accompanying muscle contraction during an incremental exercise and whether its variation is strengthened by an increase in exercise intensity.

Methods

Subjects

The subjects of this study were 6 healthy adult males. The age, height, weight, and maximum oxygen intake of the subjects were 25 ± 2.8 years, 175 ± 5.9 cm, 72 ± 10.0 kg, and 2.69 ± 0.60 l/min (means ± standard deviation, Table 1). Written consent was obtained from each subject after explaining the purpose of the experiment as well as the potential risks involved. All of the experimental procedures were also explained in detail to each subject. This study was conducted with the approval of the research ethics committee of Hokkaido University, Faculty of Education.

Experimental protocol

In this study, a progressive exercise load test was conducted using a bicycle ergometer (Ergometer Combi 232 xl, Japan). After 5 minutes of rest, exercise intensity was gradually increased at a rate of 50 w per 5 min from 50 w to 200 w (60 rpm). From 200 w, the work rate was increased by 20 w per one min. The subjects continued to exercise until a pedal rotation speed of 60 rpm could not be maintained (exhaustion).

Measurement items

Blood volume (total hemoglobin: cHb)

Using a near-infrared spectrometer (NIRO200NX, Japan), total hemoglobin (cHb) in the vastus muscle was recorded during the exercise. The measurement site was the vastus lateralis muscle and cHb was continuously measured from the resting time of the incremental load exercise test until the end of the exercise. In the probe of this device, light of three wavelengths (735, 810, 850 nm) was irradiated from the light source portion, and the light after tissue penetration was detected by the light-receiving portion separated by 3.0 cm. The sum of oxygenated hemoglobin concentration and deoxygenated hemoglobin concentration obtained by the Modified Lambert Beer (MLB) method (9) was obtained as cHb. The sampling frequency was 20 Hz. From the distance (3.0 cm) between the light source part and the light-receiving part, the theoretical measurement depth was about 2.0 cm (10). In this study, cHb was used as an index of muscle blood volume.

Oxygen uptake (oxygen consumption: VO_2)

Using a respiratory gas analyzer (AE-310S, Minato Medical Science, Japan), oxygen uptake during an incremental exercise test was measured by breath-by-breath method. Averaged VO_2 at every 20 seconds was outputted. The exhalation volume and inspiratory volume were measured with a hot-wire flow meter. A 2 L syringe was used to calibrate the flowmeter. O_2 and CO_2 concentration sensors were calibrated using standard gas of known concentration (O_2: 15.13%, CO_2: 5.068%) before the start of each test. Heart rate was monitored using a heart rate monitor connected to the respiratory gas analyzer.

Blood pH and blood lactate concentration

Blood lactate concentration and blood pH were determined from blood collected from the fingertip at each stage. The collected blood was analyzed with a gas analyzer (i-stat1, istat, Aboott point of care Ine IL, USA). At each work rate immediately before the end of stages until the time of the 200 w and at rest, blood was taken. To make it arterial blood, both hands of subjects were warmed with warm water of 40 to 45 ° C for about 10 minutes before starting each test and then warmed using gloves with a built-in heater (11,12). Regarding the range and accuracy of both measurement items, pH was 6.5 to 8.2, La was 0.30 to 20.00 mmol/L, and coefficients of variation were 0.08% and 3.27% or less, respectively.

Data analysis

Blood volume (total hemoglobin: cHb)

Changes in cHb from the beginning of exercise to the end of exercise were divided into four stages: 50 w, 100 w, 150 w, and exhaustion. Data obtained during a 10-sec period at each stage were used. The chosen times were as follows. First stage was from 4 min 50 sec to 5 min, the second stage was from 9 min 50 sec to 10 min, the third stage was from 14 min 50 sec and final stage was at exhaustion.

The dynamics of cHb was divided into three phases: the period when cHb dynamics rapidly decreased was defined as phase Ⅰ, the period when cHb dynamics rapidly increased was defined as phase II, and the period of transition from phase II to the next phase I was defined as phase III. (Fig. 3) Data for the last 10 sec of each exercise stage were averaged for each subject.

The height between the minimum value in phase I and the maximum value in phase III was defined as the amplitude of cHb dynamics (Fig. 3). The amplitude of each cycle of cHb dynamics was first obtained, and the amplitudes obtained were averaged for the last 10 sec of each stage.

Statistical analysis

Results are expressed as means ± standard deviation. Significant differences in levels of blood La, blood pH and cHb were tested by multiple comparisons (Dunnett' s method). The data for 50 w were used as the baseline in cHb in multiple comparisons. The data obtained at rest were used as baseline levels of lactic acid and pH in blood in multiple comparisons. The level of significance was set at less than 5% ($p < 0.05$).

Results

In this study, an incremental exercise test was conducted using a bicycle ergometer. The maximum work rate of six subjects are shown in Table 1. In two subjects, the exercise text ended in the fourth step (200 w).

Table 1 Age, height, body weight, maximum load and peak oxygen uptake (VO_2 peak) of each of the 6 subjects.

	Age(years)	Height(cm)	Weight(Kg)	Wmax(watts)	$\dot{V}O_2$peak(l/min)
Sub.1	30	172	67	200	2.51
Sub.2	24	173	65	240	2.2
Sub.3	23	167	63	200	2.43
Sub.4	23	180	73	320	3.63
Sub.5	23	172	72	280	3.29
Sub.6	28	185	93	280	3.68
Mean	25	175	72	253	2.96
SD	2.8	5.9	10.0	44.2	0.6

Table 2 shows the lactic acid levels and pH values in arterial blood in an incremental exercise test. The lactic acid concentration in blood increased markedly as the exercise intensity was increased. Blood pH significantly decreased with increase in exercise intensity.

Table 2 Changes in lactic acid and pH value of arterial blood during the progressive load exercise test.

		Rest	5 min (50 watts)	10 min (100 watts)	15 min (150 watts)	20 min (200 watts)
La (mmol/L)	Mean	1.27	1.925*	3.19*	6.17*	10.09*
	SD	0.27	0.34	1.06	2.57	3.78
pH	Mean	7.41	7.37*	7.38*	7.35*	7.31*
	SD	0.02	0.01	0.02	0.03	0.05

*: Significant difference from the resting value ($p < 0.05$)

Figure 1 shows VO_2 at rest and during the incremental exercise test. VO_2 increased and showed a steady state in each stage. Then VO_2 increased until VO_2peak.

Figure 2 shows the dynamics of cHb at the start of exercise. cHb before exercise did not change. However, a decrease in cHb was observed in the first pedaling after the beginning of exercise.

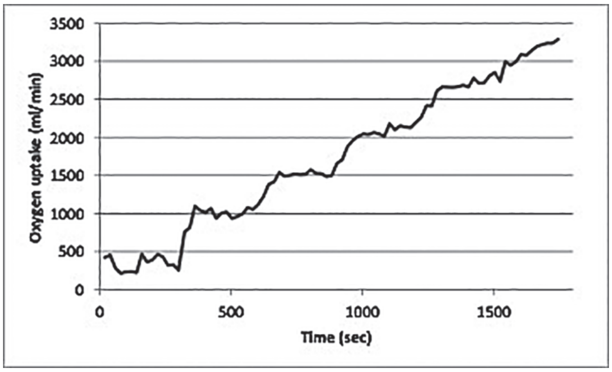

Fig. 1 Oxygen uptake during rest and exercise.

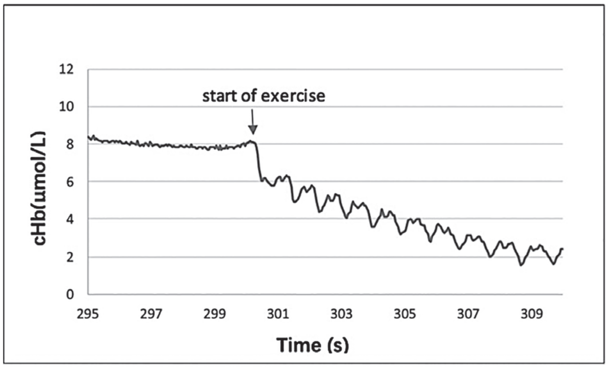

Fig. 2 Changes in blood volume (cHb) after the start of exercise.

Figure 3 shows the dynamics of cHb during two revolutions of the pedal. As explained in the methods, the change in cHb was divided into three phases. After a sharp decrease of phase I, a sharp increase phase II was observed. Then, cHb re-decrease was observed in a few subjects in phases III. As shown in Table 3, the time interval of each phase did not change regardless of the increase in exercise intensity.

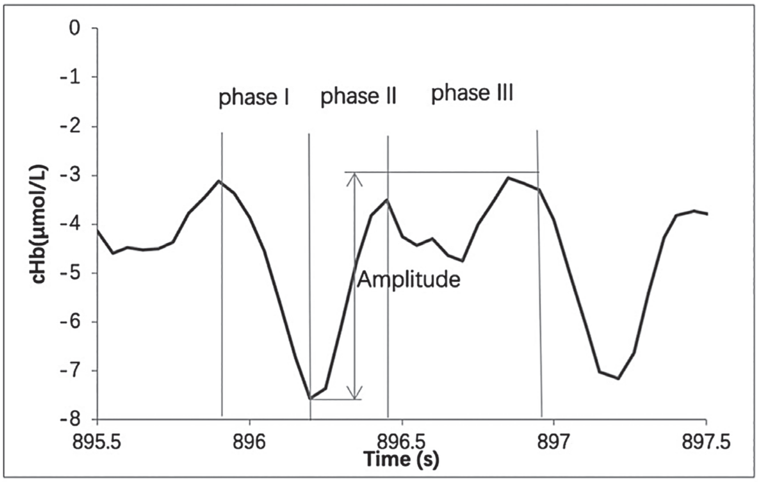

Fig. 3 Changes in blood volume (cHb) during two cycles of pedaling. Division of phase was carried out for one cycle.

Table 3 Each phase of the time of the total amount of hemoglobin at each exercise intensity (see Fig. 5).

		phase Ⅰ(sec)	phase Ⅱ(sec)	phase Ⅲ(sec)
50 watts				
	Mean	0.330	0.310	0.390
	SD	0.129	0.097	0.203
100 watts				
	Mean	0.267	0.308	0.392
	SD	0.107	0.093	0.177
150 watts				
	Mean	0.300	0.342	0.333
	SD	0.082	0.098	0.182
Exhaustion				
	Mean	0.375	0.367	0.260
	SD	0.125	0.152	0.219

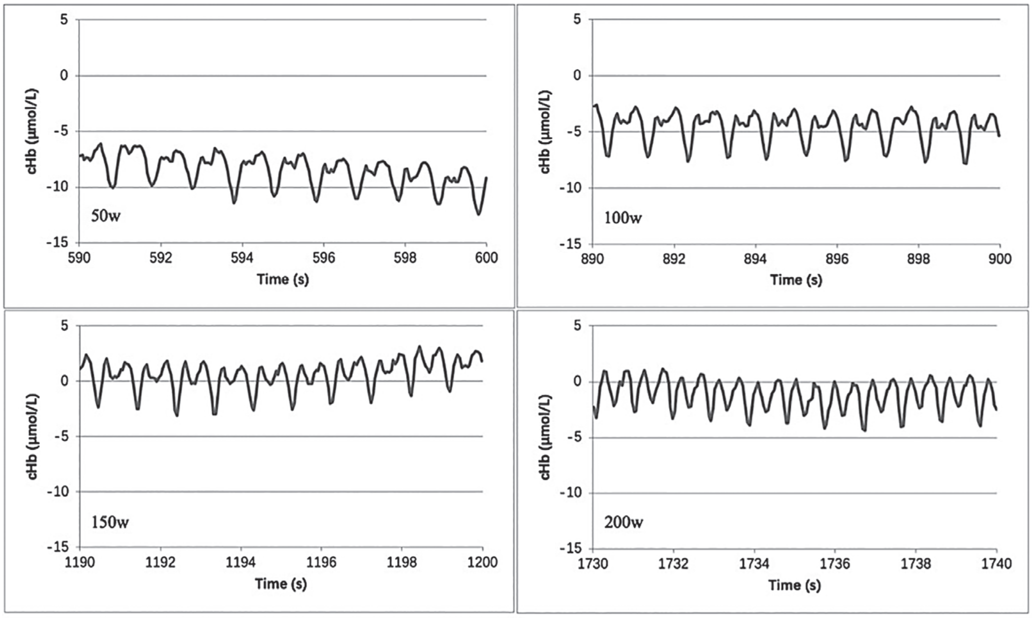

Fig. 4 One example of the temporal in the blood volume (cHb) in the last 10 sec at each stage (50w, 100w, 150w, 200w).

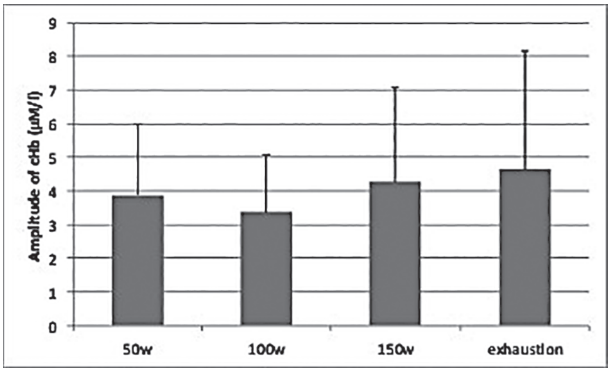

Fig. 5 Amplitude of blood volume (cHb) in the vastus lateralis from 50 w to exhaustion.

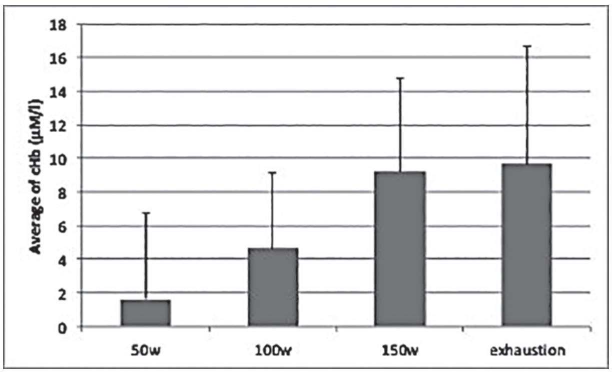

Fig. 6 Average blood volume (cHb) in the vastus lateralis from 50 w to exhaustion.

Figure 4 shows individual dynamics of cHb in the last 10 sec at each stage. One cycle of cHb consisted of 3 phases. The cHb dynamics were repeated 10 times at each stage, but the forms were not necessarily the same within each stage. Figure 5 shows the amplitude of dynamic cHb. There were no significant differences in the amplitude with increasing exercise intensity. Figure 6 shows the value averaged for dynamic cHb for 10 sec. Averaged cHb tended to increase with increasing exercise intensity, but there was no significant difference among the averaged values of cHb.

Discussion

In this study, we measured the blood concentrations of cHb in the vastus lateralis muscle during exercise using Near-infrared Spectroscopy (NIRS). A bicycle ergometer was used for exercise tests. The pedaling rate during exercise was 60 revolutions per minute. Therefore, the muscles contracted and relaxed once per second. Dynamic cHb for one second was divided into three phases. The difference between the maximum value and the minimum value during dynamic cHb was taken as the amplitude. As a result, the durations of the three phases of dynamic cHb were not affected by the increase in exercise intensity. The amplitude of dynamic cHb was also not affected by the exercise intensity.

An initial decrease in cHb was observed in the first pedaling (Fig. 2), suggesting that blood in the muscle was squeezed out by the muscle pump. McCully et al. (10) suggested that blood volume measured by NIRS is derived from capillaries, the sites of arterioles and venules. Therefore, changes in blood volume would be observed at these sites due to the muscle pump action.

In phase Ⅰ, cHb decreased rapidly (Fig. 3). Activation of muscle was observed on electromyography in this phase in a previous study (8). However, in that study, as mentioned in introduction, they shifted the rotational rate of the bicycle ergometer from measurement time of NIRS. By this shift, the blood volume at each rotation angle was determined. In this case, there are two measurements in one rotation, but if blood volume is continuously measured several times, the blood volume can be determined at different positions of the pedal rotation angle. However, in this case it is assumed that the dynamics of blood volume are the same shape despite repetition. Also, because the blood volume is measured at 2 Hz, the measured value becomes the average value of 0.5 seconds. This leads to smoothing of the dynamics of blood volume. In this study, we measured cHb at 20 Hz. As a result, the dynamics of cHb was different in each cycle, and an acute change was observed from phase I to phase II. This is different from the previous study.

Phase Ⅱ and phase Ⅲ are thought to be muscle expansion phases. In phase II, blood rapidly flows into the muscle. This phase is the recovery period from the outflow of blood in the phase I. That is, this period is thought to reflect the inflow of blood. For example, the ascending amount per unit time in phase II corresponds to the blood flow rate. However, in this study, the fact that the phase durations (Table 3) and amplitude (Fig. 5) did not change with increase in exercise intensity cannot be explained by such a simple understanding because blood flow should increase with an increase in exercise intensity, but an increase in blood flow was not observed in phase II.

In phase Ⅲ, it was shown that there was almost no change in blood storage in the muscle (Fig. 3). However, this does not mean that blood flow has stagnated. The storage capacity of blood in the muscle is constant, but the velocity of blood flow in the muscle changes. In other words, the increase in exercise intensity increases blood flow, but the intramuscular blood volume does not change.

In phase III, a decrease in blood volume was observed in a few subjects, though the magnitude of decrease was not as great as that in phase I. In phase I, a leg pushes the pedal and is extended. Phase III is the period when a leg is flexed. At that time, the reverse leg is in the push phase, that is, the extension phase. Because of this extension, the flexion does not become a mere relaxation but stretch. As a result, blood volume decreases.

The duration of each of the phases was not related to exercise intensity (Table 3). This suggests that muscle contraction and relaxation or stretching occur in relation to the position of pedaling. That is since the position itself of pedaling is independent of exercise intensity, it is considered that the duration of each of the phases become the same.

The amplitude of the dynamics of blood volume was not related to exercise intensity (Fig. 5). Intramuscular pressure increases with increase in exercise intensity. Therefore, it can be imagined that the muscle pump action increases accordingly. However, since the capacity of blood storage in muscle is not dependent on exercise intensity, the amount of blood that is ejected in one muscle contraction becomes constant regardless of exercise intensity (Fig. 5). This does not confirm that muscle pumping action is enhanced with increase in exercise intensity.

Changes in average blood volume are thought to be related to skin blood storage and blood storage capacity in the muscles (Fig. 6). Skin blood volume should increase due to a rise in muscle temperature caused by exercise. However, the results for each stage were not significantly different. Therefore, changes in skin blood volume did not affect blood volume in the active muscle. Furthermore, it seems that the blood storage capacity in the muscle during muscle relaxation is not increased by a rise in body temperature accompanying an increase in exercise intensity.

Conclusions

The results of this study suggested that muscle pump action can affect the dynamics of blood volume in the active muscle. However, it seems that the amplitude of dynamics of blood volume does not increase with increase in exercise intensity.

Conflict of interest statements

The authors declare that there is no conflict of interests regarding the publication of this article.

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運動強度が筋ポンプ作用によって生じる血液量の動態に及ぼす影響

陳　璐[*]・波多野　慶[*]・柚木　孝敬[*]・矢野　德郎[*]

【要旨】 本研究の目的は，近赤外線分光法で測定される大腿四頭筋の総ヘモグロビン濃度（cHb）が漸増負荷運動時の筋収縮に伴う筋ポンプ作用に依存して変動するか，またその変動が運動強度によって増強されるかどうかを明らかにすることであった。健康な成人男性６名は，５分間の安静後，自転車ペダリング（回転数：60 rpm）による漸増負荷運動を行い，その間にcHbが20Hzで連続測定された。運動開始後最初のペダリングでcHbの減少が観察された。１ペダリング中のcHbの動態は，急激な低下（第Ⅰ相），急激な上昇（第Ⅱ相），第Ⅱ相から次の第Ⅰ相への移行期（第Ⅲ相）の３相に分けられた。１サイクル中のcHbの最低値と最高値の間の高さをcHb振幅と定義した。cHb振幅および各相の持続時間に，運動強度の増加に伴う有意な変化は認められなかった。以上より，漸増負荷運動中のcHb動態には筋ポンプ作用があるが，cHb振幅は，運動強度の増加とともに増加しないようであった。

【キーワード】総ヘモグロビン濃度，漸増負荷運動，運動強度，筋血液量

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[*] 北海道大学大学院教育学院運動生理学研究室