Prediction of Blood Lactate Accumulation from Excess CO₂ Output during Constant Exercise

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Abstract. To determine the predictability of blood lactate accumulation from excess CO₂ output derived from bicarbonate buffering of lactic acid during constant exercise, eight normal active volunteers were studied during three stages of constant exercise on a cycle ergometer. Three work rates consisted of 100% (stage I), 120% (stage II) and 150% (stage III) of each subject's anaerobic threshold (AT), each of which was lasted for 4 min. Excess CO₂ output (Ex CO₂, ml) at each stage of constant exercise was estimated from the integral of difference between total V̇CO₂ and aerobic V̇CO₂ (from regression line for V̇CO₂ and V̇O₂ at exercise intensities below the AT obtained in incremental exercise test). Ex CO₂ per body mass (Ex CO₂-mass⁻¹) was increased progressively with blood lactate (La) accumulation from rest to each stage of constant exercise. Mean values (±SD) in the measured La accumulation (ΔLa,measured) and predicted La accumulation (ΔLa,predicted) at three stages of constant exercise were 1.82 ± 0.83 vs 3.19 ± 1.70 for stage I, 5.58 ± 3.47 vs 7.09 ± 3.28 for stage II and 12.19 ± 2.36 vs 12.74 ± 1.83 mmol·l⁻¹ for stage III, respectively. There was a significant difference between ΔLa,measured and ΔLa,predicted at stage I (p<0.05), but no significant differences between these two variables at stage II and III. The averaged difference from ΔLa,predicted to ΔLa,measured at stage III (0.55 mmol·l⁻¹) showed a tendency to be smaller than stage I (1.38 mmol·l⁻¹) and II (1.50 mmol·l⁻¹). On the other hand, ΔLa,predicted was found to correlate very closely with ΔLa,measured (r=0.954, P<0.001, n=20). The results of this study suggest that the changes of La accumulation could be predicted from excess CO₂ output generated in constant exercises above the AT.


Keywords: aerobic CO₂, bicarbonate buffering, anaerobic threshold

Introduction

Since Harrison and Pilcher (1930) described that excess carbon dioxide (excess CO₂) was released from bicarbonate (HCO₃⁻) when acids formed during anaerobic metabolism were buffered, it has been well accepted that the volume of excess CO₂ output, resulting mainly from bicarbonate buffering of lactic acid accumulated in exercising muscle and in blood, would be stoichiometrically equivalent to lactate accumulation (Jones, 1980; Sutton and Jones, 1979; Wasserman et al., 1991). Clode and Campbell (1969), using CO₂ balance technique, quantified the excess CO₂ output during steady state exercise and estimated an increase of blood lactate (La) concentration. Their method used in the estimation of La changes during steady state exercise would be thought to have a confidence theoretically on one side and to make complicated procedures practically on the other. Consequently, a new simplified prediction method of La changes during exercise should be developed for practical application such as exercise prescription, because it is important to predict La accumulation in evaluating the magnitude of exercise load to the body from a metabolic aspect.

It has been reported that excess CO₂ output, which is estimated from the difference between total V̇CO₂ and aerobic V̇CO₂ (calculated by a regression line for V̇CO₂-V̇O₂), correlated closely with blood La accumulation and bicarbonate decrease (Hirakoba et al., 1993b). From this point of view, it would be possible to predict La accumulation from excess CO₂ output during exercise. We have therefore tried to predict an accumulation of La during constant exercise by a different method from that of Clode and Campbell (1969). This work has been briefly reported previously (Hirakoba et al., 1993a). The purpose of the present study was to examine a new method for predicting La accumulation from excess CO₂ output during constant exercise.

Methods

Subjects

Eight healthy active male volunteers, who were the same subjects as reported in previous study (Hirakoba et al., 1993b), participated in this study. The average (±
SD) values in physical characteristics and maximal oxygen uptake per body mass (\(\dot{\text{V}}\text{O}_2\) max) and oxygen uptake at anaerobic threshold (AT-\(\dot{\text{V}}\text{O}_2\)) of the subjects are listed in Table 1. They were all informed of the aims and procedures of this experiment, and gave their informed consent.

**Incremental exercise test**

A 1-min incremental exercise test, which has been described previously (Hirakoba et al., 1992), was carried out to determine AT-\(\dot{\text{V}}\text{O}_2\) and \(\dot{\text{V}}\text{O}_2\) max before performing constant exercise. In brief, after 4-min unloaded pedaling, each subject performed on a cycle ergometer (Monark-Crescent AB, Sweden) a 1-min incremental exercise test with an increase of exercise intensity of 30 W every minute until the subject's exhaustion point. A regression line of \(\dot{\text{V}}\text{CO}_2\)-\(\dot{\text{V}}\text{O}_2\) relationship during exercise intensities below the AT, \(\text{CO}_2\) excess (ml) and \(\text{CO}_2\) excess per body mass per unit increase of \(\text{La} \ (\text{CO}_2\text{ excess - }\Delta\text{La})\); ml\(\text{-kg}^{-1}\cdot\text{min}^{-1}\cdot\text{l}^{-1}\)) were calculated according to Hirakoba et al. (1992) and Yano (1987). The values of \(\text{CO}_2\) excess-\(\Delta\text{La}\) of the eight subjects obtained in the incremental exercise are given in Table 1.

**Constant exercise test**

Several days after the above mentioned incremental exercise test, each subject performed three stages of constant exercise on a cycle ergometer. Three work rates consisted of 100% (stage I), 120% (stage II) and 150% (stage III) of each subject's AT-\(\dot{\text{V}}\text{O}_2\), each of which was lasted for 4 min. The work rate for constant exercise was calculated from the linear relationship established between work rate-\(\dot{\text{V}}\text{O}_2\) in the incremental exercise. Numbers of subject who could complete stage I, II and III of constant exercise were eight, seven and five of the subjects, respectively (Table 2), because exercise intensity of the last stage was too high for the three subjects to perform.

Expired gas was collected continuously throughout the incremental and constant exercise tests by an automatic gas analyzer (Aerobic Processor 391, San-ei, Tokyo) and minute ventilation (\(\dot{\text{V}}\)\(_E\)), \(\dot{\text{V}}\text{O}_2\), \(\dot{\text{V}}\text{CO}_2\) were calculated by this analyzer with a computer based-system as previously described (Hirakoba et al., 1993b).

### Table 1 Physical characteristics, maximal oxygen uptake, oxygen uptake at anaerobic threshold and \(\text{CO}_2\) excess of subjects

<table>
<thead>
<tr>
<th>Subject</th>
<th>Age (yr)</th>
<th>Height (cm)</th>
<th>Mass (kg)</th>
<th>(\dot{\text{V}}\text{O}_2) max (ml(\text{-kg}^{-1}\cdot\text{min}^{-1}))</th>
<th>AT-(\dot{\text{V}}\text{O}_2) (ml(\text{-kg}^{-1}\cdot\text{min}^{-1}))</th>
<th>(\text{CO}_2) excess - (\Delta\text{La}) (ml(\text{-kg}^{-1}\cdot\text{mmol}^{-1}\cdot\text{l}^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sub. 1</td>
<td>20</td>
<td>172.0</td>
<td>55.0</td>
<td>59.4</td>
<td>39.2</td>
<td>4.46</td>
</tr>
<tr>
<td>Sub. 2</td>
<td>19</td>
<td>172.9</td>
<td>60.6</td>
<td>55.1</td>
<td>34.9</td>
<td>5.51</td>
</tr>
<tr>
<td>Sub. 3</td>
<td>20</td>
<td>166.9</td>
<td>44.3</td>
<td>58.1</td>
<td>35.9</td>
<td>4.50</td>
</tr>
<tr>
<td>Sub. 4</td>
<td>20</td>
<td>173.1</td>
<td>59.7</td>
<td>57.0</td>
<td>33.1</td>
<td>4.33</td>
</tr>
<tr>
<td>Sub. 5</td>
<td>22</td>
<td>175.0</td>
<td>62.0</td>
<td>55.8</td>
<td>36.5</td>
<td>4.59</td>
</tr>
<tr>
<td>Sub. 6</td>
<td>18</td>
<td>163.0</td>
<td>58.3</td>
<td>63.9</td>
<td>40.6</td>
<td>4.33</td>
</tr>
<tr>
<td>Sub. 7</td>
<td>21</td>
<td>165.0</td>
<td>56.2</td>
<td>60.8</td>
<td>39.0</td>
<td>5.34</td>
</tr>
<tr>
<td>Sub. 8</td>
<td>21</td>
<td>175.7</td>
<td>60.0</td>
<td>55.9</td>
<td>35.5</td>
<td>4.82</td>
</tr>
<tr>
<td>mean</td>
<td>20.1</td>
<td>170.4</td>
<td>57.0</td>
<td>58.3</td>
<td>36.8</td>
<td>4.80</td>
</tr>
<tr>
<td>S.D.</td>
<td>1.2</td>
<td>4.7</td>
<td>5.5</td>
<td>2.8</td>
<td>2.5</td>
<td>0.40</td>
</tr>
</tbody>
</table>

Mass: Body mass, \(\dot{\text{V}}\text{O}_2\) max: Maximal oxygen uptake per body mass, AT-\(\dot{\text{V}}\text{O}_2\): oxygen uptake per body mass at anaerobic threshold, \(\text{CO}_2\) excess - \(\Delta\text{La}\): \(\text{CO}_2\) excess per body mass per unit increase (from rest to 3 min after exhaustion) of \(\text{La}\) obtained in incremental exercise.

### Table 2 Mean values of excess \(\text{CO}_2\) output, measured and predicted blood lactate accumulations at three stages of constant exercise.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Stage I (n=8)</th>
<th>Stage II (n=7)</th>
<th>Stage III (n=5)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>mean</td>
<td>SD</td>
<td>mean</td>
</tr>
<tr>
<td>Ex (\text{CO}_2) (ml(\text{-kg}^{-1}))</td>
<td>15.36</td>
<td>8.51</td>
<td>34.15</td>
</tr>
<tr>
<td>(\Delta\text{La}_{\text{measured}}) (mmol(\text{l}^{-1}))</td>
<td>1.82</td>
<td>0.83</td>
<td>5.58</td>
</tr>
<tr>
<td>(\Delta\text{La}_{\text{predicted}}) (mmol(\text{l}^{-1}))</td>
<td>3.19</td>
<td>1.70*</td>
<td>7.09</td>
</tr>
<tr>
<td>Diff. (\Delta\text{La}) (mmol(\text{l}^{-1}))</td>
<td>1.38</td>
<td>1.08</td>
<td>1.50</td>
</tr>
</tbody>
</table>

Ex \(\text{CO}_2\): Excess \(\text{CO}_2\) output, \(\Delta\text{La}_{\text{measured}}\): Measured blood lactate accumulation, \(\Delta\text{La}_{\text{predicted}}\): Predicted blood lactate accumulation, Diff. \(\Delta\text{La}\): Difference between measured and predicted blood lactate accumulation. *: P<0.05, Significantly different from measured blood lactate accumulation.
Blood samples for the determination of La during the incremental exercise were withdrawn from a superficial dorsal hand vein every minute from 4th min until the exhaustion point, as well as during the resting and recovery period (at 3 min after exhaustion point), and during the constant exercise from an antecubital vein through an indwelling needle at rest and at the end of each stage of constant exercise (i.e., 4th, 8th and 12th min from the onset of constant exercise). Immediately after blood collection, the blood samples were deproteinized in cold perchloric acid, and then La was analyzed by an enzymatic method (Hadjivassiliou and Pieder, 1968).

The AT was detected by the gas exchange parameters ($\dot{V}_E$, $\dot{V}_E/\dot{VO}_2$ and $\dot{V}_E/\dot{VCO}_2$) and La (Davis et al., 1979; Wasserman et al., 1973). The criteria of the AT detection have been shown elsewhere (Hirakoba et al., 1992).

**Prediction of blood lactate accumulation during constant exercise**

An individual regression line of $\dot{VCO}_2/\dot{VO}_2$ relationship obtained in the incremental exercise test was used to calculate aerobically produced $\dot{CO}_2$ (aerobic $\dot{VCO}_2$) during constant exercise. Excess $\dot{CO}_2$ output (Ex $\dot{CO}_2$; measured as the integral of difference between the total $\dot{VCO}_2$ and aerobic $\dot{VCO}_2$) at each stage of constant exercise was estimated as shown in Fig. 1. Prediction of La accumulation ($\Delta La_{predicted}$) from rest to each stage of constant exercise was calculated from the following formula:

$$\Delta La_{predicted} = \text{Ex CO}_2\text{-mass}^-1 / \text{CO}_2 \text{ excess-}\Delta La$$

where Ex CO$_2$-mass$^{-1}$ = the volume of excess CO$_2$ output per body mass (ml·kg$^{-1}$) above that due to aerobic metabolism at each stage of constant exercise.

CO$_2$ excess-ΔLa = CO$_2$ excess per body mass per unit increase (from rest to 3 min after exhaustion) of La (ml·kg$^{-1}$·mmol$^{-1}$·l$^{-1}$) obtained in the incremental exercise test.

**Statistics**

Statistical significance of difference between the ΔLa$_{measured}$ (calculated as increase of La from rest to each stage of constant exercise) and ΔLa$_{predicted}$ at each stage of constant exercise was tested by a paired Student's t-test (two-tailed test) and Pearson product correlation was calculated in a correlation analysis. P<0.05 was accepted as significance level.

**Results**

As shown in Fig. 1, total $\dot{VCO}_2$ was found to be approximately equal to or lower than aerobic $\dot{VCO}_2$ at 1 min after the onset of stage I. Thereafter total $\dot{VCO}_2$ started to exceed aerobic $\dot{VCO}_2$.

Mean values (±SD) in Ex CO$_2$-mass$^{-1}$ (ml·kg$^{-1}$), ΔLa$_{measured}$ and ΔLa$_{predicted}$ at three stages of constant exercise are given in Table 2. Ex CO$_2$-mass$^{-1}$ was increased from stage I to stage III, which corresponded to the increases of ΔLa$_{measured}$ at three stages of constant exercise. A high, significant correlation coefficient (r=0.939, P<0.001) was obtained between Ex CO$_2$-mass$^{-1}$ and ΔLa$_{measured}$ (Fig. 2).

Figure 3 shows a relationship between ΔLa$_{measured}$ and ΔLa$_{predicted}$ at three stages of constant exercise. ΔLa$_{predicted}$ was found to correlate very closely to ΔLa$_{measured}$ (r=0.954, P<0.001), with a regression line of the standard error of estimate (SEE) of 1.47 mmol$^{-1}$·l$^{-1}$.

Mean values (±SD) in ΔLa$_{measured}$ and ΔLa$_{predicted}$ at three stages of constant exercise were 1.82 ± 0.83 vs 3.19 ± 1.70 for stage I, 5.58 ± 3.47 vs 7.09 ± 3.28 for stage II and 12.19 ± 2.36 vs 12.74 ± 1.83 mmol$^{-1}$·l$^{-1}$ for stage III, respectively (Table 2). There was a significant difference between ΔLa$_{measured}$ and ΔLa$_{predicted}$ at stage I (P<0.05), but no significant differences at stage II and III of constant exercise. The averaged difference from ΔLa$_{predicted}$ to ΔLa$_{measured}$ at stage III (0.55 mmol$^{-1}$·l$^{-1}$) showed a tendency to be smaller than stage I (1.38 mmol$^{-1}$·l$^{-1}$) and II (1.50 mmol$^{-1}$·l$^{-1}$). However, individual differences of
Relationship between excess CO₂ output per body mass and blood lactate accumulation (from rest) at three stages of constant exercise. For definitions see Table 2.

La predicted from La measured at three stages were changed between the stages even in the same subject, ranging from −1.71 to 4.05 mmol·l⁻¹ (Fig. 4).

Discussion

It has been indicated that the change of the CO₂ stores in the body during exercise enables the calculation of changing La without taking blood samples (Bouhuys et al., 1966; Clode et al., 1967; Isserlin and Rodahl, 1961). Clode and Campbell (1969) found a good agreement (r=0.97) between La measured and La predicted for steady state exercise and indicated that a quantitative change of La from CO₂ balance technique could be estimated, with an accuracy of within ±1 mmol·l⁻¹. However, it seems in their method to be necessary to follow the complicated procedures for the prediction of La accumulation. Thus they used an assumed muscle respiratory quotient (Rqm) of 0.90 at moderate and 0.95 at heavy exercise and slope of CO₂ dissociation curve of 1 ml·mmHg⁻¹·kg⁻¹ to estimate aerobic VCO₂ and the volume of CO₂ added to or removed from tissue fluids (respiratory CO₂). The aerobic VCO₂ could be estimated from the product of total VO₂ and assumed Rqm (VO₂ × Rqm), and respiratory CO₂ from changes in oxygenated mixed venous blood CO₂ pressure (PvCO₂) and slope of CO₂ dissociation curve of the body. For the measurement of PvCO₂, CO₂ rebreathing must be carried out at the beginning and the end of each stage of exercise. Moreover, it has been indicated that the Rqm was different among subjects even in the submaximal exercise below AT, ranging from 0.84 to 1.02 (Beaver and Wasserman, 1991) and the slope of CO₂ dissociation curve

![Graph showing relationship between excess CO₂ output per body mass and blood lactate accumulation.](image)

**Fig. 2**

![Graph showing relationship between predicted and measured blood lactate accumulation.](image)

**Fig. 3**

![Graph showing individual differences of blood lactate accumulation.](image)

**Fig. 4**
of the body would vary from 0.40 to 2.10 ml·mmHg$^{-1}$·kg$^{-1}$ (Parhi, 1964).

On the other hand, the prediction of La accumulation used in this study would be thought to be a simpler method, compared with that of Clode and Campbell (1969). A high, significant correlation coefficient between $\Delta$La$_{\text{measured}}$ and $\Delta$La$_{\text{predicted}}$ was found ($r=0.964$, $P<0.001$), with a standard error of estimate of 1.47 mmol$^{-1}$. This result is comparable to that of Clode and Campbell (1969), indicating that the changes of La accumulation could be predicted from Ex CO$_2$ during constant exercise and CO$_2$ excess - $\Delta$La obtained previously in incremental exercise test.

Beaver and Wasserman (1991) have indicated that the integral of excess $\dot{V}$CO$_2$ from AT to the end of an incremental exercise, which is excess volume of CO$_2$ due to bicarbonate buffering of lactic acid ($\dot{V}$CO$_2$$_{2at}$), is a good estimator of total lactate accumulation and of bicarbonate depletion. Similarly, Yano (1987) defined the integral of excess $\dot{V}$CO$_2$ during incremental exercise as "CO$_2$ excess". It has in fact, been reported that CO$_2$ excess per body mass (CO$_2$ excess-mass$^{-1}$) correlated significantly with La accumulation and decrease of blood bicarbonate (Hirakoba et al., 1993b; Yano 1987). Furthermore, CO$_2$ excess-mass$^{-1}$ per unit increase of La (CO$_2$ excess - $\Delta$La)$_{\text{measured}}$, which would imply ml CO$_2$ excess expired per mmol increase of La, has been indicated to vary among subjects due to fitness level and training specificity (Inaki et al., 1993; Hirakoba et al., 1990, 1992; Yano, 1987), but to remain constant in the same subject in spite of $\Delta$La fluctuations due to exercise and change in blood bicarbonate concentration (Hirakoba et al., 1993b). Therefore, if individual CO$_2$ excess - $\Delta$La was obtained previously, it would be possible to predict La accumulation from Ex CO$_2$ during constant exercise which could be estimated from the difference of total $\dot{V}$CO$_2$ and aerobic $\dot{V}$CO$_2$.

However, the differences from $\Delta$La$_{\text{predicted}}$ to $\Delta$La$_{\text{measured}}$ in all stages of constant exercise varied significantly among subjects, ranging from -1.71 to 4.05 mmol$^{-1}$. This prediction error of La accumulation could not be accounted for either by subject's specificity or by difference of work rate, as shown in Fig. 4. It is, therefore, important to clarify the factor(s) which could result in the variation of the difference between the measured and predicted values of La accumulation observed in this study.

There was a significant difference between $\Delta$La$_{\text{measured}}$ and $\Delta$La$_{\text{predicted}}$ in the first stage as shown in Table 2. It is inferred from this result that the La prediction method in this study would not be sensitive enough to differentiate between the aerobic $\dot{V}$CO$_2$ and excess CO$_2$ output in stage I (100% of AT) at which muscles are beginning to depend more on anaerobic glycolysis. In addition, another possible mechanism for the greater variation in the La prediction in the first stage is the difference of efflux rate in CO$_2$ and La from muscles to blood in stage I, because intramuscular bicarbonate is indicated to be the major source of excess CO$_2$ output (Beaver and Wasserman, 1991). Therefore, it is anticipated that the greater variation of the prediction error in stage I would be caused by the faster efflux of CO$_2$ than of La from muscles to blood. However, this assumption could not account for a phenomenon that aerobic $\dot{V}$CO$_2$ (calculated by a regression line for $\dot{V}$CO$_2$-$\dot{V}$O$_2$) exceeds total $\dot{V}$CO$_2$ in the early phase of stage I (Fig. 1). It has been pointed out that some part of CO$_2$ produced by aerobic metabolism is retained in the body, resulting in the transient decrease of respiratory exchange ratio (R) within 1 min after onset of exercise.

On the other hand, the calculation of aerobic $\dot{V}$CO$_2$ during constant exercise in this study was made by the regression line for $\dot{V}$CO$_2$-$\dot{V}$O$_2$ in incremental exercise for estimating Ex CO$_2$ (measured as the difference between total $\dot{V}$CO$_2$ and aerobic $\dot{V}$CO$_2$) derived from bicarbonate buffering of lactic acid. Yano (1987) has reported that at the same metabolic rate ($\dot{V}$O$_2$), $\dot{V}$CO$_2$ during steady state exercise was higher than that of incremental exercise and the regression line for $\dot{V}$CO$_2$-$\dot{V}$O$_2$ of steady state exercise was different from incremental exercise. According to this finding, the aerobic $\dot{V}$CO$_2$ during constant exercise estimated from the regression line for $\dot{V}$CO$_2$-$\dot{V}$O$_2$ of incremental exercise might be underestimated, which would be associated with the overestimation of excess CO$_2$ output. Moreover, it is suggested that a small part of Ex CO$_2$ estimated in the present method must be attributed to respiratory compensation for metabolic acidosis (Beaver and Wasserman, 1991; Zhang et al., 1994; Wasserman et al., 1981). In fact, $\Delta$La$_{\text{predicted}}$ showed a tendency to be higher than $\Delta$La$_{\text{measured}}$ in all stages of constant exercise (Table 1). As a consequence, it may be likely that the difference in the regression line for $\dot{V}$CO$_2$-$\dot{V}$O$_2$ between both types of exercise and the volume of CO$_2$ released by hyperventilation due to metabolic acidosis could vary among subjects, which would lead to subject to subject variation of prediction error in La accumulation during constant exercise.

In summary, the present study suggests that the changes of La accumulation during exercise above the AT level could be predicted from excess CO$_2$ output generated in constant exercise. However, further studies should be carried out to determine the mechanism(s) in more detail as to why the prediction error of La accumulation occurs and to reexamine the validity of this method.

Acknowledgment. The authors gratefully acknowledge the financial support of Kagoshima Keizai University.
References


Received: January 17, 1996
Accepted: June 7, 1996
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