Skeletal Muscle Energetics, Acid-Base Equilibrium and Lactate Metabolism in Patients with Severe Hypercapnia and Hypoxemia*

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Quadriiceps femoris muscle needle biopsies were performed in ten patients with chronic obstructive pulmonary disease and acute respiratory failure and in ten age- and sex-matched healthy control subjects. The main indices of skeletal muscle cell energy metabolism, intracellular acid-base equilibrium and lactate metabolism were evaluated. Reduced ATP and phosphocreatine content, intracellular acidosis related to hypercapnia, increased muscle lactate without alterations of the muscle lactate concentration gradient were observed in the skeletal muscle of the hypercapnic-hypoxic COPD patients studied, in which group no correlation was found between hypoxia and energy or lactate metabolism parameters. These results suggest that an overall derangement of cell energy metabolism and acid-base equilibrium is present in severely hypercapnic-hypoxic chronic obstructive pulmonary disease and that in this condition skeletal muscle seems to metabolize anaerobically—even though, in addition to hypoxia, other factors interfering with both cell energy and lactate metabolism are likely to be present.

Very scant information on cell metabolism of patients with both hypercapnia and hypoxemia is presently available.

Both low adenosine 5’-triphosphate (ATP) and phosphocreatine levels and decreased intracellular pH have been observed in the skeletal muscles of patients with chronic obstructive pulmonary disease (COPD) and acute respiratory failure (ARF).

However, the problem of relationships between the presence of a hypercapnic-hypoxic condition and the consequent derangements of cell energetics, acid-base equilibrium and lactate metabolism has not yet been raised.

The present investigation focuses on a parallel evaluation of cell energy metabolism, intracellular acid-base equilibrium, and lactate metabolism in the skeletal muscle of a group of severely hypercapnic-hypoxic patients.

This was made by means of a direct biochemical study on quadriiceps femoris muscle specimens obtained through needle biopsy technique in patients with COPD and ARF.

**Materials and Methods**

**Patients**

Ten COPD patients (nine men and one woman) who ranged in age from 52 to 75 yr (mean, 62 ± 3 yr SEM) were admitted to a Pulmonary Intensive Care Unit for ARF. Blood gas measurements were as follows: arterial carbon dioxide tension (PaCO₂), 72 ± 4 mm Hg SEM, range, 54 to 99 mm Hg; arterial oxygen pressure (PaO₂), 38 ± 2 mm Hg SEM, range, 28 to 50 mm Hg; pHa, 7.32 ± 0.02 SEM, range, 7.25 to 7.37.

All patients had had an acute exacerbation of their underlying disease (two to eight days prior to admission), which was mainly due to acute bronchial or pulmonary infection.

Diagnosis of COPD, based on a positive history, clinical criteria, and standard measurements of pulmonary mechanics, had been made during previous hospitalizations. All patients had a history of chronic bronchitis (lasting from 14 to 30 yr) and dyspnea (exertional or at rest or both). Forced expiratory volume in one second (FEV₁) and the FEV₁ forced vital capacity ratio were less than 60 percent of predicted standard values.

Ten age- and sex-matched subjects (eight men, two women) who ranged in age from 40 to 79 yr (mean, 61 ± 4 yr SEM) and who did not have a history of lung disease and had normal blood gas analysis and lung function were utilized as controls.

All subjects and their next of kin, or both, were informed about the nature and the possible risks of the study; consent was obtained from each participant or relatives, or both.

**Experimental Procedures**

In all patients and control subjects, three muscle samples weighing 30 to 60 mg were obtained from the lateral portion of the same quadriiceps femoris muscle by the needle biopsy technique according to Bergström.¹

In COPD patients, muscle biopsies were taken while they were breathing air within two hours after admission and immediately before any treatment was started; in control subjects, muscle samples were obtained in the morning after an overnight rest.

Immediately before biopsy, blood samples from the ipsilateral femoral artery and vein were anaerobically drawn into heparinized syringes, and then analyzed for pH, partial oxygen pressure (PO₂), carbon dioxide partial pressure (PCO₂), bicarbonate, chloride, and total proteins.

**Analytical Procedures**

Blood. Arterial and venous pH, PCO₂, and PO₂ were measured by a radiometer AB1.1 pH meter gas analyzer (Radiometer, Copenhagen). Plasma bicarbonate concentrations were calculated with an
Skeletal HCO₃, <0.001 NS

Muscle. Immediately after needle biopsies, muscle samples rapidly were dissected free from any visible fat or connective tissue. One sample was used for muscle water compartment (extracellular and intracellular water, H₂O, and HCO₃ determination according to the chloride method, as previously described. Two samples were frozen within six to seven s in Freon 22, stored under liquid nitrogen, and then analyzed for intracellular acid-base equilibrium indexes and energy metabolism parameters, respectively.

Intracellular bicarbonate (HCO₃) and pH (pH) were obtained from total muscle CO₂ (TCO₂) measured by the acid-labile CO₂ method, as previously described in detail.

In brief, intracellular bicarbonate values were obtained by deducting from muscle TCO₂ content, measured by means of potentiometric dead-stop end-point titrations, the amount of bicarbonate found in the extracellular water compartment and the amount of CO₂ physically dissolved in both intracellular and extracellular muscle water compartments. In order to calculate pH, HCO₃ values (expressed as millimoles per liter of intracellular water) were introduced into the Henderson-Hasselbalch equation, using a pKₐ value of 6.14 for H₂CO₃ in intracellular water and a solubility coefficient of 0.6777 mmol CO₂/mm Hg of Pco₂ for CO₂ in the same compartment.

Muscle samples intended for energy metabolism parameter determinations were freeze-dried and then extracted with perchloric acid. Muscle ATP, adenosine 5'-diphosphate (ADP), adenosine 5'-monophosphate (AMP), phosphocreatine (PCr), creatine (Cr), and total muscle lactate (Lactₜ) were measured according to spectrophotometric-enzymatic techniques, and expressed in millimoles per kilogram of dry muscle weight (dmw). The total adenine nucleotide pool (TAN) (ie, the sum of ATP, ADP, AMP = TAN) and energy charge potential (ECP) (ECP = ATP + 0.5 ADP/ATP + ADP + AMP) also were calculated.

Intracellular lactate concentration (Lactₜ [expressed in millimoles per liter of intracellular water]) was calculated from muscle and femoral vein plasma lactate (Lactᵥ) values, as well as from muscle water compartment values, according to the following equation:

\[ \text{Lactₜ} = \frac{\text{Lactᵥ} - \text{Lactᵥ}_{\text{H₂O}}}{\text{H₂O}} \]

where Lactₜ = extracellular lactate concentration (millimoles per liter of extracellular water) calculated by correcting plasma lactate (Lactᵥ) for plasma water content (H₂Oₜ, H₂Oₜ = 0.984 - 7.18, ie, H₂Oₜ = 0.984 - 7.910⁻³ g of prot per liter of plasma), H₂Oₜ and H₂Oₜ, both calculated as elsewhere previously described. The muscle lactate concentration gradient (as intracellular lactate-extracellular lactate ratio) also was calculated.

| Table 1—Muscle Energy Metabolism and Intracellular Acid-Base Equilibrium indexes* |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
|                 | PCr†            | Cr†             | TCr†            | ATP†            | ADP†            | AMP†            | ECP†            |
| COPD patients   | 49.02±11.07     | 66.50±21.06     | 115.60±23.76    | 16.77±2.09      | 3.37±0.66       | 0.24±0.12       | 20.61±2.08      | 5.14±1.27       | 0.889±0.024     | 6.77±0.23       | 14.09±5.08      |
| n = 10          |                 |                 |                 |                 |                 |                 |                 |                 |                 |                 |                 |
| Control subjects| 76.87±11.05     | 51.80±11.05     | 127.68±17.26    | 23.76±2.09      | 2.94±0.49       | 0.14±0.08       | 26.84±1.10      | 8.36±1.33       | 0.937±0.013     | 7.03±0.07       | 12.50±1.62      |
| n = 10          |                 |                 |                 |                 |                 |                 |                 |                 |                 |                 |                 |
| Mann-Whitney test | <0.001 <0.05 | <0.001 NS       | <0.001 NS       | NS              | <0.001 <0.001   | <0.001 <0.001   | NS              | NS              | NS              | NS              | NS              |

*Data are expressed as mean ± SD. PCr, phosphocreatine; Cr, creatine; TCr, total creatine; ATP, adenosine 5'-triphosphate; ADP, adenosine 5'-diphosphate; AMP, adenosine 5'-monophosphate; TAN, total adenine nucleotide pool; ECP, energy charge potential; pH, intracellular pH; HCO₃, intracellular bicarbonate; H₂Oₜ, intracellular water.

*Measured in mmol/kg/dry muscle weight.

Statistics

The Mann-Whitney test was used to assess the statistical significance of differences between control subjects and COPD patients.

Standard techniques of linear regression and correlation also were utilized.

Results

Table 1 shows energy metabolism parameters and intracellular acid-base equilibrium indices of COPD patients with ARF.

A marked decrease of both muscle ATP and PCr contents as well as a significant decrease in TAN, the ATP-ADP ratio and ECP were found. No differences in ADP, AMP, or total Cr content were found between patients and controls.

Intracellular pH values were significantly (P < 0.001) reduced and correlated to PaCO₂ values (pH = 7.76 - 0.038 PaCO₂, r = 0.77, n = 10, P < 0.01), but not to arterial Po₂ (PaO₂) values, whereas HCO₃ was not significantly different from the control values. No significant relationships between the main parameters of energy metabolism and pH were found within each group of subjects studied (COPD patients, n = 10: pH/PCr, r = 0.196 NS; pH/ATP, r = 0.425 NS; pH/ECP, r = -0.002 NS. Control subjects, n = 10: pH/PCr, r = 0.355 NS; pH/ATP, r = 0.383 NS; pH/ECP, r = 0.183 NS).

Table 2 shows lactate metabolism parameters: muscle and plasma lactate both were increased with no differences from the control values for Lactₜ, concentration and the muscle lactate gradient. No relationship was found between muscle and Lactᵥ values and PaO₂. At any given low PaO₂ value, a wide scattering of muscle lactate values was found (Fig I). In six patients, muscle lactate values fell within the control range. No correlation was found between pHₜ and lactate gradient (r = 0.42, n = 10, NS).

Discussion

Low pHₜ reduced ATP and PCr contents, and increased muscle and Lactᵥ values characterize the

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Table 2— Muscle Lactate Metabolism Indexes

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<tr>
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<th>Lactₐ (mmol/kg dry muscle weight)*</th>
<th>Lactₑ (mmol/L plasma)†</th>
<th>Lactᵦ (mmol/L NaH₂O₄)‡</th>
<th>Lactᵦ (mmol/L NaH₂O₈)§</th>
<th>Lactate Gradient</th>
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<tbody>
<tr>
<td>COPD patients</td>
<td>15.41 ± 7.31</td>
<td>1.89 ± 1.16</td>
<td>2.01 ± 1.23</td>
<td>4.38 ± 2.01</td>
<td>2.52</td>
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<td>(n = 10)</td>
<td></td>
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<tr>
<td>Control subjects</td>
<td>8.59 ± 2.58</td>
<td>0.83 ± 0.12</td>
<td>0.89 ± 0.13</td>
<td>2.79 ± 0.88</td>
<td>3.13</td>
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<tr>
<td>(n = 10)</td>
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<tr>
<td>Mann-Whitney test</td>
<td>&lt;0.01 &lt;0.05</td>
<td>&lt;0.05</td>
<td>NS</td>
<td>NS</td>
<td></td>
</tr>
</tbody>
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*Data are expressed as mean ± SD. Total muscle lactate.
†Femoral vein plasma lactate.
‡Extracellular lactate.
§Intracellular lactate.

Skeletal muscle of the hypercapnic-hypoxemic COPD patients considered in our study.

The finding of an intracellular acidosis related to PaCO₂ confirms the results of a previous study on a comparable group of subjects with COPD and ARF.²

The extent of the decrease of ATP and PCr content (about 30 to 35 percent of control values) is comparable to that found in subjects with cardiogenic shock or severe congestive heart failure¹¹ and in patients with severe acute respiratory or circulatory insufficiency.¹²

Among the factors likely to result in an altered muscle cell energy metabolism, hypoxia is credited with a major role. In fact, in the COPD patients studied, the low values found for ATP, PCr, and ATP-ADP ratio, as well as the increased muscle and plasma lactate values could be regarded as a consequence of the hypoxia-related derangements of mitochondrial oxidative function in skeletal muscle.

Yet no significant relationship between cell energy metabolism and PaO₂ values was found. Therefore, other factors, in addition to the effects of the low PaO₂ levels on mitochondrial ATP production, should be taken into consideration in order to explain our findings in COPD patients with ARF.

As a matter of fact, the regeneration of ATP stores is dependent not only on ADP rephosphorylation, but also on the availability of new adenine nucleotides.¹²,¹³

Low ATP levels, as found in this group of patients, could thus be attributed to a disrupted balance between degradation and resynthesis of adenine nucleotides. Both hypoxia and acidosis are known as activating ATP-degrading sequences; in particular, AMP deaminase is activated by low pH values, reduced oxygen availability, and low ATP values.¹³

Furthermore, lowering of the skeletal muscle adenine nucleotide pool, along with low PCr values, has been observed in a study on malnourished patients.¹⁴ A reduced content of both ATP and PCr under such conditions has been attributed to a lack of precursors and calories due to reduced nutrient intake.

The nutritional status of the COPD patients studied here has not been evaluated. Nonetheless, there is now increasing evidence that malnutrition is quite common among COPD patients with ARF¹⁵,¹⁶ and that in these patients the severity of nutritional depletion is related to the severity of the respiratory illness as judged from need for hospitalization and degree of impairment of gas exchange.¹⁷

Figure 1. Relationship between muscle lactate content and arterial PaO₂ (Pao₂) values in COPD patients with ARF.
Several aspects of muscle lactate metabolism in the COPD patients considered in the present study should be emphasized.

The wide scattering of muscle lactate values and the lack of correlation between muscle lactate and PaO₂ values, at low PaO₂ levels, point to the presence of factors interfering with muscle lactate metabolism response to hypoxia; in particular, of factors interfering with O₂ delivery and of acidosis itself. It is well known that a reduction of arterial blood O₂ levels does not necessarily correlate with the degree of tissue hypoxia, since O₂ availability for cells also is influenced by systemic and local factors mainly regulating O₂ exchange and microcirculation.  Although these factors, eg, cardiac output and its distribution of local blood supply, were not measured in our COPD patients with ARF, one cannot exclude that they are altered in such severely ill patients.

In spite of severe hypoxemia, in several patients Lact₄₀ levels were normal. Muscle lactate levels also were normal in these patients; moreover, lactate gradient values were not significantly different from controls. Thus, even if an increase of extracellular H⁺ concentration has been reported as reducing lactate efflux from muscle, the presence of acidosis does not seem to have affected lactate efflux from muscle cells in the COPD patients considered here.

Nor can one rule out the possibility that acidosis could have exerted a negative effect on glucose metabolism, for low pH, is known as determining a severe inhibition of key glycolytic enzymes phosphorylase and PFK, at least in vitro. Therefore, one can surmise that, in some of the COPD patients in question, notwithstanding their severe hypoxemia, the effects of hypcapnia-related intracellular acidosis on glycolytic sequences have limited the expected increase of muscle lactate production in response to reduced O₂ availability for cells.

The negative feedback exerted by H⁺ on lactate production has recently been regarded as an important homeostatic mechanism of "metabolic buffering" and as part of a more general homeostatic system that regulates endogenous organic acid production via changes in systemic pH. A marked suppression of exercise-induced hyperlactatemia by hypercapnia also has been ascertained in man.

The role of such negative feedback in the pathogenesis and evolution of clinical lactic acidosis is still unclear especially in patients with both hypcapnia-related intracellular acidosis and hypoxemia.

On the other hand, the same servomechanism can be overridden in cases of severely hypoxemic lactic acidosis, and one cannot exclude that this applies to some of our patients in whom increased muscle lactate levels could indicate that the inhibitory effect of acidosis is overridden by the hypoxia-drive to increased lactate production.

In summary, data concerning some aspects of skeletal muscle cell metabolism in the course of severe COPD have been presented here. In particular, the presence of low high-energy phosphate compound content values, hypercapnia-related intracellular acidosis, and increased muscle and femoral vein plasma lactate has been demonstrated.

These findings suggest that an overall derangement of cell energy metabolism and acid-base equilibrium is present in severely hypercapnic-hypoxicemic patients with COPD and ARF. They also suggest that in the same patients skeletal muscle is metabolizing anaerobically, even though, in addition to hypoxia, other factors interfering with cell energy and lactate metabolism are likely to be present.

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