**Catabolism in Skeletal Muscle**  
**The Phosphagen System**

- **Overview of ATP Regeneration**
- **Anaerobic vs Aerobic Metabolism**
- **Creatine Kinase Reaction**
- **Adenylate Kinase Reaction**
- **Purine Nucleotide Cycle**
- **Creatine Phosphate Shuttle**
- **\(^{31}P\) MRS and Muscle Metabolism**

## ATP - energy currency of cell

Muscle contraction can increase the cellular demand for ATP 100-fold! Resting [ATP] of 8 mmol/kg could be depleted in 2-3s of intense exercise!

The design and function of skeletal muscle metabolism is to meet this ATP demand as well as possible.

Skeletal muscle has sensitive biochemical controls of metabolic pathways involving the sudden *activation* and *inhibition* of specific enzymes.
ATP Regeneration

Skeletal muscle can produce the ATP required to support muscle contraction from one or a combination of three metabolic reactions / pathways;

1. **Phosphagen System** - forming ATP from using creatine phosphate or two ADP molecules

2. **Glycolysis** - from blood glucose or muscle glycogen

3. **Mitochondrial Respiration** - the use of oxygen in the mitochondria

![% Energy System Contribution](chart.png)
Anaerobic vs Aerobic Metabolism...
Old Terminology!

Anaerobic metabolism - does not require the presence of oxygen - creatine kinase & adenylate kinase reactions, and glycolysis.

Aerobic metabolism - the combined reactions of mitochondrial respiration - pyruvate oxidation, the TCA cycle, and the electron transport chain.

These terms are not entirely accurate and it is inappropriate to differentiate the pathways as two extremes when they actually share a common central pathway (e.g., glycolysis) and occur simultaneously!

The Phosphagen System

The regeneration of ATP via the transfer of phosphate groups through either of two reactions:

1) Creatine Kinase Reaction (aka CrP reaction)

2) Adenylate Kinase Reaction

The creatine kinase reaction is the most immediate means to regenerate ATP. Why?

\[
\text{CrP} + \text{ADP} + H^+ \xrightleftharpoons{\text{creatine kinase}} \text{ATP} + \text{Cr}
\]

Immediate changes in ADP and ATP concentrations!
The Phosphagen System, cont’d

Note: The creatine kinase reaction is actually two separate reactions that are “coupled” together.

\[
\begin{align*}
\text{exergonic} & : & \text{CrP} & \rightarrow & \text{Cr} + \text{Pi} & + & \text{ADP} + \text{H}^+ + \text{Pi} & \rightarrow & \text{ATP} \\
\text{“endergonic”} & : & \text{CrP} + \text{ADP} + \text{H}^+ & \rightarrow & \text{ATP} + \text{Cr}
\end{align*}
\]

Consumes H+ which helps delay acidosis!

The Phosphagen System, cont’d

The ATP production capacity of the creatine kinase reaction relies on a store of CrP, ~26 mmol/kg wet wt. CrP content is slightly higher in fast twitch than in slow twitch muscle fibers.

Location of skeletal muscle creatine kinase:
- 4% on the outer mitochondrial membrane - essential for creatine phosphate shuttle
- 3% bound to myofibrillar proteins of sarcomere
- remainder free in cytosolic solution
Increased Activity

At rest, the reaction is at equilibrium, therefore, the $\Delta G \sim 0$.

\[
\begin{array}{c}
\text{CrP} + \text{ADP} + \text{H}^+ \\
\text{26 mmol/kg} \\
\text{1.0 x 10}^{-4} \text{mmol/kg}
\end{array}
\rightarrow
\begin{array}{c}
\text{ATP} + \text{Cr} \\
\text{8.0 mmol/kg} * \\
\text{4.0 mmol/kg}
\end{array}
\]

In other words, the substrate and product concentrations are approximately constant and therefore there is no net release of free energy.

* expressed mmol/kg wet wt in solution of cytoplasm

Activity, cont.

During intense exercise, the reaction is pushed / pulled to the right, thereby breaking down CrP and forming ATP. $\Delta G \sim -9.12$

\[
\begin{array}{c}
\text{CrP} + \text{ADP} + \text{H}^+ \\
\text{expressed mmol/kg}
\end{array}
\rightarrow
\begin{array}{c}
\text{ATP} + \text{Cr} \\
\text{expressed mmol/kg}
\end{array}
\]

Initially, exercise results in an $\uparrow$ in substrate concentrations (ADP and H\(^+\)) and a $\downarrow$ in product concentrations (ATP).
Dr. Robert Robergs  

**Phosphagen System**

The concentrations of molecules that are important in energy metabolism within skeletal muscle at rest and after intense exercise to fatigue are shown in Table 2.3.

<table>
<thead>
<tr>
<th>MOLECULE</th>
<th>REST (mmol/kg wet wt)</th>
<th>FATIGUE (mmol/kg wet wt)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CrP</td>
<td>24.0</td>
<td>3.0</td>
</tr>
<tr>
<td>ATP</td>
<td>5.0</td>
<td>4.5</td>
</tr>
<tr>
<td>ADP</td>
<td>0.05</td>
<td>0.5</td>
</tr>
<tr>
<td>Cr</td>
<td>4.0</td>
<td>25.0</td>
</tr>
<tr>
<td>Pi</td>
<td>3.0</td>
<td>24.0</td>
</tr>
<tr>
<td>H+</td>
<td>$1.0 \times 10^{-4}$</td>
<td>$4 \times 10^{-3}$</td>
</tr>
<tr>
<td>Lactate</td>
<td>1.0</td>
<td>25.0</td>
</tr>
<tr>
<td>Glycogen</td>
<td>200.0</td>
<td>75.0</td>
</tr>
</tbody>
</table>

Data are from references 3, 4, 6, 7, and 11.

* These concentrations apply to molecules in solution in the cytoplasm of the cell. For a concentration unit applicable for precise bioenergetic calculations, the metabolites would be expressed relative to muscle water, which is estimated at approximately 78% of wet weight muscle, hence increasing the concentration shown by a factor of 1.3.
CrP also supplements ATP regeneration from mitochondrial respiration

Note the increased reliance on “anaerobic” sources of ATP regeneration as exercise intensity increases.

The Phosphagen System, cont’d

The adenylate kinase reaction is similar to the creatine kinase reaction in that it is near equilibrium at rest.

\[ \Delta G = 0 \]

\[
\begin{align*}
0.05 \text{ mmol/kg} & \quad \text{ADP} + \text{ADP} \quad \text{adenylate kinase} \quad 5.0 \text{ mmol/kg} \\
& \quad \text{ATP} + \text{AMP}
\end{align*}
\]

In other words, the substrate and product concentrations are approximately constant and therefore there is no net release of free energy.
Changing activity

During intense exercise, the reaction is pushed/pulled to the right, thereby increasing the production of an allosteric activator (AMP).

\[
\text{ADP + ADP} \xrightarrow{\text{adenylate kinase}} \text{ATP + AMP}
\]

Exercise results in an \(\uparrow\) in substrate concentrations (ADP) and a \(\downarrow\) in product concentrations (ATP).
Adenosine monophosphate (AMP) is the activator of the allosteric enzymes phosphorylase (glycogenolysis) and phosphofructokinase (glycolysis), thus stimulating increased carbohydrate catabolism and ATP regeneration.

However, continued increases in AMP would decrease the phosphorylation potential of the cell which is detrimental to the cell.

Phosphorylation Potential = [ATP] / ([ADP] [AMP] [ATP])

Purine Nucleotide Cycle

The conversion of AMP to IMP via the AMP deaminase catalyzed reaction.

\[
\text{AMP deaminase} \\
\text{AMP + H}_2\text{O + H}^+ \rightarrow \text{IMP + ammonia (NH}_4) \\
\downarrow \\
\text{Toxic to cell and CNS and is shunted into circulation for liver metabolism, excretion by kidney, or sweat loss.}
\]

The reaction is stimulated during acidic conditions (↑ H⁺ ) and is reversed during recovery.
The creatine kinase and adenylate kinase reactions that cause rapid (ATP) regeneration in the vicinity of the contractile proteins.

\[ \text{H}^+ + \text{ADP} + \text{CrP} \rightleftharpoons \text{ATP} + \text{Cr} \]

2 ADP $\rightleftharpoons$ ATP + AMP

Contractile proteins

Sarcolemma

Mitochondria
The phosphagen system can regenerate ATP at high rates.

However, the finite store of CrP can be depleted in as little as 10 seconds.

<table>
<thead>
<tr>
<th>REACTION/PATHWAY</th>
<th>MAXIMAL RATE (mmol ATP/kg wet wt)</th>
<th>TIME</th>
<th>$O_2$ (mmol $O_2$/ATP)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CrP</td>
<td>2.4</td>
<td>&lt;1.0 sec</td>
<td>0.0</td>
</tr>
<tr>
<td>Glucose ↔ Lactate</td>
<td>1.3</td>
<td>&lt;5.0 sec</td>
<td>0.0</td>
</tr>
<tr>
<td>Glucose ↔ CO$_2$ + H$_2$O</td>
<td>0.7</td>
<td>3.0 min</td>
<td>0.167</td>
</tr>
<tr>
<td>FFA ↔ CO$_2$ + H$_2$O</td>
<td>0.3</td>
<td>30.0 min</td>
<td>0.177</td>
</tr>
</tbody>
</table>
Fig. 1. A schematic representation of the translocation of high energy phosphate between the sites of its production and its use. See text for a description and evidence for each of the reactions indication by the Roman numerals.
Magnetic Resonance Imaging and Spectroscopy

1. Magnetic field and high frequency radiofrequency energy input

2. Rf energy displaces atomic electrons, causing energy to be released on their return to a normal axis

3. Energy output from nuclei is acquired, and the frequency of the signal is specific to the atom of interest

(31P MRS)
Whole body NMR Imager

Main medical applications of NMR is for non-invasive imaging of tissues within the body.

A $^1$H image of the lower leg

The higher the proton density, the brighter the image on an MR scan.
Spectroscopy

Magnetic field and high frequency radiofrequency energy input

\[
\delta = \text{Pi-PCr}
\]

\[
pH = 6.75 + \log_{10} \left[ \frac{(8-3.35)}{(5.60-8)} \right]
\]

**Figure 4**—The measurement of intra-cellular muscle pH with \(^{31}\)P NMR spectroscopy.
Phosphagen System

Importance of $^{31}$P MRS Research
Since the CrP reaction involves free protons, the greater the acidosis, the longer the recovery period.

Muscle CrP recovery reveals a dual exponential curve having a fast and slow component.

The fast component is complete within 2 min and represents ~ 80-90% of complete CrP recovery.