

This student paper was written as an assignment in the graduate course

Free Radicals in Biology and Medicine

(77:222, Spring 2005)

offered by the

Free Radical and Radiation Biology Program

B-180 Med Labs

The University of Iowa

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Spring 2005 Term

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Lactate – a potential antioxidant

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For 77:222, Spring 2005

February 24, 2005

Abbreviations:

ADP, adenosine 5'-diphosphate; ATP, adenosine 5'-triphosphate; DMPO, 5,5-dimethylpyrroline-*N*-oxide; FAD, flavin adenine dinucleotide; HO \cdot , hydroxyl radical; MDA, malondialdehyde; Mn, manganese; NAD $^{+}$, nicotinamide adenine dinucleotide; NADH, nicotinamide adenine dinucleotide (reduced form); ROS, reactive oxygen species; O $_2^{\cdot-}$, superoxide; SOD, superoxide dismutases.

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Abstract

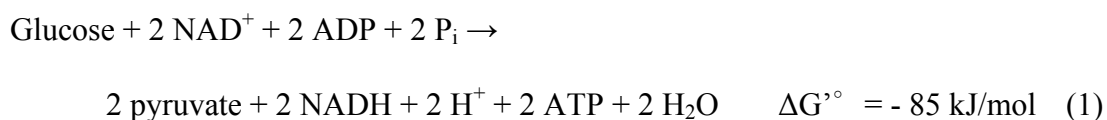
During extreme muscular activity, lactate is produced as well as reactive oxygen species resulting in oxidative stress. Data from the literature show diverse opinions about the prooxidant and antioxidant effect of lactate. Here we discuss the lactate formation, recycling, antioxidant effect in scavenging superoxide, hydroxyl radical, and attenuating lipid peroxidation. Transition metal, manganese is capable to scavenge superoxide and lactate probably can be a reductant to recycle the oxidized manganese.

Introduction

Lactate is produced during short bursts of extreme muscular activity or hypoxia. Oxidative stress and production of reactive oxygen species (ROS) are also generated during physical exercise [1, 2]. These two pieces of evidence pose the following question: what is the connection between lactate and ROS? The role of lactate could be pro-oxidant, antioxidant, or both. Rehncrona et. al. showed that lactic acidosis has a pro-oxidant effect in brain homogenates [3]. Lovlin observed a significant relationship between plasma lactate concentration and lipid peroxidation [4]. On the other hand, Anbar et. al. reported lactate ion as an antioxidant, moderate HO[•] scavenger at pH 9 [5]. Here the lactate formation, recycling, physical properties and evidences about its antioxidant effects and possible mechanisms will be discussed.

Lactate formation

Glucose is the major fuel for most organisms. In aerobic organisms, the complete oxidation of glucose to carbon dioxide and water releases 2840 kJ mol⁻¹ of energy. The first stage of glucose oxidation is glycolysis. In glycolysis, a molecule of glucose is degraded to yield two molecules of pyruvate. The overall reaction of glycolysis is [6]

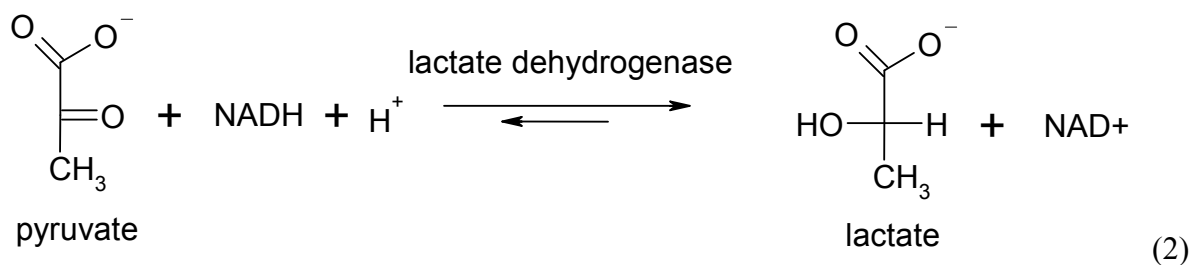


Pyruvate is degraded into Acetyl-CoA and then enters the citric acid cycle to yield NADH and FADH₂. Products of citric acid cycle, NADH and FADH₂ enter mitochondrial electron

transport chain, which uses oxygen as the final electron acceptor and produces the majority of ATP released in the glucose catabolism. Glycolysis uses NAD^+ instead of oxygen as the electron acceptor. The energy released in glycolysis is relatively small as compared to the total energy released by complete oxidation.

During extreme muscular activity, the rapid increase in energy demand is supplied by glycolysis [7]. First, the acceleration of glycolysis is fast as compared to the oxidative pathway. Second, the maximal glycolytic capacity of muscle exceeds the maximal oxidative capacity.

In glycolysis, the NAD^+ pool is consumed as electron acceptor and it must be recycled. Recycling of NAD^+ is done by converting pyruvate and NADH to lactate and NAD^+ during extreme muscular activity. This reaction is catalyzed by lactate dehydrogenase.



Lactate recycling

Skeletal muscle is the major producer of lactic acid in the body [6]. Lactate produced in muscle can exchange rapidly between muscle, blood and liver [7, 8]. Lactate in liver will be converted into glucose. This cycle of glucose-lactate-glucose is called the Cori cycle[8].

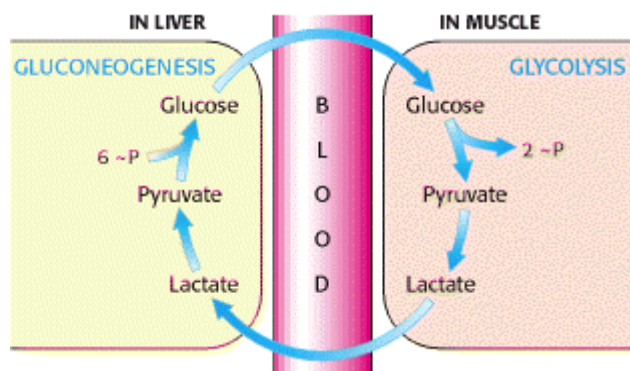
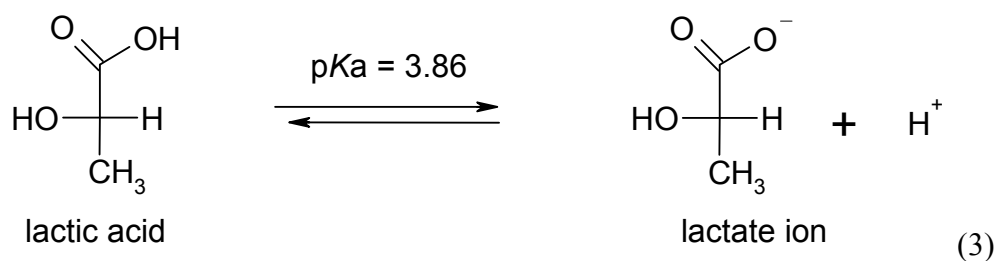


Figure 1, the Cori Cycle. Lactate formed by active muscle is converted into glucose by the liver. Adapted from [6].

Physical properties of lactic acid

Lactate ion is the major form of lactic acid under physiologic environment (pH around 7).

Lactic acid has the formula: $C_3H_6O_3$, its molecular weight is 90.08 and CAS Registry Number is 598-82-3.



Antioxidant effect

Lactate ion might be considered as a potential antioxidant agent. Carole et. al. suggested that lactate ion may scavenge free radicals such as O_2^\cdot (**figure 2**), HO^\cdot (**figure 3**) and prevent lipid peroxidation (**figure 4**) [5].

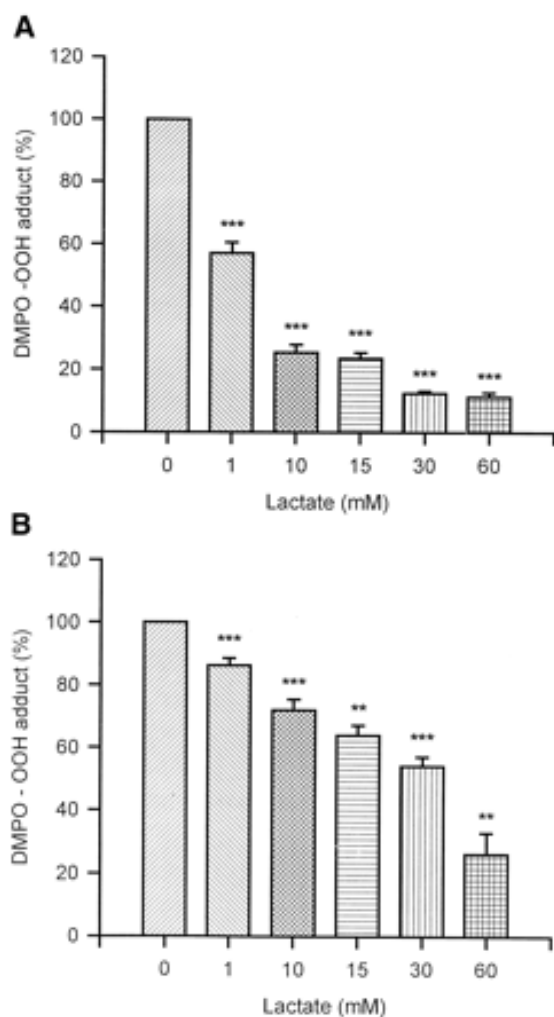
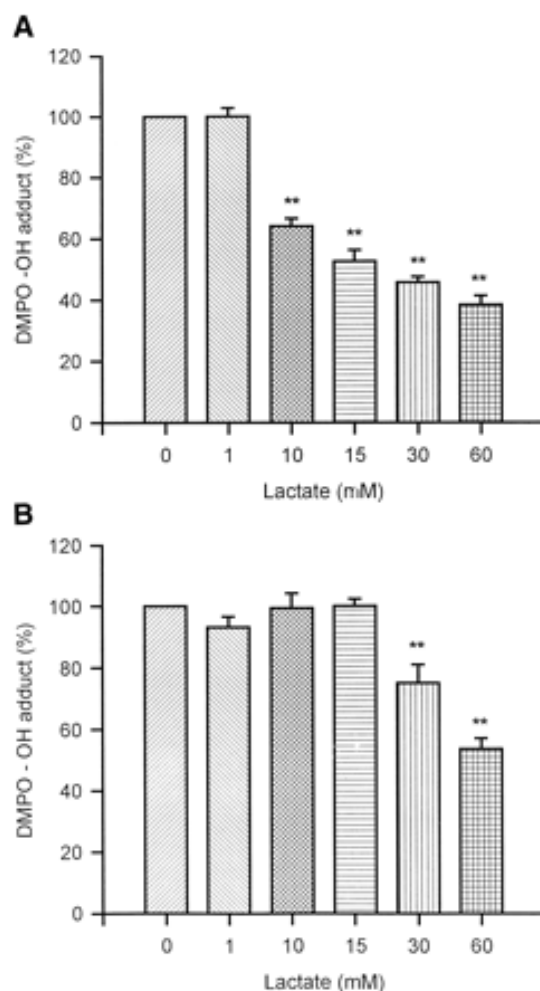


Figure 3. Scavenging of hydroxyl radical (HO^\bullet) by lactate. Effect of lactate concentrations on the electron paramagnetic resonance (EPR) signal corresponding to the HO^\bullet adduct with 5,5-dimethylpyrroline-*N*-oxide (DMPO). HO^\bullet was generated by decomposition of H_2O_2 by ferrous ion in presence or absence of lactate dissolved in water (A) or in plasma (B) at different concentrations (1, 10, 15, 30, and 60 mM). The 100% reference value corresponds to the level of DMPO-OH adduct produced in the corresponding control sample without lactate. Results are expressed as means \pm SE. ** Values significantly different ($P < 0.01$) from control without lactate. Adapted from [5].

Figure 2. Scavenging of superoxide anion ($\text{O}_2^{\bullet-}$) by lactate. Effect of lactate concentrations on the EPR signal corresponding to the $\text{O}_2^{\bullet-}$ adduct with DMPO. $\text{O}_2^{\bullet-}$ was generated by a xanthine-xanthine oxidase system in presence or absence of lactate dissolved in water (A) or in plasma (B) at different concentrations (1, 10, 15, 30, and 60 mM). The 100% reference value corresponds to the DMPO-OH adduct produced in the corresponding control sample without lactate. Results are expressed as means \pm SE. Values significantly different (** $P < 0.01$, *** $P < 0.001$) from control without lactate. Adapted from [5].



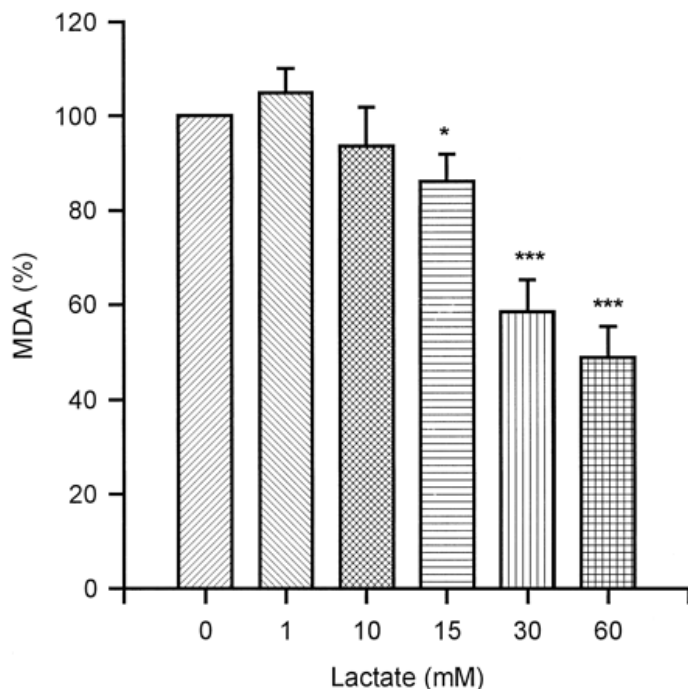


Figure 4. Effect of lactate on lipid peroxidation induced in hepatocyte cultures. Hepatocyte cultures were incubated for 4 h with ferric nitrilotriacetate (100 μM) either alone or with lactate at different concentrations (1, 10, 15, 30, and 60 mM). The malondialdehyde (MDA) value of the control culture was taken as the reference 100%. Results are expressed as means \pm SE, where 100% MDA recovery corresponds to the MDA level in cultures supplemented with iron alone. Values significantly different (* $P < 0.05$, *** $P < 0.001$) from control without lactate. Adapted from [5].

Possible mechanisms

The mechanisms underlying the antioxidant effect of lactate remain to be discovered.

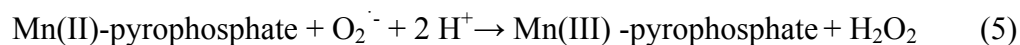
Superoxide dismutases (SOD) is known to catalyzes superoxide dismutation efficiently*.



Frederic et. al. reported *Lactobacillus plantarum*, an aerotolerant organisms contains no SOD, is able to scavenge superoxide [9]. *L. plantarum* uses Mn(II) to scavenge $\text{O}_2^{\cdot -}$.

*Buettner GR. (2005) Lipid peroxidation. class note in 77:222 Free radical and Radiation Biology. chapter 4; pp 7.

Mn(II) is known to be oxidized by $O_2^{\cdot -}$ and the product is Mn(III). In pyrophosphate buffer, the product is Mn(III)-pyrophosphate with rate constant $6 \times 10^6 \text{ M}^{-1} \text{ s}^{-1}$ [10].



The oxidized manganese needs to be recycled by reductants. NADPH has been reported as one of the reductants [11]. Lactate is another possible reductant. Lactate can convert to pyruvate in reverse direction in reaction (2) and donate electron to Mn(III).

Summary

Lactate is a metabolic intermediate formed during extreme muscular activity. *In vitro*, lactate is capable of scavenging superoxide, hydroxyl radical, and attenuates lipid peroxidation. However, the mechanisms of how it interacts with free radicals remain unclear. One of the possible mechanisms is lactate may be converted to pyruvate and donate electrons to the oxidized manganese. Then the reduced manganese can be used as antioxidant.

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