The Relationship Between Whole Blood, Plasma and Total Blood Lactate at Various Exercise Intensities.

GRADUATE THESIS

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Abstract

The purpose of this study was to examine the relationship between lactate concentrations derived from three sources: whole blood (WBLa), plasma (PLa) and total blood (TBLa) during an incremental bicycle test to exhaustion. Regression equations were computed enabling the prediction of one source from another. Thirty-two male subjects (16-25 yrs) participated in this study. Subjects performed a graded bicycle ergometer exercise test to exhaustion. Consecutive blood samples were drawn via an indwelling venous catheter at two minute intervals throughout the test, immediately post, and 5 minutes post exercise. Blood was analyzed for: WBLa, PLa, TBLa (YSI Lactate Analyser Model 23L), hemoglobin, hematocrit, bicarbonate and pH. Correlations were calculated for all variables. Simple and multiple regression equations were computed across exercise intensities. A one-way ANOVA was performed to determine if any significant differences existed between these three indices. This study illustrated that WBLa, PLa and TBLa concentrations, differed significantly and as exercise intensity increased so did the discrepancy. The regression equations were PLa=1.568(WBLa) + 0.149; WBLa=0.887(TBLa) + 0.015; PLa=1.208(TBLa) + 0.093. Although highly correlated to lactate values, the predictor variables: hemoglobin, hematocrit, bicarbonate and pH did not enhance the prediction equation.

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Chapter 1

Introduction

Purpose

The purpose of this research was to examine relationships between lactate concentrations derived from three sources: i) whole blood; ii) plasma and iii) total (lysed) blood throughout various exercise intensities. The specific aim was to provide a regression equation enabling the prediction of one source from another and to determine if hemoglobin, hematocrit, pH and bicarbonate would enhance the prediction equations.

Significance of Study

The measurement of lactate by coaches and researchers in developing and monitoring training regimens is common (Foster, Snyder, Thompson & Kuettel, 1988; Duggan & Tebbutt, 1990; O'Toole, Douglas & Hiller, 1989; Pierce, Weltman, Seip, Levine, Snead, Rogol & Weltman, 1989; Hurley, Hagberg, Allen, Seals, Young, Cuddihee & Holloszy, 1984). However, preliminary research at Lakehead University (Thayer, Allan & Burke, 1990) and recent literature (Buono & Yeager, 1986; Forrest, Morton & Lambardarios, 1990) reveals that lactate concentration in whole blood and plasma may not be comparable at all intensities of exercise. According to Buono and Yeager (1986) plasma lactate concentrations increase at a greater rate than whole blood lactate during incremental exercise.

A relationship between whole blood and plasma lactate concentrations may have implications in athletics. For example, in field tests and in some laboratories, blood samples are drawn by a finger prick and the whole blood is analyzed immediately with an automated lactate analyzer, while in other settings plasma lactate is the selected measurement. Furthermore, an alternative to whole blood and plasma is to lyse (split) the red blood cell and measure total blood lactate. Few studies have examined the difference in lactate concentrations between total blood and whole blood methods (Bishop, Smith, Murphy, & Kime, 1990). Given the inconsistencies with the various methods of blood preparation for subsequent lactate analyses, coaches would have difficulty comparing athletes outside of their own teams. The techniques used in the field, laboratories and in the clinical setting are variable and therefore contribute to this dilemma.

The results of this research will provide a quantitative comparison of lactate concentrations taken from three separate blood preparation techniques, under various levels of exercise intensity.

Limitations

1. Analysis of whole blood, plasma and total blood lactate are limited to the accuracy of the Yellow Springs Instrument Lactate Analyzer Model 23L.
2. Maximal oxygen consumption relies on the accuracy of the Beckman Metabolic Measurement Cart® and the ability of the

subjects to push themselves through a temporary feeling of discomfort as exhaustion approaches.

3. Although subjects were asked to refrain from strenuous physical activity for 24 hours prior to testing, this was not rigidly controlled. Subjects were questioned prior to the exercise test and if a subject had exercised on their test day the test was rescheduled.

4. Subjects were advised to eat at least 2-3 hours prior to testing, but the exact composition and quantity of this meal was not analyzed.

5. The subject's state of hydration could influence the results of this study. However, pre exercise hydration state was not controlled. Therefore, refraining from exercise, eating a meal and controlling the temperature of the lab were expected to reduce the effect of dehydration (Brooks & Fahey, 1984).

Delimitations

1. This study was delimited to 32 physically active men between the ages of 16 and 25 years, who were volunteers and resided in Thunder Bay, Ontario, Canada.

2. Blood measurements were: hemoglobin (Hb), hematocrit (Hct), whole blood lactate (WBLa), plasma lactate (PLa), total blood lactate (TBLa), pH, and bicarbonate (HCO_3^-).

3. The only physical performance measure was an incremental bicycle ergometer test to exhaustion.

Definitions

Anticoagulant - Anticoagulants are chemicals which prevent blood from clotting by inhibiting the conversion of fibrinogen into fibrin (Pendergraph, 1984).

Donnan's Equilibrium - A condition in which an equilibrium is established between two solutions separated by a semipermeable membrane so that the sum of the anions and cations on one side is equal to that on the other side.

Extracellular Fluid - All body fluids other than that contained within cells; includes plasma and interstitial fluid (Pendergraph, 1984).

Glycolysis - Glycolysis is defined as the degradation of a 6 carbon sugar to a 3 carbon sugar (Katz & Sahlin, 1990). The purpose of glycolysis is to produce adenosinetriphosphate (ATP) and to provide substrate for the tricarboxcylic acid (TCA) cycle for subsequent aerobic ATP production (Katz & Sahlin, 1990).

Glycolytic Inhibitor - A combination of two chemicals (sodium flouride/potassium oxalate) used to prevent glycolysis (Pendergraph, 1984).

Hematocrit - The percentage of the total blood volume that is red cells (Pendergraph, 1984).

Hemoglobin - Protein composed of four globular subunits, each bound to a single molecule of heme; the protein found in red blood cells that gives them the ability to transport oxygen in the blood (Pendergraph, 1984).

Lactic Acid - Lactic acid is an intermediate product of glucose metabolism in the glycolytic pathway. The chemical formula for

lactic acid is $C_3H_6O_3$ (Newsholme & Leech, 1983). Although lactate is the salt of lactic acid, lactic acid and lactate will be used interchangeably throughout this thesis.

Lysing Agent - A chemical substance used to lyse (rupture) erythrocytes and release lactate and intracellular fluid.

Peak $\dot{V}O_2$ ($P\dot{V}O_2$) - The highest $\dot{V}O_2$ value achieved by the subject throughout the exercise test (Kyle, Smoak, Douglass & Deuster, 1989).

pH - The measure of the acidity of a solution, pH, is calculated using the Henderson-Hasselbalch equation (pH=pka-log10[(base)/(acid)]), as the negative $\log_{10}$ of the [H+] in which 7 is neutral. Values greater than 7 are basic and values less than 7 are acidic (Hultman & Sahlin, 1980).

Plasma - When an anticoagulant is added to the blood sample, the liquid portion of the blood is termed plasma. Plasma contains fibrinogen (Pendergraph, 1984).

Plasma Lactate - Lactate contained in the plasma portion of the blood. Blood samples will be collected in Vacutainer® tubes containing an anticoagulant/glycolytic inhibitor, samples were then be centrifuged, separated and analyzed for lactate.

Total Blood Lactate - Lactate contained in the blood that has had the red blood cells lysed with buffer containing a lysing agent.

$\dot{V}O_2$max - The maximum amount of oxygen which can be consumed per unit of time by a person during a progressive exercise test to exhaustion. It is expressed as the volume per minute ($\dot{V}$) of oxygen (O_2) which can be consumed by the organism at the maximum (max)

workload which can be sustained for a criterion period of time (Brooks and Fahey, 1984).

Whole Blood - Whole blood is divided into two major components. The cellular component is red and contains red blood cells (erythrocytes), white blood cells (leukocytes), and platelets (thrombocytes). The other component is the fluid portion. This liquid portion is termed either plasma (anticoagulant added) or serum (no anticoagulant) (Pendergraph, 1984). For the purpose of this research project when referring to whole blood, the cells will remain intact.

Whole Blood Lactate - Lactate measured in whole blood. Blood samples were collected in Vacutainer® tubes containing sodium heparin (anticoagulant) and will be immediately analyzed for lactate concentration.

Chapter 2

Review of Literature

Introduction

Lactate analysis has become a valuable tool for coaches and researchers to assist in developing and monitoring an athlete's training regimen. Similarly, lactate analysis is used in the clinical setting by medical personnel to identify various metabolic disorders (Jones, 1987). However, recent research suggests that lactate concentrations in whole blood and plasma can be significantly different from each other especially at higher exercise intensities (Buono & Yeager,1986; Forrest, Morton & Lambardarios, 1990; Saltin, 1990).

This review of literature will provide an overview of the following related topics: glycolysis, factors affecting blood lactate, acid-base balance, and plasma volume. The latter part of this review will focus on the few studies which have addressed the discrepancy in lactate concentrations between whole blood, plasma, and total blood lactate concentrations and the proposed mechanisms behind this discrepancy.

Glycolysis

Lactate is a product of anaerobic glycolysis (Figure 1). Glycolysis is the splitting of a six carbon sugar into two 3 carbon molecules of lactate or pyruvate (Katz & Sahlin, 1990). The glycolytic pathway is an anaerobic energy transfer system in which six carbon sugars are used to produce adenosine triphosphate through intermediary metabolic transfer. The glycolytic pathway

can begin in one of two ways: 1), a glucose molecule enters the cytoplasm of a cell and is energized with a high energy phosphate, or 2), a terminal glycolytic residue of a glycogen molecule is cleaved from the polysaccharide linkage and phosphorylated to form glucose-1-phosphate. Regardless of whether or not the glycolytic pathway begins with glucose or glycogen, the point of convergence and essentially the start-up molecule for glycolysis is glucose-6-phosphate.

Glucose-6-phosphate is an energized hexose molecule which is eventually super-energized and subsequently degraded into two 3 carbon molecules of pyruvate. In the presence of adequate oxygen supply pyruvate is oxidized to carbon dioxide and water via the tricarboxylic acid (TCA) cycle . However, if the demands for energy from ATP exceeds normal glycolytic rates, then pyruvate molecules are temporarily converted to lactate molecules (Bowers & Fox, 1992).

The key reactions of glycolysis are catalyzed by the enzymes hexokinase, phosphorylase, phosphofructokinase (PFK), glycerol 3-phosphate dehydrogenase and lactate dehydrogenase (LDH). Substrate for glycolysis is under the control of two important enzymes; hexokinase and phosphorylase. Hexokinase is an enzyme that assists glucose in entering the cell to be phosphorylated forming glucose 6-phosphate. The breakdown of glycogen to glucose 6-phosphate is controlled by the enzyme phosphorylase. The flux of substrate through glycolysis is controlled by the rate limiting enzyme phosphofructokinase (Gollnick & King, 1969). Phosphsfructokinase catalyses the conversion of fructose 6-

phosphate to fructose 1, 6-diphosphate. The activity of PFK is modulated by levels of ATP, phosphocreatine (PC), and citrate (Ivy, Costill, VanHandel, Essig & Lower,1981). Inhibition of PFK could interrupt glycolysis and therefore slow down the rate of ATP production (Wenger & Reed, 1976). The terminal enzyme of glycolysis is lactate dehydrogenase (LDH). Lactate dehydrogenase is important in controlling glycolysis because the muscle type isoenzyme is, in effect, in competition with mitochondria for pyruvate. Because of the significant content of LDH in muscle, and it's primary role to provide for the re-oxidation of NADH+, some lactate will always be formed at rest and during the performance of exercise (Brooks & Fahey, 1984).

As the intensity of exercise increases, a point is reached where an increase in lactate concentration in blood and muscle becomes evident (Gollnick, Bayly & Hodgson, 1986). During glycolysis the mitochondria is impermeable to the $NADH^+$ formed in the cytoplasm (McArdle, Katch & Katch, 1991). At high intensities of exercise, the NAD^+/NADH ratio must be maintained at a high ratio in order for anaerobic glycolysis to continue (Newsholme & Leech, 1983). Two processes contribute to the re-oxidation of NADH. The first process important to the oxidation of NADH is that which is catalysed by glycerol 3-phosphate dehydrogenase. The second process is reduction of pyruvate to lactate in the reaction catalysed by lactate dehydrogenase (Newsholme & Leech, 1983). The concentration of glycerol 3-phosphate and lactate both increase within muscle under anaerobic conditions.

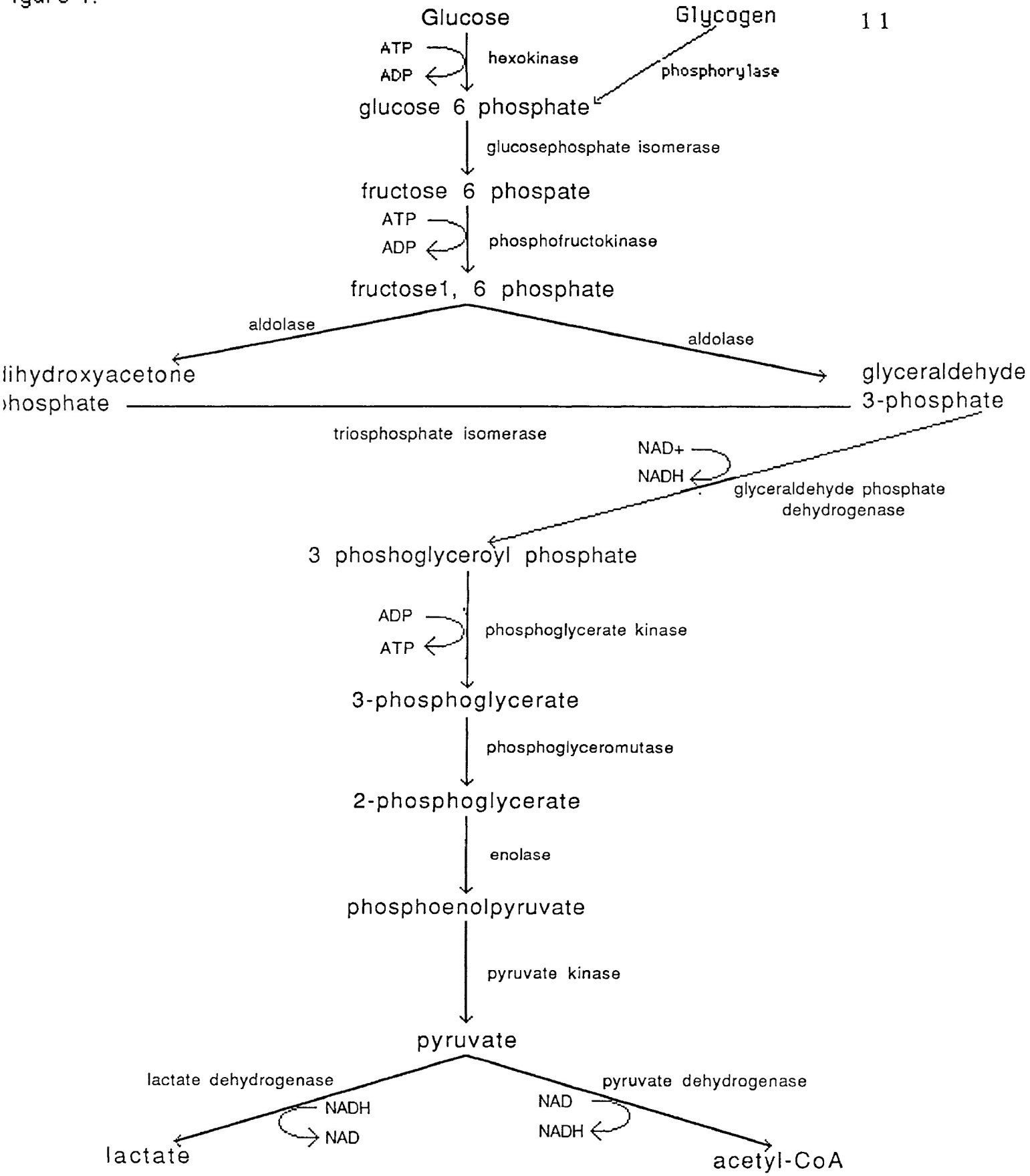
However it is the measurement of muscle and blood lactate which has been widely used to assess anaerobic metabolism

Factors Affecting Blood Lactate

Since the determination of blood lactate concentration as an index of anaerobic metabolism is a common laboratory test, it is important to interpret any blood lactate measurement in light of all known or suspected contributing factors.

Many factors influence blood lactate concentration. According to Brooks (1988) blood lactate measurements represent a balance between determinants which contribute to lactate production and those contributing to lactate removal. The term anaerobic refers to without oxygen. Under anaerobic glycolysis lactic acid is formed (Brooks,1988). There are other reasons though for lactic acid formation in working muscles it's appearance in the blood. In addition to anoxia, production will be influenced by both anaerobic capacity and lactate clearance rate. In turn, these two measures are dependent on fiber type (Tesch, Daniels & Sharp, 1982), substrate availability (Gollnick, Warwick, Bayly & Hodgson, 1986), LDH isoenzymes, buffering capacity (Bhattacharya, Panda, DasGrupta & De,1983), capillarization and mitochondrial activity (Brooks & Fahey, 1984). Any variable that affects entry and/or removal of lactate can change the level of lactate in blood.

Figure 1.



lewsholme & Leech, 1983, p. 180)

A. The Effect of Anoxia on the Glycolytic Production of ATP

Oxygen delivery, oxygen uptake (Brooks, 1988), oxygen utilization and exercise intensity (Gollnick et al., 1986) will affect the degree of hypoxia in muscle tissue. An increase in blood lactate levels is observed at approximately 50 to 70% VO_2max, well before aerobic capacity is fully utilized (Katz & Sahlin, 1990). The mechanism for the increased lactate concentration is not completely understood. One explanation is that the contracting muscle is oxygen deficient and as a result the anaerobic metabolic pathway is required to produce the necessary ATP needed to perform the exercise (Katz & Sahlin, 1990). Oxygen impairment leads to an increased reliance on the anaerobic metabolic pathways in order to produce the necessary energy. During brief periods of circulatory occlusion as in the isometric phase of a muscle contraction when oxygen supply is impaired (anoxia), 30 to 60% of ATP is produced anaerobically, illustrating the importance of this energy pathway (Katz, Sahlin & Henriksson, 1986). The fate of pyruvate depends on the presence of oxygen. In anaerobic glycolysis the pyruvate molecule receives two hydrogens from the nicotinamide adenine dinucleotide (NAD^+) molecule to form lactate and to allow glycolysis to continue (Newholme & Leech, 1983; Wilson, Ferraro & Weber, 1983). However, during long term, low intensity exercise oxygen is not limited and lactate production is reduced which results in an anaerobic glycolytic energy production of about 2% (Katz and Sahlin, 1990).

Anoxia occurs with increasing exercise intensity and is the basis of anaerobic threshold measurements. When the metabolic

demand exceeds available oxygen, anaerobic glycolysis is increased to raise ATP production and results in a concomitant increase in lactic acid production.

B. Anaerobic Capacity

As an individual increases the metabolic demand on anaerobic energy production, lactate concentrations increase. The critical factors in this energetic process include the anoxic condition and imbalances between the rate of glycolysis and the rate of pyruvate oxidation.

Recent research has revealed that lactate production is the result of the rapid breakdown of glycogen regardless of the concentration of oxygen (Brooks,1988). As noted previously, lactate dehydrogenase will always convert some pyruvate to lactate. In fact, several researchers have demonstrated in an earlier study that lactate can be produced at low, moderate and high intensities of exercise (Gollnick, Bayly & Hodgson, 1986).

Buffering capacity, percent fast twitch muscle fibres, and substrate availability, are some of the factors that affect anaerobic capacity (Saltin, Nazar, Costill, Stein, Jonsson, Essen & Gollnick, 1976). Buffering of H^+ ions is increased in the well trained athlete who specialize in events of maximal effort between 30 seconds and two minutes. This training regimen allows anaerobic glycolysis to continue for a longer duration, thereby producing higher lactate values. Athletes who compete in events which last less than 30 seconds, such as sprinters, would also demonstrate high post-maximal exercise blood lactate levels (Bhattacharya, Panda,

DasGupta & De, 1983), because of the exercise recruitment of a greater percentage of fast twitch muscle fibres (Bhattacharya et al., 1983). Anaerobic metabolism is best suited to fast twitch fibers because they have a high glycogenolytic potential and a relatively poor capillary supply. The increased production of lactic acid by fast twitch muscle has been shown to cause a lactate concentration gradient between fibre types after maximal exercise (Tesch, Daniels & Sharp, 1982). A non-uniform distribution of lactate is often seen in maximal exercise of 30 seconds duration compared to maximal exercise of 60 seconds (Tesch et al., 1982). The longer exercise work period promotes a greater reliance on muscle contraction by slow twitch muscle fibers and a lower dependence on fast twitch fibers. This adjustment in muscle fiber recruitment leads to a reduction in lactate production (Tesch et al., 1982).

Substrate availability can affect lactate production. For example, the balance between anaerobic glycolysis and pyruvate oxidation can favor oxidation if free fatty acids (FFA) are being combusted. Ivy and co-workers (1981) revealed that when FFA levels are raised during muscular activity, there is an increased reliance on lipid oxidation and a decrease in blood lactate accumulation. Citrate, a product of fatty acid oxidation, inhibits the glycolytic enzymes phosphofructokinase (PFK) and glycerol-3-phosphate dehydrogenase. Therefore, if two athletes were exercising at the same oxygen consumption the athlete with elevated levels of free fatty acids would produce less lactate.

C. Lactate Removal

The concentration of lactate in blood is determined by the balance between production in muscle and it's clearance rate from the blood. Contrary to traditional beliefs lactate should not be simply viewed as an end product of glycolysis as it may be oxidized as a fuel (Brooks, 1988). Instead it is oxidized as a fuel (Brooks, 1988). Lactate is thus an intermediate product of carbohydrate metabolism and is an important fuel source during exercise ranging from 40-75% $\dot{V}O_2$max (Brooks, 1988). During physical activity, lactate is an important oxidizable substrate (Mazzeo, Brooks, Schoeller & Budinger, 1986; Brooks, 1986). Mazzeo et al., (1986), reported that at rest, 50% of lactate production is removed by direct oxidation. During continuous exercise a greater proportion is removed with the percentage removed by oxidation being 90% at 75% $\dot{V}O_2$max (Mazzeo et al., 1986) Thus the non-accumulation of lactate does not imply that the glycolytic rate has not increased. According to Jorfeldt, Juhlin-Danndelt & Karlsson (1978), at 40% $\dot{V}O_2$max there is a 25-fold increase in glycolysis, although there is only a slight increase in the lactate concentration within the muscle. Under these conditions, the increase in glycolysis is essentially balanced (steady state) by an equal increase in pyruvate oxidation (Katz & Sahlin, 1990). Essentially, lactic acid does not build up because its removal rate is equal to the rate of production (McArdle, Katch & Katch, 1991).

Oxidation is the major fate of blood lactate and consequently there is a great deal of interaction between those factors that enhance aerobic capacity and those factors that enhance lactate

clearance. Brooks & Fahey (1984) studied the fate of lactate using tracer carbons and revealed that most of the labelled lactate carbon was recovered as expired carbon dioxide demonstrating that the lactate was oxidized. Yet the lactate which is not oxidized may serve as a glucogenic precursor or be incorporated into amino acids and proteins.

When exercise stops, the metabolic state of the body appears to determine the fate of blood lactate during the recovery period. If liver glycogen and blood glucose are at or near normal levels, high concentrations of lactate will be dissipated through oxidation (Newsholme & Leech, 1983). Conversely, prolonged exercise could encourage greater conversion of lactate to glucose. Lactate can then be viewed as a carbon reservoir to power the recovery process. In fact, the presence in skeletal muscle of the heart specific isoforms of LDH will encourage the conversion of lactate to pyruvate and it's subsequent entry into the TCA cycle. This pyruvate substitutes for that which would have been supplied by glucose or glycogen.

Recovery exercise intensity is important to lactate clearance. Evans and Cureton (1983) reported that a recovery exercise intensity of 25% VO_2max facilitated the rate and amount of lactate removal. These authors also reported that lactate removal was greater in trained than untrained subjects at this recovery exercise intensity.

Acid-Base Balance

Lactic acid production affects blood pH. Blood pH has been implicated as a factor in the transport mechanism of lactate between the plasma and red blood cell (Dubinsky & Racker, 1978).

During low to moderate exercise intensity, the body maintains an acid-base equilibrium (Hultman & Sahlin, 1980). Exercise or increased metabolism produces products such as carbon dioxide (CO_2), water (H_2O), and lactate, all of which are related to acid-base regulation (Hultman & Sahlin, 1980). The regulation of acid-base balance is actually the regulation of the hydrogen ion [H^+] concentration in the body fluids (Guyton, 1976). The hydrogen ion concentration is measured or expressed as pH.

Previous research studies have defined the patterns of acid-base balance and lactate response during incremental exercise (Wasserman, Whipp & Davis, 1981; Hughson & Green, 1982; Sahlin, Alvestrand, Brandt & Hultman, 1978). At moderate to high intensities of exercise, there is an increase in blood lactate concentration and a simultaneous increase in the [H^+] of blood and a subsequent release of hydrogen ions from muscle (Hughson & Green, 1982). As mentioned previously, under intense muscular exercise, part of the energy requirement is provided anaerobically and is evidenced by lactic acid formation. The appearance of lactate in blood is associated with an increase in hydrogen ion content. Sahlin et al. (1978), investigated lactate production, pH, and pCO_2 in three subjects who underwent exhaustive exercise on a bicycle ergometer. Sahlin et al. (1978) reported that the blood lactate and hydrogen ion concentration increased continuously throughout the exercise period. In order to maintain homeostasis the increase in [H^+] ions, resulting from intense exercise, must be buffered.

Buffer systems are present in body fluids which immediately combine with an acid to prevent changes in the hydrogen ion

concentration (Guyton, 1976). A change in the $[H^+]$ ion concentration stimulates the respiratory center via increased CO_2 production and this causes an increase in the rate of pulmonary ventilation. The net effect is a reduction in the $[H^+]$ concentration of blood.

The reaction between hydrogen and bicarbonate (HCO_3^-), a buffering agent, results in an increased production of carbon dioxide. As mentioned above, an excess of CO_2 is eliminated through increased ventilation. High intensity exercise results in increased hydrogen ion and lactate production, which exceeds the buffering capacity of blood and muscle (Wasserman et al., 1981; Sahlin et al., 1978). A decrease in pH causes an increase in pCO_2, a decrease in HCO_3^- and an increase in pulmonary ventilation (Wasserman et al., 1981).

Dubinsky and Racker (1978) conducted a study to investigate the transport mechanisms of lactate across the red blood cell membrane. The authors reported that lactate eflux was strongly pH dependent. Conversely, lactate exchange under equilibrium conditions increased with decreasing pH, which Dubinsky and Racker (1978) attributed to the direct effect of the pH upon the activity of the transport system.

The influx of lactic acid was also pH dependent (Dubinsky & Racker, 1978). Dubinsky and Racker (1978) suggest that the pH gradient was the major controlling factor of lactate flux in these experiments. The pH dependency of lactate influx and eflux indicates that the direction of the pH gradient across the membrane is the major determinant in the net lactate flux.

From a review of the literature, Dubinsky and Racker (1978) detailed two different mechanisms for the direction of lactate flux. The first mechanism originally proposed by Halstrap (1976), is a lactate/Cl^- exchange. The second mechanism suggests that lactate transport could be driven by a H^+-lactate symport (Dubinsky & Racker, 1978). Their findings suggest that a H^+-lactate symport mechanism for lactic acid transport exists independent and distinct from the inorganic anion channel.

Plasma Volume

Exercise of high intensity causes changes in plasma volume which could possibly confound the interpretation of plasma lactate values. The relationship between lactate values and plasma volume has not been adequately studied.

The total body fluids are comprised of blood plasma, interstitial fluid, intracellular fluid and miscellaneous components (deVries, 1986). During exercise, only blood plasma, interstitial fluid and intracellular fluid are important (deVries, 1986). Capillary cell walls are permeable to most substances in the plasma. As a result, plasma and interstitial fluid mix constantly and consequently are referred to as extracellular fluid (deVries, 1986).

In order to maintain homeostasis, water diffuses across the cell membrane in the direction needed (deVries,1986). Therefore, during moderate to heavy exercise, water from extracellular fluids is eliminated during sweating. Subsequently, a rise in the concentration of nondiffusible substances of the interstitial fluid results in the transfer of water across the cell membrane from the

cells of the intracellular component to the extracellular component and may facilitate dehydration (deVries, 1986).

During single efforts of exercise, there have been reports of decreased plasma volume as evidenced by an increase in hemoglobin concentration, hematocrit and red blood cells in peripheral blood (Davidson, Robertson, Galea, & Maughan, 1987; Dickson, Wilkinson, & Noakes, 1982). The degree of change of these hematological parameters depends upon the type, duration, intensity of exercise, environmental conditions and the degree of fitness of participants (Zbigniew, 1990).

The reduction of plasma volume, resulting from a shift of water from intracellular to extracellular space occurs at the onset of exercise and remains proportional to intensity (Wilkerson et al., 1977). Furthermore, Wells, Stern, Kohrt, & Campbell, (1987) suggest that the fluid shifts are more pronounced during walking and running than during bicycling.

The reduction in plasma volume depends on physical load and the degree of physical fitness. Convertino, Keil, and Greenleaf (1983) found an average reduction in plasma volume of 7.0% at 100W workload, 11% at 175W and 13.7% at 225W workload during a cycle ergometer study with untrained males. Similarly, Wells, Stern, Kohrt & Campbell (1987), reported an 11-18% decrease in plasma volume during treadmill exercise with workloads from 33-90% VO_2max. Wilkerson, Gutin, and Horvath (1977) have reported a decrease of plasma volume from 45-75% $\dot{V}O_2$max.

Wilkerson, Gutin and Horvath (1977), conducted a study to examine the changes in blood, red cell and plasma volume in human

subjects over a wide range of exercise intensities. Each subject performed a constant speed, incremental slope test at 30, 45, 60, 75, and 90% $\dot{V}O_2$max. Blood samples were taken pre and throughout the exercise bout. Percent changes in blood volume, red cell volume and plasma volume from pre exercise levels were calculated using the method of Dill and Costill (1974). The authors reported a decreased blood and plasma volume with an increased oxygen uptake and increased workload. A linear function of plasma volume and oxygen uptake from rest up to 75% $\dot{V}O_2$max was identified. No linear relationship was found at higher levels of exercise intensity. Wilkerson, Gutin, Molnar and Horvath (1976) suggested that the abrupt plasma volume change at approximately 65-70% $\dot{V}O_2$max is related to the point at which significant lactate accumulation is first observed during exercise.

While it is well established how plasma volume responds during exercise, it has not been established how changes in plasma volume effect lactate concentrations. It is possible to speculate that a decrease in plasma volume would increase the concentration of lactate. However, the subsequent increase in hematocrit associated with a decrease in plasma volume, may assist in oxygen delivery.

Whole Blood and Plasma Lactate Discrepancies

In many studies of lactate metabolism, an assumption is made that during exercise, changes in whole blood lactate concentrations accurately reflect changes in plasma lactate values. At rest, blood lactate is distributed evenly throughout the water phases of the

plasma and red blood cells (Piquard, Shaefer, Dellebach & Haberey, 1980; Forrest, Morton & Lambardarios, 1990) and a lactate gradient does not exist (Saltin, 1990; Buono & Yeager, 1986). During exercise, however, the distribution of lactate changes between the water phases of blood, with plasma lactate concentration being higher than the lactate concentration of erythrocytes (Saltin, 1990; Buono & Yeager, 1986; Forrest et al., 1990).

In a study by Huckabee (1956) the relationship of extracellular and intracellular concentrations of pyruvate and lactate were investigated in humans. Huckabee (1956) measured lactate and pyruvate concentrations in whole blood and plasma in humans during rest, mild exercise, fever and thiamine deficiency. A constant relationship was reported between the water phases of whole blood and plasma lactate. It was concluded that lactate moves across the red blood cell freely and passively.

However, Daniel, Morishima, James and Adamson (1964) postulated that under non steady-state conditions (as in high intensity exercise when the amount of O_2 available to the muscles and tissue is constantly changing) a disequilibrium of lactate and pyruvate concentrations will occur between whole blood and plasma.

Buono and Yeager (1986) carried out a study to determine plasma and intraerythrocyte lactate concentrations during graded exercise in men and women. Subjects performed a graded exercise test ($\dot{V}O_2max$) on a bicycle ergometer. Buono and Yeager (1986) reported no significant differences in lactate concentration between whole blood and plasma at rest or low intensity exercise (25% and 50% $\dot{V}O_2max$). However, as exercise intensity increased (75% and

100% $\dot{V}O_2$max), plasma lactate concentrations were significantly increased over whole blood lactate levels. These results demonstrated that during graded exercise, a significant plasma to red cell gradient forms (Buono & Yeager, 1986). Additionally, Buono and Yeager (1986) and Huckabee (1956) reported that the plasma to cell gradient was linearly related to plasma lactate concentrations.

Harris and Dudley (1989) conducted a study to determine whether the relationship between whole blood and plasma lactate concentrations are altered by exercise. Subjects underwent two incremental-load aerobic power tests performed on a cycle ergometer. Blood samples were taken pre-exercise, immediately post exercise, and after fifteen minutes of recovery. Intraerythrocyte lactate was calculated by the following equation adopted from Conn (1966, cited by Harris and Dudley,1956): Ce=[Cwb-(1-Hct) x Cp/Hct]. Hct, wb, p, e, C are hematocrit, whole blood, plasma, erythrocyte and concentration, respectively (Harris & Dudley, 1989). Harris and Dudley (1989) reported plasma lactate concentrations two times that of red blood cell lactate concentrations. All three indices showed an increase of lactate concentration from rest to immediately post exercise. Plasma lactate values post exercise were twice the value of red blood cells and 4.3 mmol/L greater than whole blood lactate. These authors noted that whole blood and plasma lactate levels did not decrease during recovery, but red blood cell lactate increased (Harris & Dudley, 1989). Therefore, the authors noted the plasma to red blood cell lactate gradient post exercise decreased by the end of recovery.

A high correlation between whole blood and plasma lactate concentrations at rest, post exercise and during recovery was reported (Harris & Dudley, 1989). When whole blood, plasma and red blood cell lactate levels were considered together, a similar correlation was reported.

Harris & Dudley (1989) postulated that the twofold greater increase in plasma lactate over red blood cell lactate would contribute to a larger relative increase in the hydrogen ion concentration of plasma. Since lactate is an acid and a strong anion, its greater accumulation in plasma would increase the total acid concentration and reduce the strong ionic difference more in plasma, than red blood cells (Stewart, 1983). If the strong ionic difference was reduced more in plasma, this would alter the distribution of cations between plasma and red blood cells which obey the law of Donnan equilibrium (Fitzsimons & Sendroy, 1961).

Plasma lactate after recovery was not different from that measured immediately post exercise. The results of this study suggest that during high intensity exercise lactate is added to the plasma portion of blood so quickly that the delay in transport across the red blood cell membrane becomes more apparent (Dudley & Harris, 1989). They were unable to explain how such an increase in plasma lactate accumulation over red blood cells during exercise could not reduce the pH gradient between plasma and red blood cells.

From this study, an equation was developed to predict plasma lactate from whole blood lactate values directly post exercise and after 15 minutes recovery. The equation: Plasma lactate (mmol/L) = whole blood lactate (mmol/L) x 1.43 - 1.11 ($r=0.95$, $p < 0.001$),

defines the relationship between these two indices immediately post exercise. However, the prediction equation generated for the data pre-exercise and recovery combined, differed from the above equation [Plasma lactate (mmol/L) = whole blood lactate (mmol/L) x 1.16 + 0.06 ($r=0.93$, $P<0.001$)]. Therefore, the authors suggested that the condition under which blood is sampled influences the nature of the relation between whole blood and plasma lactate concentrations (Dudley & Harris, 1989).

Mechanisms for Cell to Plasma Lactate Gradients

The exact mechanism that causes the red blood cell to plasma lactate gradient is still unknown. The literature reveals conflicting postulations regarding the exact mechanism of lactate transport across the red blood cell membrane.

Saltin (1990) states that prior to exercise, there is virtually no lactate gradient between the water phases of muscle cell (interstitial space), plasma and red blood cells. However, as exercise intensity increases, a gradient exists between muscle and plasma and between plasma and the red blood cells. Saltin (1990) attributed this gradient to slow facilitated diffusion of lactate through the red blood cell membrane.

Dubinsky and Racker (1978), however, propose that lactate transport across cell membranes takes place via a specific transport system and not by simple diffusion. These authors suggest that efflux and influx of lactate across the erythrocyte membrane are pH dependent and lactate transport occurs primarily via a monocarboxylate-specific transport system.

Chapter 3

Methods and Procedures

Purpose

The purpose of this research was to examine relationships between lactate concentrations derived from three sources: i) whole blood; ii) plasma and iii) total (lysed) blood throughout various exercise intensities. The specific aim was to provide a regression equation enabling the prediction of one source from another and to determine if hemoglobin, hematocrit, pH and bicarbonate enhance the prediction equations.

Subjects

Thirty-two healthy, physically active male students from Lakehead University comprised the sample population. The criteria for physically active referred to subjects who participated in at least 3 workouts per week at a moderate exercise intensity for 120 minutes total duration. Students participated on a voluntary basis and ages ranged from 16 to 25 years old. Subjects signed informed consent forms (Appendix D) and all procedures were approved by the Lakehead University Ethics Committee.

Procedures

Subjects were asked to report to the Human Performance Lab having refrained from intense exercise for at least 24 hours. Subjects were advised to eat at least 2 to 3 hours prior to testing. Anthropometric measurements were obtained prior to exercise. An

indwelling catheter was then inserted into an antecubital vein of the forearm.

A resting blood sample was obtained after catheter insertion. The subjects were then given time to familiarize himself with the equipment (i.e., seat height, toe clips, etc.). Subjects were fitted with the headgear and connected to the Metabolic Measurement Cart. The specifics of the anthropometric, physiological, and hematological tests are described below.

Anthropometric Measurements

Height was measured to the nearest 0.1 centimeter. Weight was determined to the nearest 0.1 kilogram.

Physiological Measurements

Each subject performed a graded bicycle ergometer exercise test to exhaustion ($\dot{V}O_2max$). The protocol, adopted from Buono, Clancy and Cook (1984) and Buono & Yeager (1986) consisted of a two minute warm up with 25 watts resistance. After the initial warm up period, resistance was then increased by 25 watts every two minutes until voluntary exhaustion was reached. If subjects were unable to maintain a pedaling cadence of 60 revolutions per minute, the test was terminated. $\dot{V}O_2max$ was determined using the Beckman Metabolic Measurement Cart (Horizon Systems). The exercise test was performed on a Monarch Bicycle Ergometer, model 868 (Monarch, Stockholm, Sweden).

Criteria for achievement of $\dot{V}O_2max$

1) Leveling of or decrease in $\dot{V}O_2$ with increasing exercise intensity, or,
2) Respiratory exchange ratio greater than 1.15, or,
3) Attainment of age predicted maximal heart rate

Blood Samples

Consecutive blood samples were withdrawn from an antecubital vein via an indwelling catheter throughout the exercise test. Blood samples were taken at two minute intervals (1:30 minutes into each workload). Blood was also collected at exhaustion and five minutes post exercise.

Hemoglobin and hematocrit samples were collected in 2.5 ml Vacutainers® containing ethylenediaminetetraacetic acid (EDTA). Analyses were determined using the Cell-Dyne 3000 at the Port Arthur General Hospital.

Bicarbonate (HCO_3^-) and venous blood pH were determined using the ABL 30 Blood Gas Analyzer (Copenhagen). Samples were collected in 3 ml Vacutainers® containing sodium heparin, and were placed on ice until analysis was performed.

Samples for whole blood lactate determination were collected in 3 ml Vacutainers® containing sodium flouride/potassium oxalate. Samples were analyzed immediately using the Yellow Springs Instrument Model 23L Lactate Analyzer.

Blood samples for plasma lactate analysis were collected in 3 ml Vacutainers® containing sodium heparin. Samples were centrifuged using a serum separator at 3000 revolutions per minute

for 5 minutes and analyzed using the same instrument for whole blood analysis.

Total blood (lysed cells) lactate was collected in a 3 ml tube containing sodium flouride/potassium oxalate. Samples were analyzed using the Yellow Springs Instrument Model 23L Lactate Analyzer. The buffer contained a lysing agent (Triton X®).

Plasma volume change was calculated using the method of Dill and Costill (1974).

Statistical Analysis

Descriptive statistics (mean±standard deviation and range) of physical characteristics and Peak $\dot{V}O_2$ were performed. Data was divided into stages according to percentage of Peak $\dot{V}O_2$ (rest-49%, 50-59%, 60-69%, 70-79%, 80-89%, 90-100% P$\dot{V}O_2$max, and 5 minute recovery). The mean score and standard deviation of each variable (whole blood lactate, plasma lactate, hemoglobin, hematocrit, pH and bicarbonate) was computed across all stages. Paired t-tests between the absolute difference of whole blood and plasma lactate were performed at each stage. A one-way ANOVA with repeated measures was computed using the difference of scores (plasma lactate - whole blood lactate). Post hoc analysis employed the Scheffe method. Simple regression analysis between whole blood and plasma lactate and between total blood and whole blood lactate were performed at each stage and with all data pooled together. Stepwise multiple regression equations using predictor variables (hematocrit, hemoglobin, bicarbonate and pH) in addition to whole blood were generated at each stage and with all data pooled together

to establish if a better estimate of the criterion may be obtained. A test of significance for each correlation was done using an alpha level of $P \leq 0.05$.

Chapter 4

Results

Thirty-two men participated in this study. The mean age, height, weight and Peak $\dot{V}O_2$ are listed in Table 1.

Variable	Mean	±S. D.	Minimum	Maximum
Age (yrs)	20.8	±2.00	16.0	25.0
Height (cm)	178.6	±5.99	168.0	189.0
Weight (kg)	79.7	±9.47	62.5	101.0
$P\dot{V}O_2$ (ml/kg/min)	49.4	±8.83	33.3	67.9

Table 1. Descriptive statistics of physical characteristics and Peak $\dot{V}O_2$ ($P\dot{V}O_2$) of Thirty-two Male Subjects.

Data obtained from this study were divided into stages according to percentage of Peak $\dot{V}O_2$. The stages were as follows: stage one (rest - 49% $P\dot{V}O_2$), stage two (50-59% $P\dot{V}O_2$), stage three $P\dot{V}O_2$), stage five (80-89% $P\dot{V}O_2$), and stage six (90-100% $P\dot{V}O_2$). The final stage was data obtained five minutes post exercise (Recovery).

The mean score and standard deviation of each variable (whole blood lactate, plasma lactate, hemoglobin, hematocrit, pH, and bicarbonate) according to stages are listed in Table 2.

Whole blood lactate (WBLa) concentrations increased from stage one to stage six. The highest whole blood lactate value obtained was during 5 minutes post exercise (recovery). Mean whole

blood lactate values were 0.8 (±.26), 6.2 (±1.65), and 8.5 (± 1.47) mmol/L at stage one, stage six and recovery, respectively.

Variable	1 R -49% X±S.D.	2 50-59% X±S.D.	3 60-69% X±S.D.	4 70-79% X±S.D.	5 80-89% X±S.D.	6 90-100% X±S.D.	Recovery X±S.D.
WBLa	0.8±.26	1.1±.28	1.6±.73	2.3±.82	3.8±1.42	6.2±1.65	8.5±1.47
PLa	1.4±.37	2.0±.45	2.7±.88	3.9±1.27	5.9±1.79	10.0±2.48	13.4±2.16
Hb	149±8.7	149±8.0	152±9.0	155±7.5	157±5.6	160±9.0	158±8.1
Hct	.44±.03	.44±.03	.45±.03	.46±.02	.46±.02	.47±.03	.47±.02
pH	7.34±.02	7.33±.02	7.34±.03	7.32±.02	7.29±.03	7.21±.05	7.20±.05
HCO_3	28.2±1.7	27.6±1.5	26.7±2.4	25.9±2.5	23.5±2.9	20.3±3.2	13.7±2.7
Abs. diff WBLa & PLa	.6±.21	.8±.29	.9±.62	1.6±.61	2.1±.75	3.9±1.16	4.9±1.33

Table 2. Descriptive statistics of lactate, hematology and venous blood gas values across all stages of exercise intensity, including recovery.

Similar to whole blood lactate, plasma lactate values increased from stage one to stage six and peaked during recovery. Mean plasma lactate concentrations were 1.4 (±.37) mmol/L at stage one, 10.0 (± 2.48) at stage six and 13.4 (±2.16) mmol/L during 5 minutes post exercise.

Paired t-tests between whole blood and plasma lactate were computed for each stage. The t-tests revealed that whole blood and plasma lactate are statistically different at each stage (Table 3.)

Stage	Count	WBLa (mmol/L)	PLa (mmol/L)	ΔMean (mmol/L)	p-value
One	96	0.8	1.4	0.58	< 0.0005
Two	34	1.1	2.0	0.81	< 0.0005
Three	29	1.6	2.7	1.04	< 0.0005
Four	29	2.3	3.9	1.60	< 0.0005
Five	27	3.8	5.9	2.16	< 0.0005
Six	50	3.1	10.0	3.90	< 0.0005

Table 3. Paired t-test between whole blood and plasma lactate at each stage of exercise.

The mean absolute difference between whole blood lactate and plasma lactate was .6 (±.21) mmol/L at stage one, 3.9 (±1.16) mmol/L at stage six and 4.9 (±1.33) mmol/L during recovery. A one-way ANOVA with repeated measures was computed on the difference scores and revealed a significant main effect, $F(5,253)=198.378$, $p<.0001$ (Table 4). In other words, the absolute difference between whole blood lactate and plasma lactate increased significantly over time. Post-hoc analysis using Scheffe's method revealed that the absolute difference between whole blood lactate and plasma lactate is statistically significant between various stages at $p< 0.05$ and $p< 0.01$ (Table5).

	DF	Sum Squares	Mean Square	F-ratio
Between group	5	397.158	79.432	198.375
Within groups	253	101.303	.400	p ≤ .0001
Total	258	498.460		

Table 4. One-way ANOVA expressing the absolute difference between whole blood lactate and plasma lactate at each stage of exercise

Stage	Mean (mmol/L)	One	Two	Three	Four	Five	Six
One	(0.58)	- - -					
Two	(0.81)	- - -	- - -				
Three	(1.04)	*	- - -	- - -			
Four	(1.60)	* *	* *	- - -			
Five	(2.16)	* *	* *	* *			
Six	(3.91)	* *	* *	* *	* *	* *	- - -

Table 5. Matrix showing significant difference between stages using Scheffe's post-hoc method (*=p<.05, **=p<.01) (mean is expressed in mmol/L).

Hemoglobin and hematocrit both increased simultaneously throughout the incremental exercise (Table 2). Hemoglobin increased from a mean of 149 g/L (±8.7) at stage one to 160 g/L (±9.0) at stage six. During recovery hemoglobin decreased slightly to a mean of 158 (±8.1 g/L). Hematocrit increased from 0.44 (±.03) at stage one to 0.47 (±.03) at stage six. Hematocrit decreased slightly to .47 (±.02) during recovery, but remained elevated above resting values.

Bicarbonate and pH both decreased throughout the exercise protocol (Table 2). As exercise intensity increased, bicarbonate values decreased from a mean of 28.2 (±1.7) at stage one to 20.3 (±3.2) and 13.7 (±2.7) at stage six and recovery, respectively. Similarly, pH declined from 7.34 (±.02) at stage one to 7.21 (±.05) at stage six and 7.20 (±.05) during recovery.

A correlation matrix was computed for data from rest to 100% $P\dot{V}O_2$ and is presented in Table 6. All correlations presented are significant at $p< 0.01$. Correlation matrices were also computed for all variables at each stage (Appendix A).

Variable	WBLa	PL	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.98	1.0				
Hb	0.49	0.52	1.0			
Hct	0.51	0.53	0.95	1.0		
pH	-0.83	-0.86	-0.46	-0.49	1.0	
HCO_3	-0.87	-0.85	-0.31	-0.35	0.63	1.0

Table 6. Correlation matrix of all data combined from rest to 100% $P\dot{V}O_2$.

Table 6 does not reflect the relation between variables across the exercise stages. Bicarbonate was not significantly correlated with whole blood lactate nor plasma lactate at stages one and two, but was significant at stages 3 through 6 (Appendix A).

Regression Analysis: Part 1.

Using plasma lactate as the criterion variable and whole blood lactate as the predictor variable, a simple regression equation was computed for each stage (Appendix B), and with all data pooled together (Figure 2).

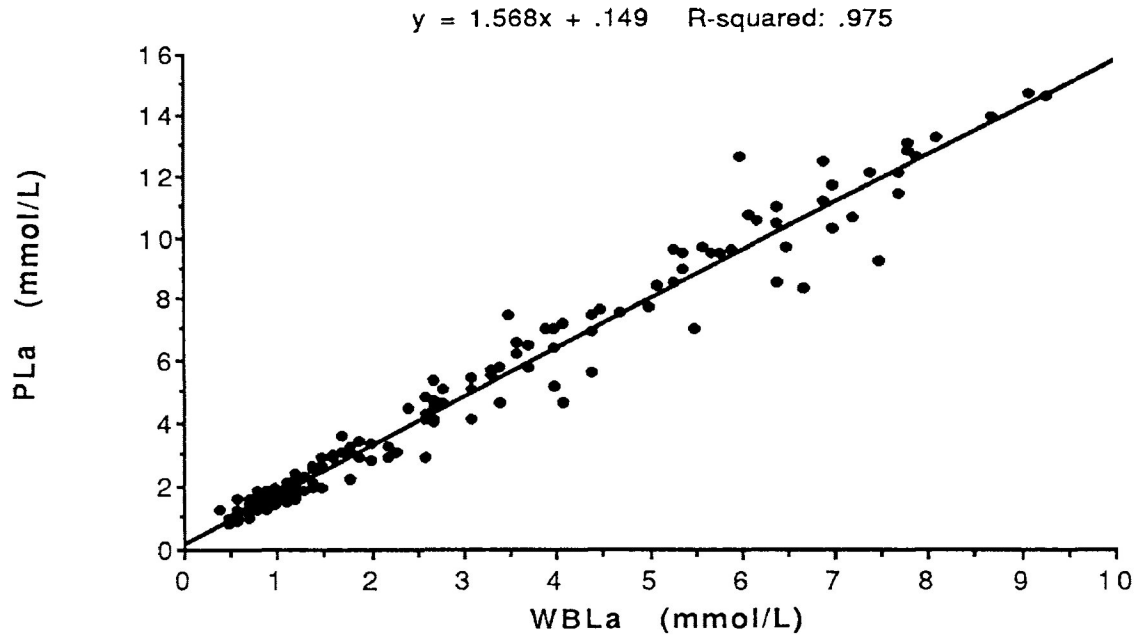


Figure 2. Scattergram and simple regression equation of all data points from rest to 100% Peak $\dot{V}O_2$ (Whole Blood and Plasma Lactate).

The simple regression equation for all data points pooled is

$$y=1.568(WBLa) + .149 \qquad \text{(Equation 1)}.$$

The R-squared value was .975 which suggests that 97.5% of the variability in plasma lactate is directly predictable from the variability in whole blood lactate. The standard error was .558. The standard error of estimate is a measure of how points deviate from the regression line and the smaller values of standard error reflect points that stay close to the regression line, while larger values indicate a greater dispersion of points away from the regression line. For simple regression analysis of all other stages refer to Appendix B.

Regression Analysis: Part 2.

Multiple regression analyses were performed on all stages and are presented in Appendix C. To determine if other predictors could

add to the predictive power of equation 1, a backward stepwise multiple regression was computed on all data points combined. The dependent variable was plasma lactate. The independent variables were whole blood lactate, hemoglobin, hematocrit, bicarbonate and pH. Each of the five predictor variables were included in the first regression analysis. The R-squared and standard error were 0.98 and .504, respectively (Table 7). The predictor that contributed the least to the equation (F-ratio) was eliminated and subsequent analyses were then performed. None of the other predictors greatly aided the predictive power of Equation 1 as indicated by the slight changes in the R-squared and standard error values (Appendix C).

DF	R-squared	Standard Error
218	.98	.504

Analysis of Variance for Regression

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	2631.879	526.376	2075.908
RESIDUAL	213	54.009	.254	$p < 0.0001$
TOTAL	218	2685.888		

Regression Results

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	44.417	9.216	4.82	
WBLa	1.437	.051	28.086	788.849
Hb	.016	.013	1.192	1.422
Hct	-1.088	4.552	-.239	.057
pH	-6.342	1.213	-5.23	27.354
HCO_3	.019	.021	.904	.817

Table 7. Multiple regression analysis of all data from rest-100% Peak $\dot{V}O_2$ including all five variables.

Regression Analysis: Part 3.

A third focus of this study was to investigate the relationship between total blood, whole blood, and plasma lactate. Total blood lactate data is presented separately because only five subjects were used to obtain total blood lactate values (40 data points). The mean age, height, weight and PVO_2 for the five subjects who provided total blood lactate data are presented in Table 8.

Variable	Mean	±S.D.	Minimum	Maximum
Age (yrs)	20.4	±2.51	16	22
Height (cm)	179.0	±4.89	173.1	186.5
Weight (kg)	75.4	±5.70	68.7	83.3
PVO2 (ml/kg/min)	49.0	±10.40	33.3	61.8

Table 8. Descriptive statistics of physical characteristics and Peak $\dot{V}O_2$ of Five Male Subjects.

Due to the small number of subjects the data obtained for total blood lactate was not divided into stages according to $P\dot{V}O_2$.

Total blood lactate (TBLa) concentrations paralleled the increase of whole blood and plasma lactate values throughout the incremental exercise. The mean total blood lactate value at rest, immediately post exercise, and after 5 minutes recovery were: 1.02 (±.179), 8.5 (±2.122) and 10.4 (±1.494) mmol/L, respectively. Paired t-tests between total blood, whole blood and plasma lactate were computed. The t-tests revealed significant differences ($p < 0.0005$) between lactate measurements. In all cases total blood lactate fell between the higher plasma lactate and the lower whole blood lactate. A mean difference of .788 mmol/L separated all total blood

lactate values from plasma lactate values while a mean difference of .362 mmol/L separated total blood lactate from whole blood lactate. This is represented graphically in Figure 3.

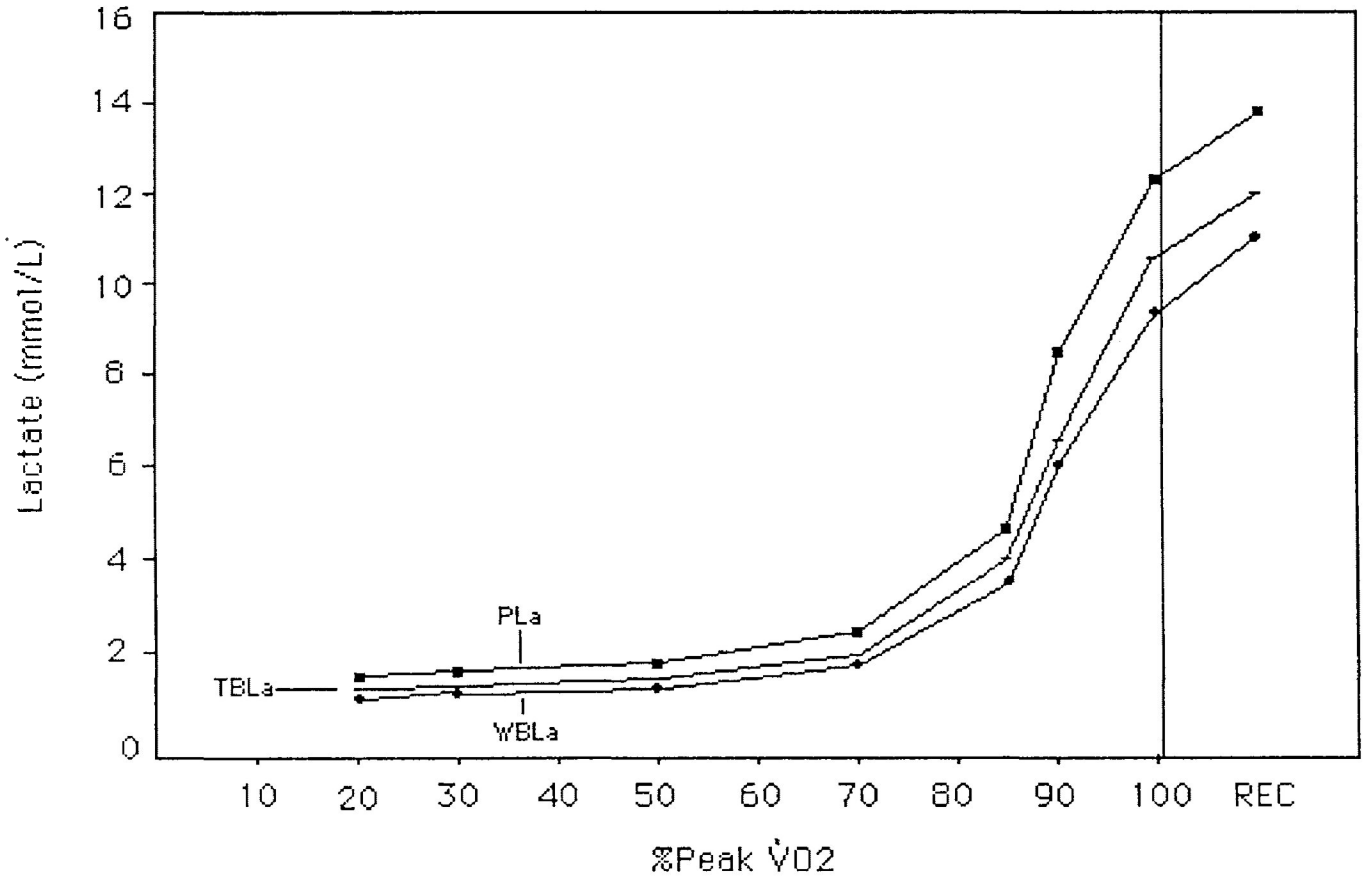


Figure 3. Typical example of the changes in whole blood lactate (WBLa), total blood lactate (TBLa), and plasma lactate (PLa) during an incremental exercise test.

Using whole blood and plasma lactate as the criterion variables and total blood lactate as the predictor variable two simple regression equations were computed.

The equation for predicting whole blood lactate from total blood lactate was

$$y=.887(TBLa) + .015 \qquad \text{(Equation 2)}$$

The R-squared value was .993 and the standard error was .225. These values suggest that 99.3% of the variability in whole blood is directly predictable from the variability in total blood lactate (Figure 4).

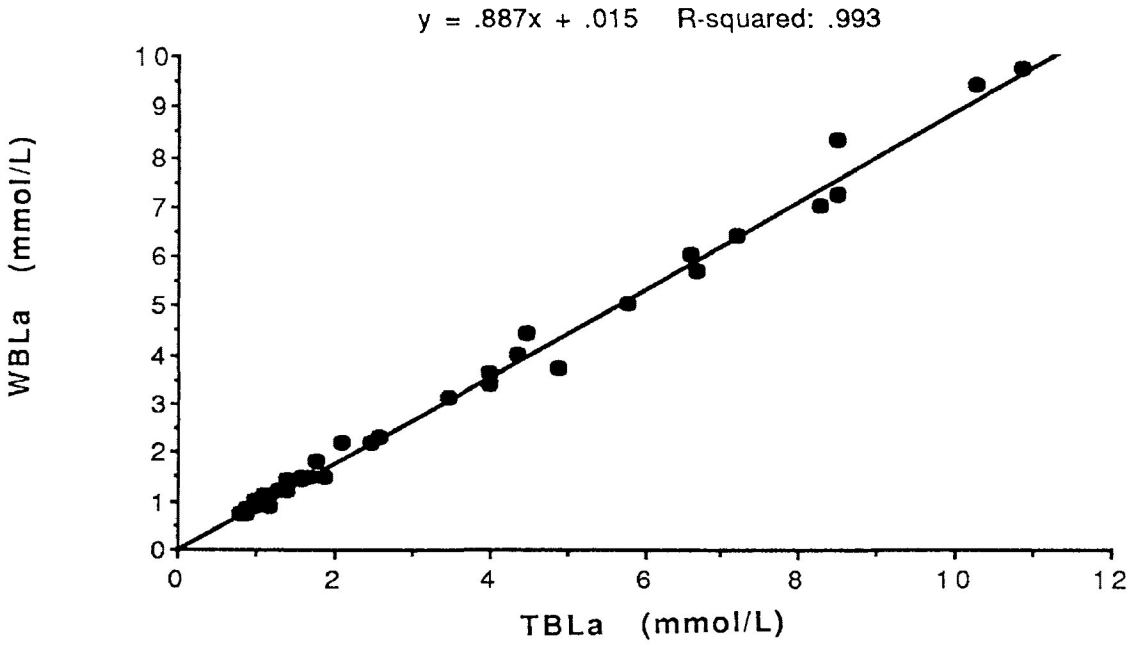


Figure 4. Scattergram and simple regression equation of all data points (total and whole blood lactate) from rest to 100% Peak $\dot{V}O_2$.

The equation computed for predicting plasma lactate from total blood lactate is

$$y=1.208(TBLa) + .093 \qquad \text{(Equation 3)}$$

R-squared was .996 and the standard error was .217 (Figure 5). Since such a high predictability exists between these three variables, it is not necessary to include other predictor variables.

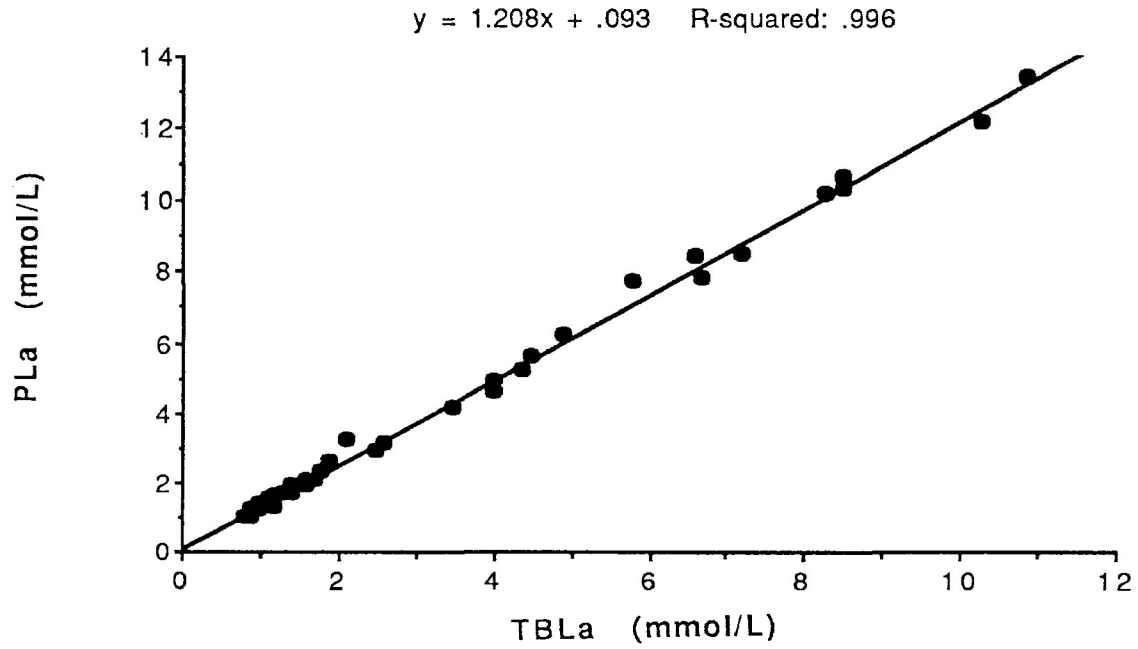


Figure 5. Scattergram and simple regression equation of all data points (total blood and plasma lactate) from rest to 100% Peak $\dot{V}O_2$.

Chapter 5

Discussion

It is well established that lactate concentrations in blood rise with an increase in exercise intensity and reach peak values during 5 to 8 minutes of recovery (Forrest et al., 1990; Harris & Dudley, 1989; Saltin, 1990). The three lactate measurements (whole blood, total blood and plasma lactate) in the current study also followed this pattern.

While the pattern for all three lactate measurements was similar, the degree of change was different. Mean plasma lactate values were consistently and significantly higher than the mean whole blood lactate concentrations at all exercise intensities, but the discrepancy increased at higher intensities. Harris and Dudley (1989) reported a similar trend in that plasma lactate concentrations immediately after high intensities of exercise were 4.3 mmol/L greater than those of whole blood lactate. In the present study the immediate post exercise mean whole blood lactate value was 6.2 (±1.65) mmol/L while the mean plasma lactate was considerably higher (10.0±2.48 mmol/L). During recovery (5 minutes post exercise) both indices continued to increase with the mean value of whole blood lactate being 8.5±1.47 mmol/L and plasma lactate rising to 13.4±2.16 mmol/L. The present results differ slightly from Buono & Yeager (1986) who reported no significant difference between whole blood and plasma lactate at 25% and 50% $\dot{V}O_2$max. Both studies are in agreement though with the situation at higher intensities.

The point at which plasma lactate begins to diverge from it's parallel tracking of whole blood lactate appears to be after 59% $P\dot{V}O2$. This becomes apparent when considering the ANOVA run on difference scores between plasma and whole blood lactate across the six stages of exercise (Table 5).

From stage one to stage two of exercise intensity, no statistical difference exists between the difference scores of whole blood and plasma lactate. After stage two there is a significant ever widening gap between the two indices.

Harris and Dudley (1989) reported high correlations between whole blood and plasma lactate values at rest, post exercise, and during recovery. In the current study whole blood and plasma lactate were highly and significantly correlated throughout all exercise intensities. Interestingly though, correlations between whole blood and other predictor variables were not significantly correlated throughout all exercise stages. For example whole blood and bicarbonate were not statistically correlated at stage one (rest-49% $P\dot{V}O_2$) and stage two (50-59% $P\dot{V}O_2$), but were statistically significant at stage three, four, five, six, and during recovery. Plasma lactate and bicarbonate followed a similar pattern. Similarly, whole blood and plasma lactate were not significantly correlated with pH until stage six and during recovery.

According to Hultman and Sahlin (1980), during low to moderate exercise intensity, the body maintains an acid base equilibrium by way of bicarbonate buffering. At high exercise intensities there is an increase in blood lactate concentrations and a simultaneous release in H^+ ions of from the muscle and a subsequent

increase of hydrogen ions in the blood (Hughson & Green, 1982). Until the blood is saturated with hydrogen ions, there is not a significant change in pH because of the bicarbonate buffering system, however when hydrogen ions are abundant and have bound to nearly all bicarbonate, a decrease in pH results. This physiological explanation seems to fit the data from this study. Since bicarbonate is continuously buffering the H^+ ions there is no significant change in pH. At higher, near exhaustion levels of exercise intensity, bicarbonate buffering has reached its capacity (Wasserman et al., 1981 and Sahlin et al., 1978) and therefore a decrease in pH results.

Plasma Volume

Davidson, Robertson, Galea and Maughan (1987) and Dickson, Wilkinson and Noakes (1982) report increases in hemoglobin concentration, hematocrit and red blood cells in peripheral blood during single efforts of exercise. Increases in both hemoglobin and hematocrit were evident in the present study. The mean hematocrit value at stage one was 0.44±.03 L/L and 0.47±.03 L/L at stage six. Hemoglobin changed from 149±8.7 g/L at stage one to 160±9.0 g/L. Using the method of Dill and Costill (1974), the mean plasma volume change from rest to immediately post exercise was a 14.7% decrease. These findings are similar to those reported by Wells, Stern, Kohrt, and Campbell (1987). Wells et al. (1987) reported a 11-18% decrease in plasma volume during treadmill exercise with workloads from 33-90% $\dot{V}O_2$max. Wilkerson, Gutin and Horvath (1977) examined the changes in plasma volume over a wide range of exercise intensities using the method of Dill and Costill (1974).

These authors reported a decrease in plasma volume with an increased oxygen uptake and increased workload which supports the findings of the present study.

While it is well established how plasma volume changes during exercise, how plasma volume change effects lactate concentrations has not yet been established. The results from this study suggest that plasma volume change during incremental exercise of approximately 17 minutes, on a bicycle ergometer, has very little effect on lactate concentrations. Hemoglobin and hematocrit were used as second predictor variables in multiple regression analysis. Both these variables contributed very little to the prediction equation. Since plasma volume change, as calculated using the method of Dill and Costill (1974), is based on hemoglobin and hematocrit values, it would therefore suggest that plasma volume change does not confound the interpretation of lactate concentrations.

Simple Regression

Harris and Dudley (1989) developed a regression equation to predict plasma lactate from whole blood lactate from data immediately post exercise. The equation computed by these authors is

Plasma lactate (mmol/L) = Whole blood lactate (mmol/L) x 1.43 - 1.11 ($r=0.95$, $p< 0.001$). A similar equation was computed in the present study using data from stage six (90-100% $P\dot{V}O_2$) which would include data immediately post exercise.

The equation computed in this particular study is Plasma lactate (mmol/L)=Whole blood lactate (mmol/L) x 1.466 + 1.126 ($r=.96$, $p<0.001$). It is interesting to note that the intercepts and slopes of these two equations are similar. The difference though is the sign before the slope value in each of the equations. The mean whole blood and plasma lactate values at stage six were 6.2±1.65 mmol/L and 10.0±2.48 mmol/L respectively. Assuming this whole blood lactate value, the predicted plasma lactate concentration using the equation presented by Harris & Dudley (1989) is 7.8 mmol/L. Using the equation from the present research, the predicted plasma lactate concentration is 10.2 mmol/L. Thus the present study suggests a greater discrepancy between whole blood and plasma lactate at high levels of exercise intensities. However, the difference in the equations could very well be a result of many factors. For instance, Harris & Dudley (1989) exercised their subjects at higher intensities (supramaximal) for a shorter duration. The number of data points used to develop the equation could also account for some of the difference. Harris and Dudley (1989) used six subjects for a total of six data points to generate the regression equation. In the present study, thirty-seven data points were used to generate the regression equation. An increased number of data points increases the power of the prediction equation.

Following the same logic, all data points from rest to 100% $P\dot{V}O_2$ were pooled to compute one equation to predict plasma lactate from whole blood lactate at any exercise intensity up to 100% $P\dot{V}O_2$. The equation generated is Plasma lactate (mmol/L)=Whole blood lactate (mmol/L) x 1.568 + .149. Using the mean whole blood lactate

value as above (6.2 mmol/L), the predicted plasma lactate value is 9.9 mmol/L. There is thus not a large change in the predicted value whether all data points or just stage six data points are used. Both equations were just 0.1 mmol/L off of the true value.

Harris and Dudley (1989) pooled rest and recovery (15 minutes post exercise) data and developed a second equation that was reported as significantly different from the equation computed from immediately post exercise data. These authors suggest that the condition under which blood is sampled influences the nature of the relation between whole blood and plasma lactate concentrations. This is an accurate statement based upon the fact that these authors took recovery samples at fifteen minutes post exercise. The relationship between whole blood and plasma lactate at five minutes recovery follows the pattern established during the exercise stages. It is thus possible to extrapolate and use the equation generated from all data points up to 100% $P\dot{V}O_2$ and to predict plasma lactate values at 5 minutes post exercise. To illustrate this point, the mean 5 minute recovery whole blood and plasma lactate values are 8.5 (±1.47) mmol/L and 13.4 (±2.16) mmol/L, respectively, and the predicted plasma lactate value derived from the overall equation is 13.5 mmol/L. The robustness of this equation for predicting lactate values from rest to 100% $P\dot{V}O_2$ to 5 minute post exercise is of practical significance to coaches who may wish to equate lactate values

Multiple Regression

The extremely high correlation between whole blood and plasma lactate precluded the contribution that other predictor variables could possibly make. This is despite the fact that other physiological variables also follow a predictable pattern through an incremental exercise test. Forrest, Morton and Lambardarios (1990) reported a 10 percent difference in whole blood and plasma lactate prior to exercise and a 30 percent difference in these two indices immediately after exercise (half marathon) and suggest that as hematocrit rises due to some dehydration that the difference between whole blood and plasma lactate will increase. For this reason, hematocrit and hemoglobin were included as predictor variables when computing the regression equations. Based on a stepwise multiple regression analysis, neither hemoglobin or hematocrit contributed to the predictive power of the equation. Hematocrit and hemoglobin may be better predictors over a longer duration exercise protocol.

Bicarbonate and pH were also included in the multiple regression analysis and contributed little to the equation. As stated previously, the high correlation and predictability between whole blood and plasma lactate could limit the contribution of other predictor variables to the regression equation. Although pH did not add to the prediction equation, it is possible that pH could contribute to a mechanism which is responsible for the discrepancy between these indices. More research is needed to identify mechanisms responsible for these discrepancies.

Total Blood Lactate

The Yellow Springs Instrument (YSI) Model 23L Lactate Analyzer is capable of analyzing whole blood, plasma, and total blood lactate samples. Total blood lactate values are arrived at by lysing the red blood cell membrane which expels any intraerythroctye lactate as well as other components. Little research has been published which reports total blood lactate values during incremental exercise testing.

Total blood lactate values paralleled the increase of whole blood and plasma lactate concentrations throughout the incremental exercise. In all cases total blood lactate fell between the higher plasma lactate and the lower whole blood lactate concentrations.

Buono, Cook and Clancy (1984) measured lactate levels in subjects who performed an incremental bicycle ergometer test to exhaustion. The total blood lactate values at rest, immediately post exercise and during 5 minutes recovery in the present study were very similar to those reported by Buono et al. (1984). At all three time periods, the lactate values reported are within 1.3 mmol/L between the two studies.

Since total blood lactate values are significantly different from whole blood and plasma lactate, and are an indice measured by the YSI analyzer, it is thus desirable to develop regression equations to predict both whole blood and plasma lactate from total blood lactate (Equation 2 and 3).

To illustrate the relationship between total blood, plasma and whole blood lactate, a total blood lactate value of 4 mmol/L equates to a whole blood lactate value of 3.6 mmol/L (Equation 2) and a

plasma lactate value of 4.9 mmol/L (Equation 3). The true values of whole blood and plasma lactate at this point are 3.4 and 4.6 mmol/L. respectively. The practical significance of this similarity is that total blood lactate measurements may be the lactate measurement of choice for coaches using the YSI Lactate Analyzer. It offers the most direct comparison to the complex Sigma method.

Chapter 6

Summary, Conclusions and Recommendations

Summary

The purpose of this study was to examine the relationship between whole blood, plasma, and total blood lactate levels at various exercise intensities. Regression equations were thus computed to predict both plasma lactate from whole blood lactate concentrations and to predict whole blood and plasma lactate concentrations from total blood lactate values. Hemoglobin, hematocrit, bicarbonate and pH were included in multiple regression analysis to determine if these variables contribute to the power of the prediction equations.

It was established that at low levels of exercise intensity, no statistical difference exists between the different scores of whole blood and plasma lactate. As exercise intensity increases though, the difference between whole blood and plasma lactate becomes greater.

The three lactate indices (whole blood, plasma and total blood lactate) were all significantly different from each other at all exercise intensities. Simple regression equations were computed across all stages of exercise and with all data points pooled together to predict plasma lactate from whole blood lactate concentrations. Multiple regression analyses were then performed on the same data and it was concluded that there is a high correlation and predictability between whole blood and plasma

lactate and therefore the contribution of other predictor variables is limited.

Conclusions

The current study, in conjunction with others (Buono & Yeager, 1986; Harris & Dudley, 1989; Forrest et al., 1990) illustrates that differences exist between whole blood, plasma and total blood lactate and that as exercise intensity increases, so does the discrepancy. From a coaches perspective, these discrepancies have some far reaching implications. Without knowledge of these discrepancies and a means for equating them coaches would be unable to draw valid comparisons between their athlete's values and values reported in the literature. The regression equations resulting from the present research will assist coaches and researchers in this regard when comparing whole blood, plasma and total blood lactate values obtained using the YSI Model 23L Lactate Analyzer.

Recommendations

Although the present research clearly identified the fact that discrepancies exist between whole blood, plasma and total blood, it is not known what mechanism(s) contribute to this difference. Future research could investigate potential mechanisms.

Additional research could examine the relationship between whole blood, plasma, and total blood lactate using different protocols. For example, treadmill running, swimming and longer and shorter durations of exercise.

The present research was delimited to YSI instrumentation. Future research could further compare the YSI method of lactate determination to other methods such as Sigma (Sigma Chemicals, St. Louis, MO).

APPENDIX A.

Correlation matrices for all dependant variables at different stages of exercise

Stage One

Variables	WBLa	PLa	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.85	1.0				
Hb	- - -	0.31	1.0			
Hct	- - -	0.27	0.96	1.0		
pH	- - -	- - -	- - -	- - -	1.0	
HCO_3	- - -	- - -	- - -	- - -	- - -	1.0

Correlation matrix for stage one (rest - 49% PVO_2). Data presented in matrix is significant at $p<0.01$.

Stage Two

Variables	WBLa	PLa	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.78	1.0				
Hb	- - -	0.51	1.0			
Hct	- - -	- - -	0.85	1.0		
pH	- - -	- - -	- - -	- - -	1.0	
HCO_3	- - -	- - -	- - -	- - -	- - -	1.0

Correlation matrix for stage two (50-59% PVO_2). Data presented in matrix is significant at $p<0.01$.

Stage Three

Variables	WBLa	PLa	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.90	1.0				
Hb	- - -	- - -	1.0			
Hct	- - -	- - -	0.94	1.0		
pH	- - -	- - -	- - -	- - -	1.0	
HCO_3	-0.62	-0.57	- - -	- - -	- - -	1.0

Correlation matrix for stage three (60-69% PVO_2). Data presented in matrix is significant at $p<0.01$.

Stage Four

Variables	WBLa	PLa	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.92	1.0				
Hb	- - -	- - -	1.0			
Hct	- - -	- - -	0.92	1.0		
pH	- - -	- - -	- - -	- - -	1.0	
HCO_3	-0.66	-0.67	- - -	- - -	- - -	1.0

Correlation matrix for stage four (70-79% PVO_2). Data presented in matrix is significant at $p<0.01$.

Stage Five

Variables	WBLa	PLa	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.92	1.0				
Hb	- - -	- - -	1.0			
Hct	- - -	- - -	0.91	1.0		
pH	- - -	- - -	- - -	- - -	1.0	
HCO_3	-0.90	-0.84	- - -	- - -	- - -	1.0

Correlation matrix for stage five (80-89% PVO_2). Data presented in matrix is significant at $p<0.01$.

Stage Six

Variables	WBLa	PLa	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.92	1.0				
Hb	- - -	0.41	1.0			
Hct	- - -	0.44	0.96	1.0		
pH	-0.58	-0.64	-0.45	-0.47	1.0	
HCO_3	-0.70	-0.58	- - -	- - -	- - -	1.0

Correlation matrix for stage six (90-100% PVO_2). Data presented in matrix is significant at $p<0.01$.

Recovery

Variables	WBLa	PLa	Hb	Hct	pH	HCO_3
WBLa	1.0					
PLa	0.80	1.0				
Hb	- - -	- - -	1.0			
Hct	- - -	- - -	0.96	1.0		
pH	-0.72	-0.83	- - -	- - -	1.0	
HCO_3	-0.81	-0.64	- - -	- - -	0.62	1.0

Correlation matrix for recovery. Data presented in matrix is significant at $p<0.01$.

APPENDIX B

Simple regression analysis of whole blood and plasma lactate at different stages of exercise.

Stage One

DF	R-squared	Standard Error
82	.725	.198

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	1	8.373	8.373	213.472
RESIDUAL	81	3.177	.039	$p < 0.0001$
TOTAL	82	11.550		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T
INTERCEPT	.355	.076	4.68
WBLa	1.262	.086	14.611

Stage Two

DF	R-squared	Standard Error
33	.609	.287

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	1	4.120	4.12	49.86
RESIDUAL	32	2.644	.083	$p < 0.0001$
TOTAL	33	6.765		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T
INTERCEPT	.488	.213	2.286
WBLa	1.277	.181	7.061

Stage Three

DF	R-squared	Standard Error
27	.807	.395

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	1	16.929	16.929	108.381
RESIDUAL	26	4.061	.156	$p < 0.0001$
TOTAL	27	20.990		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T
INTERCEPT	.947	.185	5.132
WBLa	1.053	.101	10.411

Stage Four

DF	R-squared	Standard Error
28	.851	.501

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	1	38.791	38.791	154.314
RESIDUAL	27	6.787	.251	$p < 0.0001$
TOTAL	28	45.579		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T
INTERCEPT	.562	.285	1.974
WBLa	1.443	.116	12.422

Stage Five

DF	R-squared	Standard Error
26	.840	.731

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	1	70.031	70.031	131.083
RESIDUAL	25	13.356	.534	$p < 0.0001$
TOTAL	26	83.387		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T
INTERCEPT	1.580	.410	3.854
WBLa	1.151	.101	11.449

Stage Six

DF	R-squared	Standard Error
37	.919	.713

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	1	207.945	207.945	408.946
RESIDUAL	36	18.306	.508	$p < 0.0001$
TOTAL	37	226.251		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T
INTERCEPT	1.126	.467	2.410
WBLa	1.466	.072	20.222

Recovery

DF	R-squared	Standard Error
26	.639	1.326

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	1	77.685	77.685	44.164
RESIDUAL	25	43.975	1.759	$p < 0.0001$
TOTAL	26	121.660		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T
INTERCEPT	3.252	1.528	2.127
WBLa	1.177	.177	6.646

APPENDIX C

Multiple regression analysis of plasma lactate and predictor variables at different stages of exercise.

Stage One.

DF	R-squared	Standard Error
81	.783	.181

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	9.001	1.8	54.696
RESIDUAL	76	2.501	.033	$p < 0.0001$
TOTAL	81	11.503		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	3.063	7.333	.418	
WBLa	1.231	.086	14.286	204.095
Hb	.009	.008	1.139	1.296
Hct	-1.165	2.723	-.428	.183
pH	-.617	.989	-.624	.389
HCO_3	.036	.012	2.934	8.608

Stage Two (50-59% $P\dot{V}O_2$).

DF	R-squared	Standard Error
28	.746	.191

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	2.466	.493	13.478
RESIDUAL	23	.842	.037	$p < 0.0001$
TOTAL	28	3.308		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	-20.384	15.19	-1.342	
WBLa	.953	.158	6.038	36.453
Hb	.032	.017	1.937	3.751
Hct	-5.462	5.359	-1.019	1.039
pH	2.462	2.015	1.222	1.493
HCO_3	.025	.029	.850	.723

Stage Three (60-69% $P\dot{V}O_2$).

DF	R-squared	Standard Error
24	.824	.433

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	16.682	3.336	17.814
RESIDUAL	19	3.558	.187	$p < 0.0001$
TOTAL	24	20.240		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	18.609	30.247	.615	
WBLa	.996	.191	5.216	27.210
Hb	.013	.037	.365	.133
Hct	-3.475	12.624	-.275	.076
pH	-2.350	3.958	-.594	.352
HCO_3	-.031	.064	-.486	.236

Stage Four (70-79% $P\dot{V}O_2$).

DF	R-squared	Standard Error
21	.869	.518

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	28.527	5.705	21.236
RESIDUAL	16	4.299	.269	$p < 0.0001$
TOTAL	21	32.826		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	13.174	37.979	.347	
WBLa	1.134	.205	5.543	30.728
Hb	.009	.045	.191	.037
Hct	5.415	17.222	.314	.099
pH	-1.843	5.145	-.358	.128
HCO_3	-.088	.069	-1.277	1.632

Stage Five (80-89% $P\dot{V}O_2$).

DF	R-squared	Standard Error
21	.856	.768

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	56.332	11.266	19.082
RESIDUAL	16	9.447	.590	$p < 0.0001$
TOTAL	21	65.778		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	14.500	57.506	-.252	
WBLa	1.045	.293	3.564	12.704
Hb	.042	.077	.550	.302
Hct	-10.483	23.470	-.447	.199
pH	2.167	7.543	.287	.083
HCO_3	-.051	.150	-.339	.115

Stage Six (90-100% $P\dot{V}O_2$).

DF	R-squared	Standard Error
37	.947	.611

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	214.291	42.858	114.677
RESIDUAL	32	11.959	.374	$p < 0.0001$
TOTAL	37	226.251		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	9.219	21.51	.429	
WBLa	1.479	.134	11.055	122.204
Hb	.059	.037	1.565	2.449
Hct	-10.646	13.955	-.763	.582
pH	-1.952	2.776	-.703	.494
HCO_3	.075	.055	1.382	1.909

Recovery (5 minutes post-exercise).

DF	R-squared	Standard Error
19	.792	1.242

Analysis of Variance

	DF	Sum Squares	Mean Square	F-value
REGRESSION	5	82.418	16.484	10.687
RESIDUAL	14	21.594	1.542	$p < 0.0001$
TOTAL	19	104.012		

Beta Coefficient Table

	Regression Coefficient	Standard Error	T	Partial F
INTERCEPT	221.089	84.18	2.626	
WBLa	.545	.432	1.263	1.594
Hb	.004	.133	.027	.001
Hct	12.548	45.189	.278	.077
pH	-30.437	11.369	-2.677	7.167
HCO_3	.016	.181	.086	.007

Appendix D

Lakehead University
School of Physical Education and Athletics
Human Performance Laboratory

Dr. Bob Thayer
343-8653

Donna Newhouse
343-8187 or
344-0786

Subject Consent Form

At the present time lactate analysis is used extensively by coaches and researchers to determine whole blood lactate levels as an indicator of exercise intensity. However, preliminary research at Lakehead University suggests that there is a discrepancy between whole blood and plasma lactate levels. At rest and low intensity exercise, plasma and whole blood lactate levels are fairly equipoise. As exercise intensity increases the distribution of lactate changes and a disequilibrium occurs with plasma lactate concentrations being greater than whole blood (Thayer & Allan, 1990). Therefore, based upon this evidence the purpose and objectives of this research are:

1. to examine the relationship between whole blood and plasma lactate concentrations at various percentages of VO_2max during incremental exercise to exhaustion

2. to develop regression equations to predict plasma lactate and total blood lactate concentrations from whole blood lactate values at various percentages of VO_2max

3. to investigate the physiological rationale behind a discrepancy between whole blood lactate and plasma lactate should one exist

Subjects will perform an incremental workload test on a cycle ergometer until voluntary exhaustion. Expired gases will be collected during the exercise test to aid in the determination of VO_2max (maximal amount of oxygen consumed). The length of the test will be approximately 10 to 15 minutes. Subjects may experience a feeling of slight discomfort due to fatigue. Results

from this test will provide subjects with a measure of their cardiovascular fitness level.
Blood samples will be taken before, during and after exercise. Blood sampling involves the insertion of an indwelling catheter in a forearm vein. This simple procedure will be performed by a qualified technician and it may cause slight discomfort and bruising at the point of insertion. The amount of blood drawn is approximately 150 ml. Blood will be assayed for whole blood lactate, plasma lactate, total blood lactate, hematocrit, pH, HCO_3,and pCO_2.

You will be asked to report to the laboratory having refrained from intense exercise for at least twenty-four hours. It is recommended that you do not eat for two hours prior to your test. Height and weight will be recorded at this time.

Individual results will be held confidential to the researchers as subjects will be referenced by number in any form of publication.

I have read and understand the above mentioned purposes and procedures of this study. In agreeing to be a participant, I assume all responsibilities, and waive any claims resulting from personal injuries, against Lakehead University or any member of the research team. I also understand that I am free to withdraw from any part of the study at any time.

_____
Signature of Participant

_____
Witness

_____
Date

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