The effects of β-alanine supplementation in aerobic exercise

A way to delay the onset of muscular fatigue?

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Abstract

Muscle fatigue has always been of vital importance in most sports (Mohr, Krstrup & Bangsbo, 2005). A few possible factors have been reported to be the cause of muscular fatigue during high intensity exercise; depletion of glycogen, oxidative stress, disruption of contractile mechanisms and accumulation of metabolites (Begun et al., 2005; Nimmo & Ekblom, 2007). One of the theories of the cause of muscular fatigue, both in endurance and intermittent sports, is decreased pH levels due to increased concentration of H⁺ ions dissociated from lactic acid in muscle (Derave et al., 2007). Carnosine, a fairly unnoticed ergogenic aid, taken in the form of β-alanine has shown to potentially delay the onset of fatigue. Supplementation of β-alanine, would increase carnosine levels in muscle and may counteract the decrease in pH since carnosine functions as a H⁺ buffer (Derave et al., 2007). The purpose of the present study was to examine the effects of 8 weeks supplementation of β-alanine in distance runners and Swedish division four soccer players on aerobic capacity, intermittent recovery and muscular fatigue. The runners (n = 15) were tested in lactate profiling tests and the soccer players (n = 22) were tested in the Yo-Yo intermittent endurance test pre and post the 8-week test-period. The yo-yo test did not result in significant difference between the soccer players’ β-group and control-group (p = 0,29). Neither did the lactate test result in significant differences between the distance runners’ β-group and control-group in any of the five variables measured. However, a trend in difference was seen between groups in both velocity at lactate threshold (V_LT) (p = 0,11) and recovery blood lactate (RBL) (p = 0,14) where the β-group had increased slightly from 16,8 ± 1,6 km/h to 17,0 ± 1,2 km/h in V_LT and decreased from 4,5 ± 1,6 mmol∙L⁻¹ to 3,1 ± 1,0 mmol∙L⁻¹ in RBL. The results suggested that β-alanine may delay the onset of fatigue and improve performance in endurance sports such as running by increasing the removal of lactate acid from muscle.
Contents

1 Introduction .................................................................................................................. 5
  1.1 Muscular fatigue ................................................................................................. 5
  1.2 Carnosine and β-alanine ..................................................................................... 7
    1.2.1 Carnosine and performance ......................................................................... 7
  1.3 Evaluation and requirements of sports ............................................................... 8
    1.3.1 Distance running ......................................................................................... 8
    1.3.2 Soccer ......................................................................................................... 11

2 Purpose and Objective ................................................................................................. 13
  2.1 Aim ...................................................................................................................... 13

3 Materials and methods ............................................................................................... 14
  3.1 Information retrieval ......................................................................................... 14
  3.2 Subjects ............................................................................................................... 14
  3.3 Experimental protocol ....................................................................................... 14
    3.3.1 The Yo-Yo intermittent endurance test protocol ......................................... 14
    3.3.2 Lactate testing protocol .............................................................................. 15
    3.3.3 Equipment .................................................................................................. 15
  3.4 Statistical analysis ............................................................................................... 16

4 Results ......................................................................................................................... 17
  4.1 The Yo-Yo intermittent endurance test ............................................................... 17
  4.2 Lactate test ......................................................................................................... 17
    4.2.1 Velocity at lactate threshold (VLT) ............................................................. 17
    4.2.2 Heart rate at lactate threshold (HRLT) ......................................................... 18
    4.2.3 Blood lactate at lactate threshold (BLLT) .................................................... 18
    4.2.4 RPE at lactate threshold (RPELT) ............................................................... 18
    4.2.5 Recovery blood lactate (RBL) ..................................................................... 19
  4.3 Training diary ...................................................................................................... 19

5 Discussion ................................................................................................................... 20
  5.1 Discussion of results ........................................................................................... 20
    5.1.1 The Yo-Yo intermittent endurance test ....................................................... 20
    5.1.2 Lactate test ................................................................................................ 21
    5.1.3 Mechanisms of muscular fatigue ............................................................... 22
  5.2 Discussion of methods and materials ................................................................. 22
    5.2.1 The Yo-Yo intermittent endurance test ....................................................... 22
    5.2.2 Lactate test ................................................................................................ 22

6 Conclusion .................................................................................................................. 24

7 Acknowledgements ..................................................................................................... 24
References...........................................................................................................................................25
Appendix 1 ........................................................................................................................................28
  Information to subjects .........................................................................................................................28
Appendix 2 ........................................................................................................................................31
  Written consent .................................................................................................................................31
Appendix 3 ........................................................................................................................................32
  Lactate test protocol .........................................................................................................................32
Appendix 4 ........................................................................................................................................33
  Yo-Yo Intermittent Endurance test protocol .....................................................................................33
1 Introduction

Muscle fatigue is and has always been of vital importance in most sports (Mohr, Krustrup & Bangsbo, 2005). During moderate to high intensity exercise, muscular fatigue will reflect performance and thereby influence the result of an exercise session, game or competition (Royal et al., 2006; Oliver, Armstrong & Williams, 2008). Several investigators have researched the components of muscular fatigue. To date there are numerous of theories of the cause of fatigue but no single cause have been found.

Over the years, various supplements have been used to delay the onset of muscular fatigue, though without greater success. The only ergogenic aid supported by scientific studies in human is creatine (Stout et al., 2000). If one could find a legal ergogenic aid that could enhance an athlete’s performance by delaying muscular fatigue it would be a breakthrough in the world of sports. Lately, a fairly unnoticed ergogenic aid, carnosine, taken in the form of β-alanine has shown to potentially delay fatigue (Suzuki, Mukai, Takahashi & Takamatsu, 2007; Begun, Cuncliffe & Leveritt, 2005). Previous authors suggest there is an interesting theoretical base to carry out well-controlled research into any effects that carnosine could have on performance, and that “further research in humans is warranted to provide better understanding of the effects of carnosine in vivo in man” (Begun et al., 2005, p. 506).

The purpose of the present study is to examine the effects of 8 weeks supplementation of β-alanine in distance runners and Swedish division four soccer players on aerobic capacity, intermittent recovery and muscular fatigue. The runners will be tested by lactate profiling tests and the soccer players will be tested in the Yo-Yo intermittent endurance test. The aim of this study is to get preliminary results which can be used to further enhance the knowledge about potential causes of muscular fatigue during exercise and how to increase performance in sports. It will also include a brief review of the causes of muscular fatigue, and needs analyses of the two chosen sports to clarify ways to improve performance in each sport.

1.1 Muscular fatigue

Muscular fatigue is a decline in performance and can happen during all types of muscular activity, both long-term moderate and short-term intense exercise (Begun et al., 2005). Muscular fatigue can be defined as a brief and recoverable reduction in muscle force with continuous or repeated muscle contractions (McKenna, Bangsbo & Renaud, 2008). A few possible factors have been reported to be the cause of muscle fatigue during high intensity exercise; depletion of glycogen, oxidative stress, disruption of contractile mechanisms and accumulation of metabolites (Begun et al., 2005; Nimmo & Ekblom, 2007).

There are some basics of the regulation of fat and carbohydrate metabolism that has to be understood before we get deeper in the theories of fatigue. Fat and carbohydrates are our most important energy sources when the duration of exercise extends beyond 15 seconds. Fat and carbohydrates represent 92% and 2% of our endogenous energy stores, respectively (Jeukendrup, 2003). Fat is stored as adipose tissue and is comparatively unlimited as energy source while carbohydrates are stored in limited amounts as glycogen in liver and muscle. Muscle glycogen stores usually contain about 400g of glycogen, although well trained athletes can store up to 900g after carrying through carbohydrate loading protocol, and the liver contains about 100g of glycogen (Aasen et al., 2006).
The two energy sources, carbohydrates and fat, are always oxidized at the same time, however, the relative contribution depends on a couple of variables such as exercise intensity, exercise duration and aerobic capacity of the athlete. Lipolysis, fat oxidation, is a relatively slow process since it is dependent on the availability of oxygen. Free fatty acids (FFAs) are broken down from adipose tissue, the FFAs oxidizes in the mitochondria and generate ATP. Glycogen on the other hand can generate ATP faster due to the anaerobic pathways. However, the stores of glycogen are as mentioned limited. Glycogen depletion is a major mechanism for fatigue when it comes to moderate and long duration exercise above low intensity. Since fat oxidation is dependent on liver glycogen (for delivery of blood glucose) and the duration of moderate to high exercise intensities are dependent on limited glycogen stores, depleted glycogen stores can onset muscular fatigue (Jeukendrup, 2003). The rate of lipolysis does increase parallel to the intensity of the exercise as long as the intensity is < 65%\(VO_{2\text{max}}\). At intensities above 65%\(VO_{2\text{max}}\) lipolysis will be reduced and a higher ratio of glycogen will be oxidized (Jeukendrup, 2003). As the exercise intensity is kept moderate, the ratio of lipolysis will increase and carbohydrate contribution will decrease with prolonged duration. Hence lipolysis can contribute with 90% of the energy expenditure during prolonged exercise. An athlete with a high aerobic capacity, i.e., high anaerobic threshold, will be able to work at higher intensities for a longer time since the athlete can use fat as fuel even though the intensity is relatively high (Jeukendrup, 2003). In contrast, an athlete with a low aerobic capacity will deplete the glycogen stores relatively fast at moderate to high intensities and thus onset muscular fatigue (Hausswirth & Lehénaff, 2001).

The athlete’s lactate threshold, above which lactate will accumulate, has long been suggested to be one main fatigue factor in sports with moderate to long duration exercise and high intensity. During fairly short, high-intensity (>50-60 % of \(VO_{2\text{max}}\)) exercise, the mitochondria can not oxidize all the pyruvate produced, thus the excess pyruvate will convert into lactic acid. The production of lactic acid in muscle releases hydrogen ions (\(H^+\)) and lactate accumulation will lead to metabolic acidosis and a fall in the intramuscular pH (Begun et al., 2005; Cairns, 2006). Lactate will accumulate as a result of increased exercise intensity, hence the lactic acid production is greater then its elimination (Billat, Sirvent, Koralsztein & Mercier, 2003). By decreasing the intracellular pH, muscle fatigue is caused through a number of mechanisms such as relaxation and reduced tension of the skeletal muscle (Suzuki et al., 2007). A study by Coyle et al (1988 in Bassett & Howley, 2000), investigated the relationship between the lactate threshold and time to fatigue in a group of 14 trained cyclists with similar \(VO_{2\text{max}}\). The subjects were divided into two groups depending on their lactate threshold, high-LT group (mean LT=81,5% \(VO_{2\text{max}}\)) and low-LT group (mean LT=65,8% \(VO_{2\text{max}}\)). Both groups conducted a test at 88% \(VO_{2\text{max}}\) which resulted in significant differences in performance (60,8min vs. 29,1min) and lactate concentration (7,4mmol\(\text{L}^{-1}\) vs. 14,7mmol\(\text{L}^{-1}\)) post exercise for the high-LT and low-LT groups, respectively. More over, the investigators examined the difference in metabolic response between the high-LT and low-LT groups in a 30min cycling test at 79% \(VO_{2\text{max}}\). The results showed that the low-LT group used 69% more glycogen and they reduced their vastus lateralis muscle glycogen concentration 134% more than the high-LT group (Bassett & Howley, 2000). Even in sports where the aerobic system is dominant, e.g. soccer, it is the repeated random short high intensity anaerobic activities that are the main reason for muscular fatigue (Reilly, Bangsbo & Franks, 2000; Bloomfield, Polman, & O'Donoghue, 2007). Long duration endurance exercise also relies heavily on anaerobic energy production since the anaerobic glycolysis significantly contributes with energy production as the intensity increases (Oöpik et al., 2003). However, more recent studies have suggested that \(H^+\) and low pH may only be two of several determinants of fatigue. It has also been proposed that metabolic acidosis is more correctly
caused by an increased reliance on non aerobic ATP turnover and adenosine diphosphate (ADP), inorganic phosphate (Pi) and H⁺ production.

An increase in Pi is thought to inhibit the calcium release from the SR and impair the crossbridge cycling rate which in turn impairs the muscle contraction. Elevated levels of magnesium, as an outcome of reduced pH and ATP, may also reduce the SR calcium release (Nimmo & Ekblom, 2007).

Several studies have suggested that progressive accumulation of potassium (K⁺), due to progressive exercise intensity, may be involved in the process of fatigue by reducing membrane excitability (Nielsen, Kristensen, Hellsten, Bangsbo & Juel, 2002). More recent studies in the same area have investigated the muscle’s disturbances of K⁺, Na⁺ and Cl⁻ on Na⁺ - K⁺ pump activity and the membrane inexcitability as important variables in muscle fatigue (for review see McKenna, Bangsbo & Renaud, 2007).

In conclusion, the process of muscle fatigue is still not fully understood. What is clear is that there are several components involved such as glycogen depletion, decreased pH, high extracellular potassium etc. For coaches and trainers at elite levels it’s essential to have knowledge about this since it will affect the outcome of a training program. The athlete should also have some knowledge about fundamental theories for causes of muscular fatigue to be able to apply them in his or her exercise regime towards improved performance.

1.2 Carnosine and β-alanine

Around the year 1900, Gulewitsch identified an intracellular muscle component named carnosine (Balcombe, Batheja & Manninen, 2007). About 40 years later, the first findings of carnosine’s hydrogen ion buffering capacity and increase in performance were made. It was not until 2003 researchers started to study β-alanine, a component of carnosine, more thoroughly. Well established scientists in the exercise science field have investigated the effects of β-alanine supplementation, among them Dr. Robert C Harris who made the groundbreaking study on creatine in 1992 (Balcombe et al., 2007). As a dietary source, β-alanine is found naturally as carnosine in protein rich foods as fish, beef and chicken. An increased intake of these foods will not have a great effect on the carnosine levels since the carnosine breaks down to its constituents and then re-synthesizes to carnosine, though with a great loss in concentration levels (Balcombe et al., 2007).

Carnosine has been indicated in other areas which could indirectly influence fatigue. Several studies investigating reactive oxygen species and the effect of carnosine as an antioxidant agent show that carnosine might have an effect on the uptake and release of calcium in the SR (Begun et al., 2005). Since a muscle contraction more or less depends on calcium release, it seems as carnosine may have an important part in the process of muscular fatigue even though more studies in the area have to be done.

1.2.1 Carnosine and performance

One of the theories of the cause of muscular fatigue, both in endurance and intermittent sports, is decreased pH levels due to increased concentration of H⁺ ions dissociated from lactic acid in muscle. Supplementation of β-alanine, would increase carnosine levels in muscle and may counteract the decrease in pH. By buffering the H⁺ ions thus potentially delay fatigue (Derave et al., 2007). The body has an H⁺ buffering capacity, which means that the...
body can contain the intracellular pH even though levels of H\(^+\) increase. Carnosine, a naturally occurring histidine and multifunctional dipeptide, synthesized from the amino acids L-histidine and β-alanine, functions as a buffer over the physiological pH range (Derave et al., 2007). Carnosine is found in many tissues, but most significantly in muscle and nerve cells, up to 20mM (Begun et al., 2005). Studies have shown that the availability of β-alanine is the rate-limiting step in intramyocellular synthesis of carnosine, whereas histidine is found in more than sufficient concentrations, relative to the demand for carnosine synthesis in the skeletal muscle. It has been reported that muscle carnosine levels is a crucial factor in both dynamic and isometric exercise performance at high intensity (Hoffman et al., 2008).

The efficiency of β-alanine supplementation on high-intensity exercise performance has been shown in several studies. In a study by Hill et al., (2007, in Derave et al., 2007) untrained males were supplemented with β-alanine over 10 weeks which improved time to exhaustion and total work performed throughout a high-intensity-cycle-test. Stout et al., (2000) reported that 3.2g·d\(^{-1}\) of β-alanine supplementation during a graded physical test in untrained males considerably increased lactate threshold and physical work capacity (Derave et al., 2007). Harris et al., (1997 in Derave et al., 2007) found that an oral intake of 6.0g·d\(^{-1}\) of β-alanine during 4 weeks resulted in increased levels of carnosine content in the vastus lateralis of untrained humans by about 60%, a prolonged supplementation over a total of 10 weeks showed an increase of about 80%. No toxic effects of carnosine or its derivates have been reported (Begun et al., 2005: Derave et al., 2007). Further more, carnosine and its constituents are legal ergogenic aids and are not on the 2009 prohibited list of international standard (WADA, 2009).

Studies have shown that Type II muscle fibers contain twice as high levels of carnosine as Type I fibers, and the proportion of Type II muscle fibers of total muscle fibers is strongly linked to the carnosine concentration (Suzuki et al., 2007). A study by Hoffman et al., (2008) investigated the effect of 4.5g·d\(^{-1}\) of β-alanine supplementation over 30 days on anaerobic performance in collegiate American football players. The subjects were divided in to one β-alanine group and one placebo group. Performance was tested in a 60 s Wingate anaerobic power test, 3 shuttle-runs with rest in between sprints, a training diary and a questionnaire on subjective feelings of fatigue, soreness and practice intensity was carried out. Results showed a lower fatigue rate for the β-alanine group during the Wingate anaerobic power test and a trend in significantly greater volume was reported during the resistance exercise sessions. Furthermore, the ratings of perceived exertion and fatigue were significantly lower for the β-alanine group than the placebo. In addition, several studies have investigated the effects of ingestion of other buffering agents, such as sodium bicarbonate and sodium citrate, on performance with equivocal results (Oöpik et al., 2003).

To my knowledge, no study of the effects of β-alanine supplementation in soccer or distance running have been made. A supplement with β-alanine’s characteristics and potential gains would potentially increase performance and thus be of great interest in these sports.

### 1.3 Evaluation and requirements of sports

#### 1.3.1 Distance running
In distance running, the aerobic system is predominant, the contribution of the anaerobic system depends on the capacity of the runner and the distance of the race (Saunders, Pyne, Telford & Hawley, 2004).

Exercise physiology has a few basic principles. A primary principle is that work requires energy, and that a certain amount of energy needs to be supplied for an athlete to keep a specific work rate or running velocity during a distance run. During low to moderate intensity work, the predominant system to provide ATP to the cross bridges is the aerobic, oxidative phosphorylation system. As the intensity of the work increases the faster the ATP needs to be supplied, and the more the anaerobic system contributes to the synthesis of ATP to maintain cross bridge cycling (Bassett & Howley, 2000). Since the anaerobic system synthesizes ATP via glycolysis, time of duration at this intensity is limited, due to glycogen depletion (Hausswirth & Lehénaff, 2001). Therefore, a prerequisite for superior endurance performance has shown to be the ability to metabolise energy via aerobic processes (Saunders et al., 2004). A common and accepted variable used to determine middle- and long-distance runners’ performance is the maximal oxygen uptake (VO_{2max}) (Saunders et al., 2004). Studies have shown that the VO_{2max} sets the upper limit for endurance performance, but that it is not the greatest predictor of athletic capacity (Bassett & Howley, 2000). The VO_{2max} is an important factor for distance running, however there are other physiological and performance factors that have to be considered (Saunders et al., 2004). Lactate threshold, VO_{2} measured at lactate threshold, running economy and anaerobic capacity are the factors included (Midgley, McNaughton & Wilkinson, 2006). An athlete with a high lactate threshold (LT)/onset of blood lactate accumulation (OBLA) can run at a higher percentage of his VO_{2max} without accumulating lactic acid in comparison with an athlete with a low LT. The ability to breakdown and utilize fat as fuel at higher intensities spares glycogen and will enable the athlete to keep up a high velocity without depleting the glycogen. An established indicator of endurance athletes’ performance is the velocity at LT (Saunders et al., 2004).

![Figure 1. Simplified chart of how VO_{2max}, percentage of VO_{2max} at LT and running economy relate to distance running performance. From Bassett, D.R. JR., and Howley, E.T. (2000).](image-url)
The VO$_{2\text{max}}$ is the maximum rate that an athlete can take up oxygen from the air, transport to and utilize by the cells for aerobic processes during physical activity (Midgley et al., 2006). The VO$_{2\text{max}}$ is affected by a mixture of factors, such as haemoglobin mass, muscle capillary density, stroke volume, muscle fibre composition and aerobic enzyme activity (Saunders et al., 2004). An athlete’s VO$_{2\text{max}}$ is directly associated with the rate of ATP production that the athlete can maintain throughout a distance race. However, the significance of this fact is relative since distance runs are not run at 100% of VO$_{2\text{max}}$. The rate of ATP production depends on the relative VO$_{2}$, i.e. ml·kg$^{-1}$·min$^{-1}$, that can be sustained during the run. This means that the VO$_{2\text{max}}$ is an important factor for energy production in endurance events, but it will not determine the final performance (Bassett & Howley, 2000).

Running economy (RE) is an important factor in distance running. RE is expressed as the submaximal VO$_2$ at a given work rate (Saunders et al., 2004), or as “…the energy required per unit mass to cover a horizontal distance” (Bassett & Howley, 2000, p. 78). A runner with a high RE will be able to run at a higher intensity at a low percentage of the runner’s VO$_{2\text{max}}$ in comparison with an athlete with a lower RE who will have a higher submaximal VO$_2$ at the same velocity even though the two runners have similar VO$_{2\text{max}}$. Studies have shown that elite runners have a better RE than untrained or less trained runners (Hausswirth & Lehénaff, 2001). Some physiological factors affecting the RE are core temperature, ventilation, muscle fibre type, heart rate and lactate levels. When the core temperature increases, so does the sweating, therefore the metabolic cost increases due to raised blood circulation and increases ventilation, this in turn will result in increased submaximal VO$_2$ and a decreased RE. Biomechanical factors, such as height, pelvis- and foot size, stride length, leg morphology and other kinematics are also important for RE. It has been suggested that anthropometric measures such as body mass, percentage body fat and limb dimensions will influence the RE (Saunders et al., 2004). In addition, a recent study in well trained triathletes showed that heavy weight training in combination with endurance training enhanced the RE and improved the running performance (Bassett & Howley, 2000). The study was over a period of 14 weeks, one group trained heavy weight training and endurance while the control group only did endurance training. The endurance/strength group showed an 11% lower submaximal VO$_2$ in comparison to the endurance-only group after the 14 weeks as well as the endurance/strength group had improved their RE compared to the pre-tests (Bassett & Howley, 2000). The percentage of VO$_{2\text{max}}$ that can be sustained during prolonged exercise is as mentioned an important variable for endurance performance. Study result shows that trained subjects could work at 87% and 83% of their VO$_{2\text{max}}$ for 1 and 2 hours respectively, in comparison with untrained subjects who only could work at 50% and 35% for the same time (Bassett & Howley, 2000) (see simplified chart, fig. 1).

Several studies have shown that the lactate threshold is a good predictor of performance in endurance sports such as running and cycling as for both trained and untrained subjects. A classical model proposes that “…the ability to maintain a high running speed is linked to the ability to maintain a high rate of oxidative ATP production” (Bassett & Howley, 2000, p. 80). The most valid way to determine a runner’s lactate threshold is by standardized lactate testing.

**Lactate test**

“Quite simply, true lactate testing is the gold standard and ultimate form of testing available” (Mauro, 2006, p. 1). A lactate test measures both the aerobic and anaerobic capacity of the athlete. By progressive increases in intensity and continuous blood sampling, the lactate concentration in the blood is measured and represents the energy systems. The blood samples clearly indicate if the athlete is working aerobic or anaerobic. The purpose of the test is to find
the anaerobic threshold and thereby determine the aerobic and anaerobic capacity of the athlete (Caputo, Stella, de Mello & Denadai, 2003).

1.3.2 Soccer

Energy systems
Soccer integrates periods of high-intensity activity combined with periods of lower-intensity activity. A soccer player needs to be physiologically competent in a number of aspects of fitness such as agility, muscle strength, flexibility, aerobic and anaerobic capacity etc. (Svensson et al., 2005). According to Reilly et al. (2000), the distribution of energy systems in elite soccer players is as following:

<table>
<thead>
<tr>
<th>Predominant energy systems used in soccer</th>
<th>Anaerobic</th>
<th>Aerobic</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATP-PCr</td>
<td>2 %</td>
<td>O2</td>
</tr>
<tr>
<td>Glycolytic</td>
<td>18 %</td>
<td>80 %</td>
</tr>
</tbody>
</table>

The predominant metabolic pathway during soccer is aerobic. Most of the activity during a game is without the ball, deceiving opponents, creating space to play the ball for teammates or following runs by opposition players (Reilly et al., 2000). A Danish study showed that a male elite soccer player runs an average of 10.8 km (between 10.1 and 11.4 km) per game, the study also showed that the soccer player runs about 5% longer during the first half of the game compared to the second half (Bangsbo et al., 1991).

Approximately 80-90% of total time during competitive play is spent in moderate and low intensity activities while 10-20% is high intensity activities. Though the aerobic system is the predominant during an average game it is the repeated random short high intensity anaerobic activities that’s producing and increasing the levels of lactic acid and thereby the accumulation of H+ ions which is the major factor influencing fatigue. The activity distribution has been reported to be 19.5% standing, walking 41.8%, jogging 16.7%, running 16.8%, sprinting 1.4%, and other 3.7%. A session of high-intensity activity was observed to occur every 30 seconds and all out sprinting every 90 seconds during match play (Reilly et al., 2000 & Bloomfield et al., 2007).

Several studies have determined the maximum oxygen uptake for elite male soccer players to be 56.0-69.0 ml·kg⁻¹·min⁻¹ (Reilly et al., 2000) and 38.6-57.6 ml·kg⁻¹·min⁻¹ for female soccer players (Stolen et al., 2005). Midfielders and full-backs had the highest values. The average energy expenditure during a match for a male with a oxygen uptake of ml·kg⁻¹·min⁻¹ and weighing 75 kg has been estimated to about 5700 kJ (1357 kcal) (Reilly et al., 2000). A player who can sustain a high work-rate throughout a whole game has a big advantage comparing to those whose energy, in the form of glycogen stores, will be close to depleted towards the end of the game (Svensson et al., 2005).

The aerobic base is also very important for the player in recovery purpose. There are a lot of multiple sprints during match play and the player needs to be able to recover rapidly in between sprints. Opposed to single sprints, physical demands in multiple sprints are higher, as phosphocreatine and ATP must be resynthesized during recovery. The better aerobic capacity and higher anaerobic threshold, the faster and better the aerobic glycolysis can resynthesize
phosphocreatine stores and remove blood lactate from the muscle. A low ability to eliminate the lactate will reduce the phosphocreatine levels in the following sprints and result in poor resynthesis of ATP (Svensson et al., 2005).

The Yo-Yo intermittent recovery test
The Yo-Yo intermittent recovery test and Yo-Yo intermittent endurance test were designed for testing soccer players’ physical capacity in a way that is similar to match play (Krustrup et al., 2003). Krstrup et al. (2003) established the validity and reliability of the Yo-Yo intermittent recovery test. They found significant correlations between the amount of high intensity exercise performed during soccer match-play and performance on the Yo-Yo intermittent recovery test. The test simulates both aerobic and anaerobic glycolysis, since the blood lactate concentration and the heart rates of the players were elevated at the end of the test. Furthermore, a high level of phosphocreatine was also broken down at the end of the test which confirmed the use of the ATP-PCr system. The distribution and use of energy systems recorded in the test were very similar to those that support match-play. It was determined that the Yo-Yo intermittent recovery test provides a more accurate and valid indication of soccer specific activity patterns and aerobic fitness during soccer match-play than for example direct assessment or field predictions of VO$_{2\text{max}}$. A high score in the Yo-Yo intermittent recovery test has been shown to be linked to a high physiological capacity in soccer (Krustrup et al., 2003).
2 Purpose and Objective
The purpose of the present study was to examine the effects of 8 weeks supplementation of β-alanine in distance runners and Swedish division four soccer players on aerobic capacity, intermittent recovery and muscular fatigue. The runners were tested in lactate profiling tests and the soccer players in the Yo-Yo intermittent endurance test. A secondary objective of this study was to get preliminary results which can be used to potentially improve performance in sports.

2.1 Aim
Specific aim 1: Determine if β-alanine supplementation decreases the lactate acid production and in turn delays the onset of muscular fatigue. 
Hypothesis 1: After 8 weeks of β-alanine supplementation, I believe that the subjects will be able to work at the same intensity as pre test period but with a lower level of lactate acid concentration in the blood.

Specific aim 2: Assess if β-alanine supplementation results in a faster recovery between shuttles during the Yo-Yo intermittent endurance test in soccer players and a lower level of blood lactate acid concentration at post recovery lactate test in distance runners due to faster blood lactate acid removal.
Hypothesis 2: After 8 weeks of β-alanine supplementation, I believe that the soccer players’ β-group will increase the number of shuttles in the Yo-Yo intermittent endurance test more than the control group due to increased blood lactate acid removal between the shuttles, and that the distance runners’ β-group will have a lower level of blood lactate acid accumulation than the control group at the recovery blood sample due to a decreased lactate accumulation during the test.
3 Materials and methods

3.1 Information retrieval
In the search of literature and articles, the databases Science Direct, Sport Discus, PubMed and Google Scholar were used. Searching word used were e.g. physical demands, lactate threshold, muscular fatigue, potassium accumulation, lactate test protocol, yo-yo intermittent endurance test etc. I also used Halmstad University’s library in the search for literature.

3.2 Subjects
I recruited the subjects via the coach of the soccer team and the leader of the runners.

The initial subject pool \( n = 37 \) included 36 males and 1 female. The subject pool consisted of 22 male soccer players (group S, mean age 22 years; age range 18-29 years), playing in a Swedish division four team, and 1 female and 15 male long distance runners (group DR mean age 30 years; age range 17-43 years), all at a competitive level. Subjects signed an informed consent form (see appendix 2) approved by the Ethical Review Board at the SET at Halmstad University for the use of human subjects. The subjects were informed about the study in writing (see appendix 1) and by a presentation held by the author. The two groups were randomly divided into two additional groups, a β-alanine group and a control group, respectively. The S-group consisted of the β-alanine group \( n = 8 \), mean age 20 years; age range 18-29 years) and the control group \( n = 14 \), mean age 24 years; age range 18-29 years), and the DR-group consisted of the β-alanine group \( n = 8 \), mean age 35 years; age range 27-42 years) and the control group \( n = 7 \), mean age 26 years; age range 17-43 years).

Drop out from post-testing occurred in both groups in the study. Of 22 subjects in the S-group (pre-test; β-group \( n = 8 \) and control-group \( n = 14 \)), 12 subjects (post-test; β-group \( n = 6 \) and control-group \( n = 6 \)) accomplished the post-test and of the initial 15 subjects in the DR-group (pre-test; β-group \( n = 8 \) and control-group \( n = 7 \)) 12 subjects (post-test; β-group \( n = 6 \) and control-group \( n = 6 \)) accomplished the post-test. The S-group consisted of, at post test, β-group \( n = 6 \) mean age 21 years; age range 18-29 years) and the control group \( n = 6 \), mean age 22 years; age range 18-28 years and the DR-group consisted of, at post-test, β-alanine group \( n = 6 \) mean age 35 years; age range 29-43 years) and the control group \( n = 6 \), mean age 33 years; age range 19-43 years).

3.3 Experimental protocol
The β-alanine groups were supplemented with 5g·d\(^{-1}\) of β-alanine during eight weeks, whilst the control groups did not take any supplements. All subjects participated in two test sessions, pre and post the test period (at week 0 and week 8). The S-group was tested in the Yo-Yo intermittent endurance test and the DR-group was tested on a lactate test on treadmill. All the subjects trained as usual during the test period. To avoid source of errors, all subjects were instructed to keep training diary, some exercise sessions were monitored and a dialog was held with coaches and leaders throughout the test period. The training diary was to include variables such as pre training meal, exercise volume and intensity, feelings of muscle soreness and fatigue and the perceived quality and exertion of the session using the Borg CR10 Scale (Borg, 1998).

3.3.1 The Yo-Yo intermittent endurance test protocol (see appendix 4)
The test was conducted in the sport stadium at Halmstad University. Subjects were advised to come to the tests rested, hydrated, not to eat within 1 hour prior the test and to avoid strenuous exercise in the 48 hours preceding the test session (Denadai, Figuera, Favaro & Gonçalves, 2004). The test was conducted according to standard protocol, level two of the test was chosen considering the level of the athletes (Bangsbo, 2005). Prior the test, the subjects were asked to warm up for five minutes and further run the course at an optional speed for three minutes. The test consisted of repeated 20m shuttle runs interspersed with short, 2.5m x 2, recovery. The subjects jogged during the recovery section and had to come to a complete stop before the next shuttle. The time allowed for a shuttle was progressively decreased dictated by audio bleeps from a CD-player. The aim of the test was to complete as many shuttles as possible. The bouts were run until the subject could not keep up the present pace for 2 bouts. The first time the subject did not make the bout in time a warning was given, he then continued until he failed to keep up the pace a second time. When the subject failed to continue, final velocity, number of the final 2x20m interval and the total distance covered were recorded.

![Figure 2. The Yo-Yo intermittent endurance test's course.](image)

### 3.3.2 Lactate testing protocol (see appendix 3)
The test was conducted in the bio lab at Halmstad University. Subjects were advised to come to the tests rested, hydrated, not to eat within 1 hour prior the test and to avoid strenuous exercise in the 48 hours preceding the test session (Denadai, Figuera, Favaro & Gonçalves, 2004). The test was conducted according to standard protocol (Caputo, Stella, de Mello & Denadai, 2003). The test was performed on a treadmill (NordicTrack, 9800 Incline Trainer), blood samples was taken in the finger capillary and analyzed in a lactate analysis machine (Analox instruments Ltd). The subject was seated for 5 minutes to measure the “resting” heart rate, using a heart rate monitor (Garmin Forerunner 305), prior the test. No warm up jog was allowed, only stretching and walking. The treadmill was at a constant 1.5% grade to simulate over ground running. The test was performed over a number of stages depending on the lactate levels of the subject (range of number of stages = 4-8 stages). Stage 1 was 10km/h, 12km/h or 14km/h depending on the present fitness of the athlete. Each stage lasted 4 minutes, hence the subject reached steady state, and the speed increased with +1km/h at each new stage. The athlete straddled the treadmill belt when 30 s remained of each stage, blood sample was taken and lactate concentration was analyzed. Heart rate and RPE was also recorded. To finish the test, the athlete had to continue until 2 consecutive stages with >1mmol∙L⁻¹ increase and lactate concentration had to be > 4 mmol∙L⁻¹. A 10 minute warm down at 4-6 km/h was conducted and recovery blood lactate and heart rate were recorded.

### 3.3.3 Equipment

*The Yo-yo intermittent endurance test*
- Measuring tape, 50m
- Cones
- Stereo and the yo-yo test on audio CD
• Protocol

*Lactate test*
• Treadmill (NordicTrack, 9800 Incline Trainer)
• Lactate machine (GM Series Analyser, Analox Instruments Ltd)
• Lactate reagent buffer lysing solution (Analox Instruments Ltd)
• Other equipment such as pipettes, lancettes, sterile gloves etc.
• Heart rate monitor (Garmin forerunner 305)
• Protocol

3.4 Statistical analysis
In this study, Microsoft Excel 2007 was used for statistical analysis. All data are expressed as mean ± SD. Differences between pre- and post-tests were examined using a two-tailed student’s t-test for paired samples. Statistical significance level was set at \( p < 0.05 \) and statistical trend level was set at \( p < 0.15 \).
4 Results

4.1 The Yo-Yo intermittent endurance test
For the soccer players, the 8-week test period resulted in significant distance increases for both groups \((p = <0.05)\). However, it did not result in significant difference between the β-group and the control-group \((p = 0.29)\). Even though there is no significant difference or a trend, the β-group had a higher total increase in meters run between their pre- and post-test in comparison to the control-group, see figure 3. The mean distance increase for the β-group was 953 ± 396m and for the control-group 726m ± 337m. Significant changes was evident between pre- and post-tests tests but not between groups, see figure 3.

![Figure 3. Pre- and post-results from the Yo-yo intermittent endurance test. The β-group ran a mean distance of 1020 ± 390m and 2033 ± 670m at the pre- and post-test respectively. The control-group ran a mean distance of 1493 ± 657m and 2220 ± 743m at the pre- and post-test respectively.](image)

4.2 Lactate test

4.2.1 Velocity at lactate threshold (\(V_{LT}\))
There was no significant change after the 8-week test period in \(V_{LT}\) in the two groups \((p = 0.75)\). However, there was a trend in difference between the groups \((p = 0.11)\). The mean \(V_{LT}\) for the first and second test (figure 4) increased slightly from 16.8 ± 1.6 km/h to 17.0 ± 1.2 km/h in the β-group and decreased from 17.3 ± 1.0 km/h to 16.8 ± 1.5 km/h for the control-group.

![Figure 4. Mean results from of \(V_{LT}\) from pre- and post-lactate test in control-group and β-group. The vertical axis represents velocity in km/h.](image)
4.2.2 Heart rate at lactate threshold (HR<sub>LT</sub>)

HR<sub>LT</sub> did not change significantly during test period in the two groups (\(p = 0.82\)) nor between the groups (\(p = 0.67\)). As figure 5 shows, mean HR<sub>LT</sub> for the first and second test was 179.6 ± 5.0 b·min<sup>-1</sup> and 179.6 ± 5.0 b·min<sup>-1</sup> for the β-group and 173.2 ± 7.6 b·min<sup>-1</sup> and 172.0 ± 4.7 b·min<sup>-1</sup> for the control-group. There was no significant change in HR<sub>LT</sub> between tests or between groups.

![Figure 5](image)

**Figure 5.** Mean results of HR<sub>LT</sub> from pre- and post-lactate test in the control-group and β-group. The vertical axis represents heart rate in b·min<sup>-1</sup>.

4.2.3 Blood lactate at lactate threshold (BL<sub>LT</sub>)

No significant change in BL<sub>LT</sub> was seen in the two groups (\(p = 0.32\)) nor between the groups (\(p = 0.69\)). The mean BL<sub>LT</sub> for the first and second test was 8.9 ± 1.8 mmol·L<sup>-1</sup> and 7.8 ± 1.8 mmol·L<sup>-1</sup> for the β-group and 8.6 ± 2.7 mmol·L<sup>-1</sup> and 7.9 ± 2.5 mmol·L<sup>-1</sup> for the control-group. Figures 6A and B shows the pre- and post-results of the individual subjects in both groups. There was no significant change in BL<sub>LT</sub> between tests or between groups.

![Figure 6](image)

**Figure 6.** Results of BL<sub>LT</sub> from pre- and post-tests in subjects of the control-group (A) and the β-group (B), respectively. The vertical axis represents blood lactate in mmol·L<sup>-1</sup>.

4.2.4 RPE at lactate threshold (RPE<sub>LT</sub>)

RPE<sub>LT</sub> did not change significantly between the first and second test in the two groups (\(p = 0.18\)) nor between the groups (\(p = 0.20\)). The mean RPE<sub>LT</sub> for the first and second test was 8.7 ± 1.8 and 8.5 ± 1.8 for the β-group and 8.6 ± 0.7 and 7.9 ± 5.4 for the control-group. No significant change in RPE<sub>LT</sub> was evident between tests or between groups.
4.2.5 Recovery blood lactate (RBL)

RBL did not change significantly between the first and second test in the control group \((p = 0.76)\). In contrast, a trend in difference between the first and second test in the \(\beta\)-group was noticed \((p = 0.11)\). Furthermore, there was a trend in difference between the groups \((p = 0.14)\). Figures 7A and B shows the results of the individual subjects in both groups at pre- and post-test. The mean RBL for the first and second test was 4.5 \(\pm\) 1.6 mmol\(\cdot\)L\(^{-1}\) and 3.1 \(\pm\) 1.0 mmol\(\cdot\)L\(^{-1}\) for the \(\beta\)-group and 4.6 \(\pm\) 1.6 and 5.0 \(\pm\) 3.0 for the control-group. No significant change in RBL was evident between tests or between groups, nevertheless a trend could be detected in decreased RBL in \(\beta\)-group at the post test.

\[
\begin{array}{c|c|c|c|c|c|c|c|c|c|c|c}
0 & 2 & 4 & 6 & 8 & 10 & 12 \\
\hline
\end{array}
\]

\[
\begin{array}{c|c|c|c|c|c|c|c|c|c|c|c}
C1 & C2 & C3 & C4 & C5 & C6 \\
\hline
\end{array}
\]

\[
\begin{array}{c|c|c|c|c|c|c|c|c|c|c|c}
B1 & B2 & B3 & B4 & B5 & B6 \\
\hline
\end{array}
\]

\[\text{Figure 7. Results of RBL from pre- and post-tests in subjects of the control-group (A) and the \(\beta\)-group (B), respectively. The vertical axis represents blood lactate in mmol \cdot L^{-1}.}\]

4.3 Training diary

According to interviews and training diary of subjects, a majority of the subjects in the distance runner’s \(\beta\)-group perceived an increase in performance during the test period. Most significant was the effect during high intensity interval training where some of them noticed that heart rate recovered twice as fast between intervals in comparison to pre-test period and that the feeling of “lactate removal” seemed faster between intervals. Subjects also felt that they could run at very high intensities without accumulating as much lactic acid. Furthermore, two of the six \(\beta\)-group subjects lowered their sub maximal heart rate (at a given work load) with 5-10 bpm during the test period which meant they could run at higher velocities with a lower heart rate and hence increasing their LT. However, since this was not consistent in all the \(\beta\) -group the difference in LT did not reach statistical significance.
5 Discussion

5.1 Discussion of results
The major finding of this study was a trend in decreased recovery blood lactate and increased \( V_{LT} \) in the distance runners-group after 8-week of \( \beta \)-alanine supplementation. In addition, \( \beta \)-alanine supplementation did not seem to have any effect in soccer players as evaluated by the yo-yo test. Important to notice is that, this was the first study to my knowledge to investigate the effects of \( \beta \)-alanine in distance runners and soccer players.

The aims of this study were (1) to determine if \( \beta \)-alanine supplementation decreases the lactate acid accumulation and therefore increase the LT and in turn delay the onset of muscular fatigue and (2) to assess if \( \beta \)-alanine supplementation results in a faster recovery between shuttles during the Yo-Yo intermittent endurance test in soccer players and a lower level of blood lactate acid concentration at post recovery lactate test in distance runners due to increased removal of blood lactate acid. These aims were assessed on a Swedish division four soccer team and distance runners at elite levels. The results from this study differed slightly from previous studies (Hill et al., 2007 & Hoffman et al., 2008) possibly due to a different study design where variables such as different tests, training status of the subjects, training season, illness, drop outs and mismanagement of beta alanine intake could play a role.

5.1.1 The Yo-Yo intermittent endurance test
In the soccer players, there was no significant difference or trend between the \( \beta \)-group and the control-group’s results \((p\text{-value} = 0.29)\) from the yo-yo intermittent endurance test. The \( \beta \)-group improved their results more than the control-group, although not statistically significant. This improvement could be an indication that a significant significance would have been found with a larger subject pool. An alternative theory for the \( \beta \)-group’s improvement is that they performed poorly during pre-test and therefore would be expected to do better in absolute terms during post-test. The pre-test was carried out during early pre-season. For some of the soccer players the test session was the first practice since last season, hence the fitness status varied between players. Only six of the 22 subjects had done maintenance training during off-season. The low fitness status of the players during pre-season may be a reflection of the division (4) they are in. The post-test was conducted 8 weeks after pre-test in competition season, which meant that the fitness-levels of the subjects were higher due to 8 weeks of consistent training. Therefore, improved results at post-test were expected for both \( \beta \)-group and control-group.

Furthermore, two of the subjects in the \( \beta \)-group did not follow through the \( \beta \)-alanine intake according to protocol. The reasons for inconsistent \( \beta \)-alanine intake were vacation and illness. In comparison to the runners, the soccer players seemed less motivated in testing the supplement. This may be since they are not as serious with their sport, that soccer is a team sport, a lower mean age of the subjects and that the soccer players were not as knowledgeable about their physiological capacities as the individual runners were since the soccer players may focus more on the game of soccer itself. Anyhow, the choice of a division four soccer team was a conscious choice. There would have been a greater risk that the training protocol would have affected the results of the yo-yo test more in a team that plays in a higher division since they are more likely to periodize their training to an even greater extent. It would be optimal to test soccer players during competition season when their training intensity and volume are similar during a longer period of time.
In addition to two people being inconsistent with their β-alanine intake, drop out from post-testing occurred in the study and may have affected the result. Of 22 subjects (pre-test; β-group n= 8 and control-group n= 14), 12 subjects (post-test; β-group n= 6 and control-group n= 6) accomplished the 2 tests.

5.1.2 Lactate test

There were no significant differences between the two distance running groups in HR\textsubscript{LT}, BL\textsubscript{LT} or RPE\textsubscript{LT}. However, a trend in difference was seen between groups in both V\textsubscript{LT} (p = 0.11) and RBL (p = 0.14) where the β-group had increased slightly from 16.8 ± 1.6 km/h to 17.0 ± 1.2 km/h in V\textsubscript{LT} and decreased from 4.5 ± 1.6 mmol·L\textsuperscript{-1} to 3.1 ± 1.0 mmol·L\textsuperscript{-1} in RBL. In contrast to these results, most studies that show significant improvements from β-alanine supplementation however, other studies have generally examined the effect of the supplement in high intensity exercise (Hill et al., 2007; Suzuki et al., 2007). Furthermore, HR\textsubscript{LT}, BL\textsubscript{LT} and RPE\textsubscript{LT} do not point out any representative results as single values since they are dependent on at least 1 other variable. On the other hand an increase in-, V\textsubscript{LT} and decrease in RBL do point out representative results as single variables since they are not dependent or in direct relationship to other variables than the LT. The V\textsubscript{LT} is a major variable for performance in distance running, an increase in V\textsubscript{LT} will potentially improve performance since min·km\textsuperscript{-1} will be reduced. Efficiency of lactate transportation from muscle is of vital importance in both running and soccer. A low RBL is an indication of efficient lactate removal.

The variation in fitness status from off-season to competitive-season is not as high in runners as in soccer players, since many runners train all year around. Therefore, a smaller increase in performance was expected in the runners at the post-test in comparison to the soccer players. However, runners tend to increase the intensity of their training towards competitive-season and decrease their volume training which results in higher performance. Since the runners trained somewhat similar, according to training diaries, during the test period, training protocol was not expected to affect results between β-group and control-group. In contrast to the results of Hill et al., (2007) this study did not show significant improvements from β-alanine supplementation which might in part be due to that they used untrained subjects.

The results from the post-test may not be representative since two subjects in the β-group had just come back from illness and still felt a bit tired, and 1 other subject in the β-group ran a 10km competition 1 day prior to the test which may have affected the result and been a reason that the result did not show a significant difference in LT. In addition 1 subject in the control-group had also been ill a couple of days earlier as well as 1 subject ran a 4km competition the day prior the test.

The drop out ratio was smaller in the runners than soccer players, however three subjects dropped out. 12 subjects (post-test; β-group n= 6 and control-group n= 6) of 15 (pre-test; β-group n= 8 and control-group n= 7) accomplished the post test.

Supplementation of β-alanine could potentially increase the buffering of H\textsuperscript{+} and thus increase the removal of blood lactate acid. The fact that there was no significant difference (p-value = 0.69) between groups in BL\textsubscript{LT} could have been that LT was reached at lower velocities and heart rates in two of six subjects in the control-group and therefore they did not produce as much lactate acid rather than having an effective removal. Even though no significant difference was shown in BL\textsubscript{LT}, a trend (p = 0.14) in lower RBL was evident. One could speculate in that the control-group had performed better at the post-test, the mean BL\textsubscript{LT} may
had been increased and thus the RBL would potentially been higher and the statistics in RBL could have reached significant difference.

5.1.3 Mechanisms of muscular fatigue

There are several potential mechanisms for muscular fatigue, such as depletion of glycogen, oxidative stress, disruption of contractile mechanisms, decrease in pH, increased levels of inorganic phosphate and accumulation of metabolites as lactate, hydrogen and potassium (Nimmo & Ekblom, 2007; McKenna, Bangsbo & Renaud, 2007; Oöpik et al., 2003; Reilly, Bangsbo & Franks, 2000; Bloomfield, Polman, & O'Donoghue, 2007; Begun, Cuncliffe & Leveritt, 2005). In this study only one possible cause was investigated indirectly. One of the major theories of the cause of muscular fatigue, both in endurance and intermittent sports, is decreased pH levels due to increased concentration of H⁺ ions dissociated from lactic acid in muscle. Derave et al., (2007) suggested that supplementation of β-alanine, thus increase carnosine levels in muscle and may counteract the decrease in pH and by buffering the H⁺ ions potentially delay fatigue. Since there was a trend in the DR-group in decreased RBL ($p = 0.14$) and increased $V_{LT}$ ($p = 0.11$), this may potentially represent the H⁺ buffering effect of the β-alanine. In addition, it seems as supplementation of β-alanine increases the elimination of blood lactate, hence low RBL, which is of major importance in all sports where the anaerobic system is used.

5.2 Discussion of methods and materials

5.2.1 The Yo-Yo intermittent endurance test

Since the Yo-Yo intermittent recovery test was not accessible, I chose to test the soccer players in the Yo-Yo intermittent endurance test. The two tests are similar and both adjusted for soccer with differences in velocity and length of the recovery jog (Reilly, Bangsbo & Franks, 2000).

The soccer players were instructed to keep a training diary, however they did not. General training session information from the subjects was verbal recollected. The results from the Yo-Yo intermittent endurance test showed that the β-group did improved their results more than the control-group. It would have been of interest to analyze the training diaries since significant differences in training levels between the two groups may have been found. It may be that the subjects found it burdensome to keep the diary and that a pre made training diary form would have been a wiser choice.

The results from the soccer players may not be as representative depending on the facts that all of the subjects in the β-group did not follow the supplement intake protocol due to some possible causes, with lack of motivation being the major possible cause. The level of the players is also a problem.

5.2.2 Lactate test

The subjects in the DR-group kept detailed training diaries which made it possible to exclude changes in training protocol as a major variable for the results.
In this study a lactate machine was used instead of portable lactate measurement equipment. This procedure is slightly more advanced and cumbersome, but the reliability is higher in the lactate machine which made it worth the effort.

To estimate the perceived exertion of the subjects, the Borg CR10 Scale was used without complication. However, the Borg RPE Scale may have been more appropriate since it’s created for use in physical tests.

To analyze the results from the lactate test in the DR-group more thoroughly, it would have been necessary to have conducted maximal tests, such as a VO$_{2\text{max}}$-test. With values from a maximal test, the potential maximal capacity of the subjects would have been known, hence the percentage of the subject’s max could have been analyzed in relation to the variables. Coyle et al., (1988 in Bassett & Howley, 2000) used values from VO$_{2\text{max}}$-tests when they investigated the relationship between the LT, as a percentage of VO$_{2\text{max}}$, and time to fatigue in cyclists. Even though this would have favored the study, valid equipment was not available.
6 Conclusion
A trend toward lower levels of RBL in and increased $V_{LT}$ after eight weeks of β-alanine supplementation was found in distance runners whereas no differences between soccer player group could be detected, however, no statistically significant ($p < 0.05$) differences were found. Due to not optimal circumstances, the results can not be taken as definite. A larger study with increased sample size may have reached significant difference. Considering the trend found in the study with lower RBL for the runners’ β-group part of the results indicates faster removal of lactate acid which is of major importance in many sports. These results suggested that β-alanine may delay the onset of fatigue and improve performance in endurance sports such as running by increasing the removal of lactate from muscle.

7 Acknowledgements
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References


http://www.trainingsmartonline.com/images-Free_Triathlon_Articles/Lactate_Testing_Triathlon.pdf
Appendix 1

Information to subjects

Informationsbrev och förfrågan om medverkan i en labbstudie till triatleter, löpare och fotbollsspelare.


Syftet med den planerade studien är att undersöka effekten av 8 veckors intag av aminosyran beta (β) alanin på aerob och anaerob kapacitet. Min fråga är därför om du/ni vill delta i denna studie.

För att skapa förståelse och intresse kring studien följer en genomgående presentation och bakgrund:

Presentation och bakgrund


Studier visar att testpersoner som intagit β-alanin ökade tiden till utmattning samt den totala utförd arbetsmängden under ett intensitetscykeltest signifikant. Andra studier har visat att β-alanintillskott minskar den upplevda tröttheten under arbete, testpersoner orkar med längre träningspass, högre volym och intensitet mm. i jämförelse med placebo- och kontrollgrupper.

**Tillvägagångsätt**

Deltagandet i studien innebär att laktat- och/eller YoYo-test kommer att genomföras på dig/er (protokoll följer nedan). Laktattest beräknas ta omkring 30 minuter, likaså beräknas YoYo-testet ta ca 30 minuter. Du ombedes även att under testperioden, 8 v, föra träningstestbok med följande variabler; Typ av träningsspass, volym, intensitet, upplevd trötthet (RPE-skala 1-10), muskelkondition (skala 1-5), sömn (god/dålig), mat före passet, allmänt välbefinnande på träningssvårighetsgrader samt kvalitet på passet (skala 1-5). Du för träningstestbok för att jag sedan ska kunna utesluta att träningen kan ha påverkat testresultaten. All personlig information kommer att behandlas konfidentiellt vilket betyder att protokoll och rapport kommer att avidentifieras och behandlas i enlighet med bestämmelser i Sekretesslagen. Din medverkan är frivillig och kan när som helst avbrytas. Studien är granskad och godkänd av den lokala etikgranskningsgruppen vid Högskolan i Halmstad och genomförs som en del av min kandidatexamen i Biomedicin inr. fysisk träning vid Högskolan i Halmstad.

**Protokoll och genomförande**

Du kommer som testperson få en burk à 300g pulver β-alanin. Du kommer att bli ombedd att inta 5g/1 skopa av β-alanin per dag under 8 veckor. En servering (1 skopa) intages 45 min före träning. På icke träningssvårigheter intar du 1 servering i samband med måltid, exempelvis frukost eller middag. Som deltagare i kontrollgruppen kommer du ej att inta något preparat, utan enbart utföra testerna. Testernas resultat kommer att skriva 0 och 8 under testperioden. Intaget av preparatet sker först efter första testomgången.


Laktattestet kommer att ske på löpband enligt följande protokoll:

- Du som testperson ombedes att; komma till testet utvilad, äta en bra kost tidigare under dagen och ej närmare än 1 timma innan testet, vara välhydrerad och ej ha tränt hårt under de senaste 48 timmarna.

**Utrustning**

- Löpband
- Laktatmätarinstrument inkl nödvändiga vätskor, pipetter, lancetteter, steril handskar mm.
- Pulsklocka
- Tidtagarur

**Procedur**
• Testpersonen lägger sig ner i 5 minuter, vilopuls och laktatnivå i blodet protokollförs
• Stretching och gång är tillåtet innan testet påbörjas, ej någon uppvärmningsjogg
• Konstant 1,5% lutning för att simulera utomhuslöpning
• Steg 1, hastigheten sätts på 10km/h och hålls i 4 minuter eller tills testpersonen hittar steady state
• Efter steg 1 är varje steg 4 minuter och vid varje nytt steg ökas hastigheten med +1km/h
• Testpersonen gränslar löpbandet medan blodprov tas, ca 30sek
• För att testet ska kunna slutföras måste 2 på varandra följande steg öka med >1mmol och laktatkoncentration måste vara >4mmol
• Ett sjätte steg, steg 6 kan läggas till om det ej har blivit en signifikant ökning av laktatnivån och testpersonen ej upplevde steg 5 som väldigt jobbigt. Testet kan likaså avslutas efter steg 4 om testpersonen upplever det som väldigt jobbigt och en signifikant ökning av laktatnivån har observerats, vanligen >4mmol.

Blodproven kommer att tas i fingerspetsarna på testpersonen, det kommer att ske ca 6 blodprovstagnningar å 7µl. Innan varje blodprov kommer fingerspetsen torkas av med desinfektionsmedel samt vatten. Testledare som utför blodproven kommer att ha på sig plasthandskar.

Yo-Yo Intermittent Recovery Test kommer att ske i idrottshall och enligt följande protokoll:
• Du som testperson ombedes att; komma till testet utvilad, äta en bra kost tidigare under dagen och ej närmare än 1 timma inpå testet, vara välhydrerad och ej ha tränat hårt under de senaste 48 timmarna.

**Procedur**

YoYo-testet består av upprepade löprundor fram och tillbaka mellan start-, vänd- och mållinje med en progresivt ökande hastighet kontrollerad av pipsignaler från en CDspelare. Mellan varje runda har testpersonen en 10 sekunders viloperiod, bestående av 2x5 m jogging. Då testpersonen ej klarar av att nå mållinjen två gånger i följd protokollförs den avklarade distansen, vilken representerar testresultatet. Testet kommer att utföras i grupp.

**Kontakt**

Om du accepterar att medverka i studien kommer du att kontaktas per telefon eller e-post för att komma överens om en tid för test.

Med vänlig hälsning

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Appendix 2

Written consent

Skriftligt, informerat samtycke till medverkan i labbstudien gällande, effekten av 8 veckors intag av β-alanin på aerob och anaerob kapacitet.

Jag har informerats om studiens syfte, om hur informationen samlas in, bearbetas och handhas. Jag har även informerats om att mitt deltagande är frivilligt och att jag, när jag vill, kan avbryta min medverkan i studien utan att ange orsak. Om jag skulle känna av någon form av allergisk reaktion från preparatet kontaktar jag Johan Arnerlind omgående. Jag samtycker härmed till att medverka i denna labbstudie som handlar om vad 8 veckors intag av β-alanin kan ge för effekt på aerob och anaerob kapacitet.

Ort/Datum/År

________________________
Namnunderskrift

________________________
Namnförtydligande

________________________
Studentens underskrift

________________________
Namnförtydligande

________________________
Handledarens underskrift

________________________
Namnförtydligande
Appendix 3

Lactate test protocol

Name:

Time and date:

Tester:

Starts step 1 at: 10km/h 12km/h 14km/h

<table>
<thead>
<tr>
<th>Steps</th>
<th>Time (Min)</th>
<th>HR (BPM)</th>
<th>Lactate (Mmol/l)</th>
<th>RPE (1-10)</th>
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<tr>
<td>Recovery</td>
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General comments:
## Appendix 4

**Yo-Yo Intermittent Endurance test protocol**

**Resultatschema:**
**Yo-Yo Intermittent uthållighetstest**
**Testtillfälle #**

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Datum:  
Nivå:  
Underlag:  
Testledare: