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Relationship Between Amino Acids and Performance Measures of Elite Soccer Players

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Relationship Between Amino Acids and Performance Measures of Elite Soccer Players

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I dedicate this paper to my amazing daughter Alexis!

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ABSTRACT

Current theories suggest serum amino acids concentration can be used as an index of fatigue. The purpose was to quantify changes in the biomarkers, and performance measures over time and determine the ability of amino acids to detect changes in athletic performance. Twenty male collegiate soccer players (age, 20.5 ±1.2 y; height, 180 ±6cm; body mass, 78.2 ±6.3kg; body fat percentage, 12.1 ±2.4%; HR_{max}; 200 ±7 $b \cdot min^{-1}$; VO2max, 51.5 ±5.1 $mL \cdot kg \cdot min^{-1}$) were analyzed during this study. Amino acids were analyzed via an extensive panel of blood biomarkers, while performance measures via GPS, accelerometry, and heart rate. Athletes were monitored for 69 training sessions and 24 matches over a period of 4 months. The athletes were asked to provide a total of five blood draws consisting of 147 separate blood and urine biomarkers at various time points throughout the season. Three amino acids (Phenylalanine, Ethanolamine, and Beta-Amino Isobutyric Acid) entered the regression equation and were significantly related to sum playerload/min, F (3,73) = 12.847, p <.001, r = .588, r2 = .346. Five amino acids (Arginine, Alpha-Amino Adipic Acid, Taurine, Aspartic Acid, Histidine) entered the regression equation and were significantly related to sum avg maxvel, F (5,70) = 14.013, p <.001, r = .707, r2 = .500. Of the 19 regressions conducted Phenylalanine resulted in the most significant predictor six times, and Taurine was next with four suggesting further research is needed to identify the mechanism relating these amino acids so strongly to performance.

Chapter 1

Introduction and Review of Literature

Introduction

Athletes are constantly seeking ways to maintain performance over the course of a season. For this, modifications are required to combat performance reductions. One of the factors commonly associated with athletic performance reduction is fatigue.[1] This has led to a scientific approach to monitoring performance measures to understand athletic performance responses and determining which athletes are ready for the demands of competition. However, performance measures do not provide a measure of the intensity of fatigue, and due to the variability between sports what may be classified as a significant performance measure in one sport may not hold true for another sport. Ensuring the appropriate performance measures are being monitored will depend on the sport being monitored, and if a strong enough negative relationship exist between performance measures and fatigue then a reduction in performance may suggest fatigue.

Soccer, a high-intensity intermittent team sport,[2] requiring players to perform a number of physical actions for the duration of approximately 90-minutes per game. Understanding fatigue and how to prevent it, can be beneficial to sustaining athletic performance over the course of an 18-game regular season for a men's collegiate soccer team.

Fatigue is a very important biological alarm that signals the need for rest, but in today's athletic culture of "what have you done for me lately," coaches are constantly under pressure to do what it takes to win. Even if provided with the knowledge of fatigue

and its impact on athletic performance the question begs, does the risk outweigh the reward? In a fatigued state, most first-string athletes are more effective than their non-fatigued second-string teammates. What incentive does this provide for coaches to rest athletes to mitigate the effects of fatigue on athletic performance. Of the many current theories, it is thought that serum amino acids concentration can be used as an index of fatigue,[3] and therefore used as a predictor of fatigue.

While most athletes have felt it, the challenge with fatigue is that nobody knows exactly what it is. The study of the underlying biological mechanisms of fatigue (Fig 1.4) along with the search for biochemical indexes of fatigue has stimulated the efforts of many research groups.[4] By examining the relationships between serum amino acids and performance measures our hope is to identify biochemical indexes of performance measures, that may one day lead to a better understanding of the mechanisms of fatigue. The aim of this paper is to examine the relationship between performance measures and serum amino acid profiles.

Previous Studies

Many previous studies that have examined the relationship between athletic performance and amino acids have only tested single timepoints, before and after an event (Tab 1.1). Athletes don't perform a sporting event once then stop, these studies fail to show the impact an entire season may have on performance. We focused our search on studies where athletes were exposed to multiple athletic performances/training.

Among the various inconsistencies of these studies is the scientific approach to measuring amino acid concentrations. Utilizing urine samples is the most common approach, mainly due to its lower cost and accessibility. It's much easier to urinate in a cup then it is to acquire personnel and a facility capable to draw and store blood respectively, providing a practical and non-invasive means by which teams/organizations can acquire samples. The concern with urine sampling of amino acids is that it represents a very small fraction of the circulating amino acids.[5]

Changes in serum amino acid concentration has been shown to be related to the type of exercise, intensity, and duration rendering previous studies practically irrelevant.[6] Therefore, the findings of previous studies are only specific to those populations and under those specific circumstances and renders them practically irrelevant from an external validity standpoint. Investigations examining amino acid concentrations have utilized various modalities including swimming,[7] triathlon,[8] and cycling.[9] Given the large variation in exercise mode, duration, and intensity across studies, currently there is a gap in the body of knowledge related to sport-specific amino acid concetrations over the course of an athletic season especially in soccer athletes at the division I NCAA level.

Another approach researchers have utilized is supplementing with amino acids based on characteristics that support an ability to inhibit a mechanism of fatigue such as glycogen depletion. Campos-Ferraz examined glucogenic amino acids because of their role to increase muscle alanine production during exercise and the increased release of alanine in the bloodstream resulting in a higher contribution to glucose synthesis.[10] Based on the effect that Branched Chain Amino Acids (BCAA) have on the amount of

serotonin synthesized a separate study involving supplementation of amino acids examined the effect of BCAAs on central fatigue.[11] Future studies of amino acids should examine the effect supplementing with BCAA's have on recovery due to their ability to intervene in muscle protein synthesis by stimulating mRNA translation and preventing muscle proteolysis by inhibiting specific mechanisms of rapamycin.

Amino Acids

During this study 34 amino acids were examined, there were nine essential amino acids; Phenylalanine (phe), Valine (val), Threonine (thr), Tryptophan (trp), Isoleucine (ile), Methionine (met), Leucine (leu), Lysine (lys), Histidine (his), 10 non-essential amino acids; Alanine (ala), Glutamine (gln), Glycine (gly), Glutamic acid (glu), Arginine (arg), Tyrosine (tyr), Serine (ser), Asparagine (asn), Aspartic Acid (asp), Proline (pro), and 15 other amino acids; Hydroxyproline (hyp), Alpha-AminoAdipic Acid (aaa), Sarcosine (sar), Beta-Alanine (bala), Taurine (tau), Citrulline (cit), 1-Methylhistidine (1-mh), Gamma Amino Butyric Acid (gaba), 3-Methylhistidine (3-mh), Beta Amino Isobutyric Acid (baib), Ethanolamine (eta), Alpha Amino Butyric Acid (aaba), Cystathionine (cys), Homocysteine (hcy), Ornithine (orn).

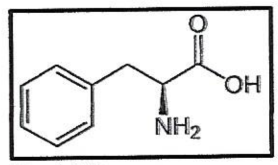
Amino acids are organic compounds containing amine (-NH2) and carboxyl (-COOH) functional groups, along with a side-chain (R group) specific to each amino acid. They can be classified according to the core structural functional groups' locations as alpha- (α-), beta- (β-), gamma- (γ-) or delta- (δ-) amino acids. (Fig 1.1) Proteins are made of amino acids linked together by chains called peptide linkages. During digestion proteins are metabolized to form amino acids, in the same way carbohydrates are

broken down to monosaccharides.[12] In the form of proteins, amino acids comprise the second-largest component of human muscles, water being the largest. Amino acids perform critical roles in processes such as neurotransmitter transport and biosynthesis. Of the large number of amino acids, 20 are common in humans. Of these, eight have been found to be essential for the adult human and have thus been termed, essential amino acids (phenyl-alanine, tryptophan, methionine, lysine, leucine, isoleucine, valine and threonine). A ninth amino acid, histidine, is required for growth and is essential for infants and children. Histidine may also be necessary for tissue repair.

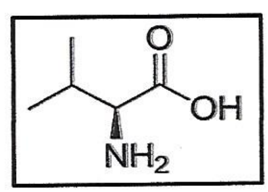
Essential amino acids are amino acids that can't be synthesized in the body thus must be consumed via the diet. These amino acids play a critical role in protein synthesis, and are necessary for a variety of physiological functions.[13] There are also Non-essential amino acids which are broken down into two categories 1) conditionally essential meaning they can be synthesized under special pathophysiological conditions, such a maturity and catabolic stress, 2) dispensable meaning they can be synthesized in the body.[14] BCAA's are considered the most relevant amino acids for exercise physiology. Proteinogenic BCAA (Leuicine, Isoleucine, & Valine) intervene in muscle protein synthesis by stimulating mRNA translation and preventing muscle proteolysis by inhibiting specific mechanisms of rapamycin.[15] Based on the byproducts of their metabolism amino acids are classified as either glucogenic or ketogenic amino acids (Fig 1.2). Glucogenic amino acids can be converted to glucose through gluconeogenesis, the metabolic pathway that generates glucose from non-carbohydrate carbon substrates. Gluconeogenesis mainly used by humans to maintain blood glucose levels, especially during athletic performances when there is a high demand for glucose

as an energy source. Ketogenic amino acids make their contribution to energy production after metabolism, by converting to Acetyl Coenzyme A (ACoA). [16]

Plasma amino acid concentrations change throughout the course of a workout depending on type, intensity, and duration of the workout being performed. While following a single workout session, there will likely be an increase in most amino acids, over the course of an entire season amino acid concentrations will decrease.[17] Some researchers believe this evidence is directly related to fatigue, and that some amino acids are indexes of activity associated with fatigue.[181920]

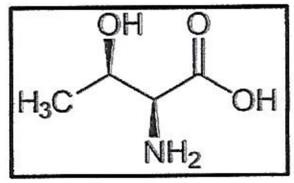


Phenylalanine (Phe) is an α-amino acid with the formula $C_9H_{11}NO_2$. There are three forms of phenylalanine: D-phenylalanine, L-phenylalanine, and the mix made in the laboratory called DL-phenylalanine. D-phenylalanine is not an essential amino acid, and its role in humans needs further research. L-phenylalanine known for its analgesic and antidepressant properties, is an essential amino acid and is the only form of Phe found in proteins. L-phenylalanine is found primarily in eggs, chicken, liver, beef, milk, and soybeans. Phenylketonuria (PKU) a rare metabolic disorder present in individuals missing the enzyme necessary to metabolize Phe. This causes a build-up which if left untreated could cause irreversible brain damage. Newborns are tested for PKU within 48 to 72 of birth, older untreated children become hyperactive with autistic behaviors, including purposeless hand movements and rhythmic rocking. Phe is a precursor for tyrosine.

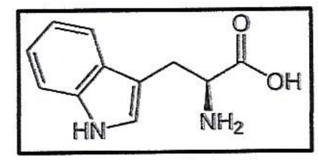


Valine (Val) is an α-amino acid with the formula $C_5H_{11}NO_2$, that competes with tryptophan for transport into the brain. Additionally this competetition for transportation into the brain has

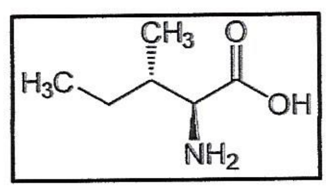
been shown to decrease brain synthesis of 5-hydroxytryptamine (5-HT), commonly known as serotonin.[21]



Threonine (Thr) is an α-amino acid with the formula $C_4H_9NO_3$. Thr is metabolized in two ways: 1) It's converted to pyruvate via threonine dehydrogenase, 2) it's converted to α-ketobutyrate the more common pathway, via the enzyme serine dehydratase and thereby enters the pathway leading to succinyl-CoA.[22]

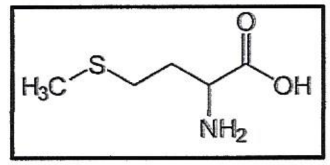


Tryptophan (Trp) is an α-amino acid with the formula $C_{11}H_{12}N_2O_2$. The transport of trp across the blood-brain barrier is the rate-limiting step in the synthesis of serotonin, the neurotransmitter responsible for arousal, sleepiness, and mood. Trp is transported across the blood-brain barrier via the L-system, the amino acid transporter that transports other large neutral amino acids including valine. Therefore the amount of trp transported into the brain depends not only on the concentration of tryptophan in the bloodstream, but also on the concentration of other amino acids, mainly val.[23]

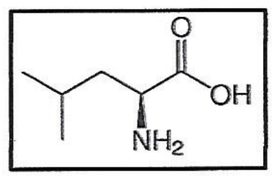


Isoleucine (Ile) is an α-amino acid with the formula $C_6H_{13}NO_2$. Ile is one of the two amino acids that possess both glucogenic and ketogenic properties. The glucogenic pathway begins with transamination with alpha-ketoglutarate then converts to either Succinyl CoA and fed into the TCA cycle for oxidation, or converted into oxaloacetate (OAA) for gluconeogenesis. The ketogenic pathway occurs when Ile is converted to Acetyl CoA and fed into the TCA cycle by condensing with oxaloacetate to form citrate. Acetyl CoA

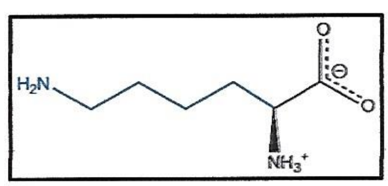
cannot be converted back to glucose in humans but can be used in the synthesis of ketone bodies or fatty acids.[24]



Methionine (Met) is a sulfur containing α-amino acid with the formula $C_5H_{11}NO_2S$ important in angiogenesis, the growth of new blood vessels. Met metabolism has multiple health benefits to include reducing blood cholesterol levels, increasing levels of antioxidants to combat free radicals in the body, and regeneration of liver and kidney tissue. Unfortunately, when it comes to factors of fatigue Met metabolism has not been shown to contribute.[25] However, there is research that suggest Met's impact on new blood vessel growth may impact recovery and muscle adaptations to increased activity.[26]

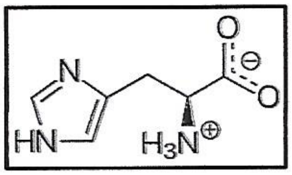


Leucine (Leu) is an α-amino acid with the formula $C_6H_{13}NO_2$. Leu has long been thought to be responsible for muscle growth, however this is all just a misinterpretation. Studies have shown Leu to be associated with the transition of muscle from a catabolic (breaking down) state to an anabolic (building) state. This transition is regulated by Leu's stimulation of mammalian target of rapamycin (mTOR). Stopping muscle from breaking down is not the same as building muscle, however this is the interpretation often given in an attempt to explain functionality of Leu. [27]

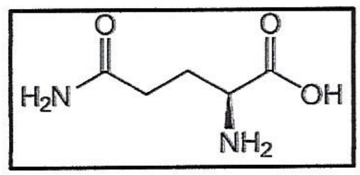


Lysine (Lys) is an α-amino acid with the formula $C_6H_{14}N_2O_2$. Lys acts as an antagonist of serotonin by reducing activity of serotonin receptors.[28] Another study showed Lys to be negatively associated with serotonin levels as well as other perceived factors knows to be regulated by the neurotransmitter. This may not have an impact on

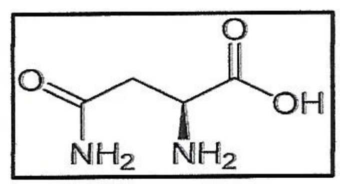
force production but it will impact perceived effort necessary to exert a desired force ultimately effecting athletic performance.



Histidine (His) is an α-amino acid with the formula $C_6H_9N_3O_2$. Previously thought to be essential only for infants has been shown to be essential for adults also.[29] His's most important role is as a precursor to histamine which regulates sleep, and more recent studies have shown to reduce an individual's cognitive function which can impact perceived effort decreasing athletic performance.

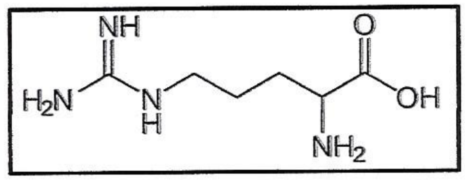


Glutamine (Gln) is an α-amino acid with the formula $C_5H_{10}N_2O_3$. Once inside the cell Gln has two primary pathways 1) hydrolysis then excretion as ammonium + glutamate (a neurotransmitter). Glutamine also has the ability to be synthesized from ammonium + glutamate via enzyme glutamine synthetase 2) partially oxidized to aspartate. This partial oxidation can be connected to the formation of ATP.[30]

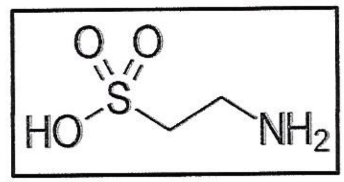


Asparagine (Asn) is an α-amino acid with a chemical formula $C_4H_8N_2O_3$. It's structure is identical to aspartic acid (Asp), except asp's acidic sidechain carboxyl group has been coupled with ammonia. Like gln, asn plays a critical role in the metabolism of toxic ammonia in the body, as well as development and function of the brain.[31] The precursor to asparagine is oxaloacetate. Oxaloacetate is converted to aspartate using a transaminase enzyme. The enzyme transfers the amino group from glutamate to oxaloacetate producing α-ketoglutarate and aspartate. The enzyme asparagine synthetase produces asparagine, AMP, glutamate, and pyrophosphate from

aspartate, glutamine, and ATP [32] the molecule that transports chemical energy within cells for metabolism and other cellular functions.



Arginine (Arg) is an α-amino acid with a chemical formula $C_6H_{14}N_4O_2$. Its most important function with regards to athletic performance is as a precursor to nitric oxide (NO) which stimulates vasodilation. Arg's other functions include reducing healing time of injures[33] and decreasing blood pressure in individuals with hypertension. Arginine is an agonist of the mTOR protein kinase that regulates growth.[34]



Taurine (Tau), a sulfonic acid with a chemical formula $C_2H_7NO_3S$ is commonly characterized as an amino acid, however does not meet the standard criteria of a compound containing both an amino and a carboxyl group. One of Tau primary functions is as a diuretic within the cell, keeping potassium and magnesium in the cell and excess sodium out.[35] Tau's is also essential for cardiovascular function, development and function of skeletal muscle, and the central nervous system.[36] Among individuals with congestive heart failure Tau has been shown to increase the myocardial contractility.[37] A study of mice hereditarily unable to transport Tau showed a reduction of more than 80% of exercise capacity compared to controlled mice without a Tau deficiency, suggesting that it is needed for proper maintenance and functioning of skeletal muscle.[38]

Performance Measures

When monitoring athletic performance, training loads are often measured to quantify athletic performance and can be classified as either external or internal. External loads are work completed by the athlete, measured independently of his/her internal characteristics.[39] An example of an external load in soccer would be average maximal velocity. While external measures are important to understanding athletic performance and capabilities of the athlete, internal measures reflect the relative physiological and psychological stress imposed on the athlete such as HR. Both measures have merit for understanding athletic performance, however a combination of both is important for performance monitoring.[40] For example, if you have a soccer player whose maximum velocity has decrease, this may suggest that the player is fatigue. However, if you have the same soccer player and now you know his heart rate has also decrease, now this suggest he simply isn't putting forth the effort. If this soccer player showed a decrease in maximum velocity coupled with an increase in heart rate this suggests it's costing the athlete more energy to do less work painting a better picture that they may be fatigued.

Given that peak power a muscle can produce during sustained exercise declines as the duration of the exercise increases,[41] duration as a performance measure is important to monitor. Duration should be monitored not to determine if but rather when it significantly affects performance. An alternative factor of athletic performance gaining support by research shows that the duration of time between events "recovery time" is just as, if not more important than the duration of the actual event.[4243]

Time motion analysis, including global positioning system (GPS) tracking and movement pattern analysis are becoming increasingly popular to monitor athletes,

particularly during competition.[44] How fast an athlete runs is underappreciated, however measuring maximum velocity is a reliable representation of how prepared an athlete is for competition. Velocity is a representation of anaerobic performance while distance is a measure of aerobic performance.

Heart rate (HR) is one of the most common means of assessing internal loads in athletes. The use of HR monitoring during exercise is based on the linear relationship between HR and the rate of oxygen consumption. HR has the advantage over oxygen consumption being that it is easier to assess. Heart Rate Reserve (HRR) which is the difference between an individual's measured or predicted maximum heart rate and resting heart rate is used as a method of measuring exercise intensity.

Banister et al[45] proposed a method of quantifying the training load of a training session into a unit dose of physical effort. He suggested that a person's HR response to exercise, along with the exercise duration, collectively called a training impulse (TRIMP), may be a plausible measure of effort, because it is based on the extent to which exercise raises the HR between resting and maximal levels.[46] Further derivations of Banister's initial TRIMP model have been developed. These include Edwards' TRIMP, which uses accumulated time in five arbitrary HR zones multiplied by a weighting factor. Lucia's TRIMP model is like Edwards' however, there are three HR zones that are based on individually determined lactate thresholds and onset of blood lactate accumulation. Further, the use of an individualized TRIMP (iTRIMP) has been developed for use in runners and recently tested in soccer players. The use of iTRIMP reduces issues associated with arbitrary zones and generic weightings and has been

shown to relate better than previous TRIMP models to changes in velocity at 2 mmol L^{-1} in soccer players.

Fatigue

Angelo Mosso, considered the pioneer of the study of fatigue, considered the industrial revolution as the stimuli for modern study of fatigue. From his work A.V. Hill developed an interest in the insight that could be gained from studying human performance. In the 1920's, Hill analyzed world records for both men and women in several sports and concluded, based on how speed decreased as event distance increased, that fatigue was a hard thing to define.[47] Despite acceptance of the notion that fatigue can limit human performance, there are considerable gaps in knowledge of the underlying mechanisms and how they can be managed to maximize athletic human performance.[48] All too often, a single factor is described as the cause of fatigue when in reality fatigue is a combination of factors that contribute to the sequence of events that results in decreased performance. These factors contributing to fatigue are broken down into two categories central and peripheral. These categories both contribute to fatigue but their relative contribution is not easily suitable for quantification or measurement.

To date there has yet to be a consensus as to the definition of fatigue, however Roger Enoka and Doug Stuart have developed one of the more commonly accepted definitions of fatigue as: *an impairment of performance that includes both an increase in the perceived effort necessary to exert a desired force and an eventual inability to produce this force.*[49]

One key advancement in defining fatigue can be seen by the proposed taxonomy of Kruger et al, that acknowledge fatigue as having two attributes: 1) performance fatigue – the decline in an objective measure of performance over a discrete period, and 2) perceived fatigue – changes in the sensation that regulate the integrity of the performer.[50] Two ideas similar to the concepts of peripheral fatigue and central fatigue respectively.

Defining fatigue may not be as critical as quantifying fatigue to understand the influence fatigue has on human performance. First, select a measurable activity strongly associated to the human performance that is effected by fatigue, second choose a reliable laboratory test, valid to predict human performance on the activity, third research relative significance of mechanisms of fatigue to the activity. With such an array of sporting events, it would be irresponsible to establish one test as a predictor of athletic performance for all athletes. To determine how fatigue influences human performance, it is necessary to identify the mechanisms most responsible for establishing levels of fatigue for each activity of human performance.

Kluger's proposed taxonomy suggest that fatigue be defined as a self-reported disabling symptom derived from performance and perceived fatigue. A word of caution is necessary about the use of self-reported measures to assess the level of fatigue, especially in athletes. The competitive nature of these individuals will more than likely lead to under reporting of responses to questions regarding fatigue.[51] Still, measuring perceived fatigue is essential in understanding how fatigue impacts athletic performance.

Measuring peripheral fatigue presents its own challenges due to the inherent match-to-match variability associated with soccer. It is difficult to make meaningful inferences about the effectiveness of interventions during actual games. A number of laboratory based protocols that replicate the demands of a soccer game have been developed. Despite being both valid and reliable, these protocols lack some ecological validity because of the unidirectional nature of the treadmills and the inability to incorporate skills such as dribbling of a soccer ball.[52]

Central Fatigue

Perceived, or more commonly referred to as central fatigue has limited knowledge of its underlying mechanisms due to a lack of research. However, there are several theories as to the mechanisms of central fatigue (Fig 1.3), including changes of glutamatergic and GABA-ergic synaptic signal transmission, as well as increased serotonin.[53] In 1986 it was hypothesized that changes in plasma amino acid concentration could play a role in central fatigue by influencing the synthesis, concentration and release of neurotransmitters, primarily serotonin.[54] Serotonin has multiple functions including the regulation of mood, arousal, and sleepiness. Serotonin is synthesized via the metabolism of L-tryptophan. The rate-limiting step in the synthesis of serotonin is the transport of tryptophan (Trp) across the blood-brain barrier in the brain. Trp is transported via the amino acids transporter system, which also transports other large neutral amino acids including BCAA. Competition between these amino acids for entry into the brain are possible, therefore the amount of Trp transported into

the brain depends not only on the concentration of Trp in the bloodstream, but also on the concentration of BCAA.

Studies have shown that supplementing with BCAA as an effective method to reduce central fatigue, however was shown to be ineffective in preventing peripheral fatigue.[55]

Peripheral Fatigue

Of the two classifications of fatigue, peripheral is better understood as there have been numerous studies on the mechanisms of peripheral fatigue. From a metabolic aspect adenosine-triphosphate (ATP) depletion, accumulation of hydrogen ions (H^+) and inorganic phosphate (Pi), calcium (Ca^{++}) reduction, electrolyte imbalance, and glycogen depletion are hypothesized to have an impact on peripheral fatigue (Fig 1.4). Each of these factors contribution is dependent on the nature of exercise; however glycogen depletion is thought to be the primary mechanism for fatigue. Amino acids, or lack thereof appear to play a role in glycogen depletion one of the primary mechanism of fatigue. Of the 20 amino acids 18 obtain the ability to be used for gluconeogenesis (Fig 1.2), a glycogen sparring metabolic process which generates glucose from non-carbohydrate carbon substrates.

Hydrogen ions increase in response to anaerobic glycolysis, they as well as Pi bind to myosin active sites limiting force output.[56] Ca^{++} was suggested to slow muscle relaxation as the sarcoplasmic reticulum altered its rate of uptake.[57,58] However, as an energy dependent process (requiring ATP) the slowing of relaxation may be the result of a reduced availability of ATP.

Neuromuscular transmission is dependent on intra- and extracellular sodium and potassium concentrations. Therefore, any shift in electrolyte concentration may influence neuromuscular transmission, specifically action potential in the sarcolemma.

Statement of Problem and Research Hypotheses

Coaches are constantly pushing athletes to maximize athletic performance, often with no regard to the athlete's level of fatigue. Fatigue, as a measuring tool when to allow athletes to rest would be monumental. The problem with this concept is that there is no standard definition of "fatigue," nor are there standard tests established to evaluate an athletes level of fatigue. Therefore, it is relevant to ask, what is fatigue? If measures of performance are related to fatigue, can we predict fatigue? If we can identify biochemical alterations, it would provide insight as to the mechanism(s) of fatigue and the development of evaluation/treatment methods for fatigue. Even after there is an accepted definition of fatigue the issue then lies in sport specificity. Muscle activity for a soccer player is vastly different than the muscle activity of a football player, therefore performance measures would vary between sports. Previous studies conducted to measure fatigue have used a variety of test and none have established a point of fatigue nor a definitive indicator of fatigue.

Therefore, the following hypothesis is posed:

1. No relationship exists between or among performance measures and amino acid profiles.

Chapter 2

Scientific Research and Methods

Experimental Approach to the Problem

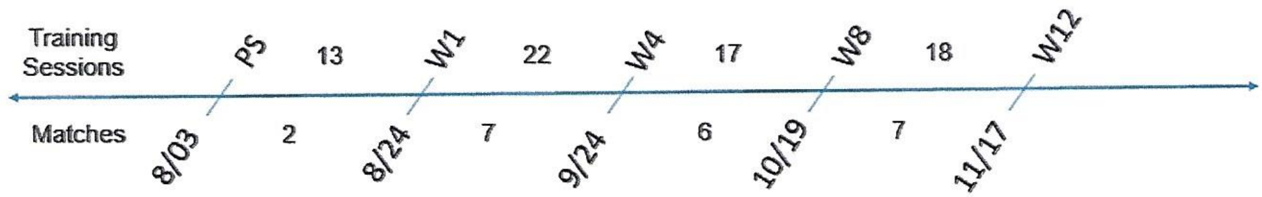
To examine the relationship between serum amino acid concentrations and performance measures of collegiate soccer players over the course of an entire season. First we aim to quantify changes in amino acids between starters and non-starters over the course of a competitive NCAA division I male soccer season, second we aim to examine which performance factors that best predict the changes in performance, third we aim to examine the relationship between amino acids and principle performance factors, and lastly we aim to determine which principal performance factors as well as amino acids should be included in future regression models. Amino acid concentrations were evaluated via an extensive panel of blood biomarkers.

Table 1.2 Amino Acid Abbreviations

Essential Amino Acids (µmol/L)		Others (µmol/L)	
Phenylalanine	(phe)	Hydroxyproline	(hyp)
Valine	(val)	Alpha-Amino Adipic Acid	(aaa)
Threonine	(thr)	Sarcosine	(sar)
Tryptophan	(trp)	Beta-Alanine	(bala)
Isoleucine	(ile)	Taurine	(tau)
Methionine	(met)	Citrulline	(cit)
Leucine	(leu)	1-Methylhistidine	(1mh)
Lysine	(lys)	Gamma-Amino Butyric Acid	(gama)
Histidine	(his)	3-Methylhistidine	(3mh)
Non-Essential Amino Acids (µmol/L)		Beta-Amino Isobutryic Acid	(baib)
Alanine	(ala)	Ethanolamine	(eta)
Glutamine	(gln)	Alpha-Amino Butryic Acid	(abu)
Glycine	(gly)	Cystathionine	(cys)
Glutamic Acid	(glu)	Homocysteine	(hcy)
Arginine	(arg)	Ornithine	(orn)
Tyrosine	(tyr)		
Serine	(ser)		
Asparagine	(asn)		
Aspartic Acid	(asp)		
Proline	(pro)		

Training load data was collected via GPS, accelerometry, and heart rate. Perceived measures were also monitored for mood, anxiety, muscle soreness, and exertion. Players were monitored over a period of 4 months, starting in early August and

ending at the completion of the season in late November. The players were asked to provide a total of 5 blood draws consisting of 34 amino acid profiles at various time points throughout the season. Blood draw collection time points were prior to the start of preseason (PS), and in season at week 1 (W1), week 4 (W4) week 8 (W8), and week



12 (W12).

The aim was to quantify changes in the biomarkers, and performance measures over time and determine the ability of amino acid concentrations to detect changes in athletic performance levels.

Subject Characteristics

Twenty NCAA Division I male soccer players competing in the 2015-2016 season were invited to participate. Prior to the start of the pre-season the subjects' age (20.5 ± 1.2 years) (mean ± SD) (n=20), height (180± 6cm), body mass (78.2 ± 6.3kg, body fat percentage (%BF) (12.1 ± 2.4%), max heart rate (HR_{max}) (200 ± 7 b·min^{-1}), VO_{2max} (51.5 ± 5.1 mL·kg·min^{-1}), and lactate threshold (LT) heart rate at 4mmol·L^{-1} (175 ± 9 b·min^{-1}) were recorded. The players were collegiate soccer players for 2 ± 1 years with several players (n=4) having played zero seasons at the collegiate level. The biomarkers collected in this study did not influence the coaching strategy or strength and conditioning plan put in place by the soccer coaches. Training consisted of 69 formal

sessions, 5-6 sessions per week, and 88 minutes (range 40-135) per training session. All twenty players who began testing at baseline remained in the study from the W1 to the W12 time point however, not all players were able to take part in all blood draws. Two players were unable to provide a sample at W12 time point due to illness and a total of four players were unable to perform the physical fitness testing due to injury or exercise restrictions; prior to season (n=1) and at the end of the season (n=3). Players attended an informational session prior to the start of data collection and provided informed consent and the study was approved by the University of Connecticut Institutional Review Board.

Blood Sampling Procedure and Processing

Blood samples were provided by the participant at 5 time points: prior to the start of preseason (PS), and in season at week 1 (W1), week 4 (W4) week 8 (W8), and week 12 (W12). All blood draws were separated by 1 month during the regular season (W4-W12) with the exception of W1, (21 days after PS). Blood draws occurred between 0700 and 0800 hours and between 32-34 hours after the completion of a match. On all occasions the 24 hours prior to the blood draw were off-days where no training took place. All players were instructed to arrive to the laboratory in a fasted state (no food 8 hours prior of the blood draw). Players were allowed and encouraged to drink fluid prior to arriving to testing. Blood samples were taken by two experienced phlebotomists (Quest Diagnostics™, ExamOne®, Rocky Hill, CT). Blood was collected from the antecubital vein using sterile aseptic technique using a blood collection set (21G, BD Vacutainer™, Safety-Lok™) while in the seated position. Samples were drawn into 8

collection tubes ranging from 2.5-7.5mL containing one of the following anticoagulant (EDTA) (n=4), clot activator (n=4), or whole blood (n=1). Appropriate blood samples were centrifuged, aliquoted, and shipped in either frozen or refrigerated overnight to a processing facility (Quest Diagnostics, Inc. San Juan Capistrano, CA). Blood samples were analyzed immediately upon arrival and results were provided back to the participant, sports medicine physician, and researcher using the MyQuest™ by Care360® online patient portal.

From PS to W1 there were 13 training sessions over the course of 12 days with 3 multi-session days, 2 matches, and two off days. From W1 to W4 there were 22 training sessions, 7 matches (2 away), and 4 off days. From W4 to W8 there were 17 training sessions, 6 matches (3 away), and 8 off days. Last, from W8-W12 there were 18 training sessions, 7 matches (4 away) and 5 off days. W12 of the regular season was four days prior to the final 2 post season games and constituted as the final blood draw time point. From a training standpoint, during PS to W1 this served as a preparation period for the players with an intense pre-season training regimen aimed at improving technical skills and drills at "near match" heart rate intensity and velocity zones. From W1 to W4, training focused on maintaining periodic match-level intensity and velocity with a close eye on GPS and accelerometry based training load. This strategy continued through W8, where maintenance and recovery along with periodic match intensity and velocity was obtained. From W8 to W12, the regular season came to an end with conference playoffs occurring during this time. Overall duration of sessions was reduced while still experiencing match level heart rate intensity and velocity zones with rest and recovery for players that demonstrated signs of fatigue or potential injury.

Anthropometrical and Physiological Testing

During laboratory blood sampling visits, players' height (PS only), body mass, and %BF were assessed. Height was recorded to the nearest centimeter using a standard tape measure while body mass was assessed using a digital scale (T51P, Ohaus, Pine Brook, NJ) and 4-site skin caliper estimate of body fat percentage using the Jackson Pollack equation.(7) Aerobic and anaerobic fitness were assessed using a VO_{2max} and lactate threshold (LT) tests, respectively. For the VO_{2max}, expired gases were collected and analyzed (TrueOne® 2400 Metabolic Measurement System, Parvo Medics, Sandy, UT) during a graded treadmill exercise test (2% grade; increased speed every 2 min) to determine VO_{2max}. Start speed was calculated based previous treadmill VO_{2max} testing performed 4 months earlier, to ensure that test duration was between 8-12 minutes in length so as not to induce muscular fatigue. VO_{2max} was confirmed if players met 2 of the 3 following criteria: (1) Within 10% of age predicted maximum HR (220-age), (2) Rating of perceived exertion greater than 17/20, and (3) respiratory exchange ratio was greater than 1.1. To assess anaerobic fitness, the LT test was conducted 24 hours after the VO_{2max} test to allow for the clearance of lactate. This test consisted of a graded treadmill exercise test (2% grade; increased speed every 3 min) to determine heart rate and velocity at $4 mmol \cdot L^{-1}$ of lactate. Blood was collected via finger prick using sterile procedure with a calibrated lactate meter (Lactate Plus, Nova Biomedical, Waltham, MA). The lactate test was complete when all of the following were satisfied; (1) a rise of $>1.0\ mmol \cdot L^{-1}$ of lactate occurred and (2) $>4.0\ mmol \cdot L^{-1}$ of lactate

was obtained. During both the VO_{2max} and LT tests, HR was obtained using a HR monitor (TIMEX, Digital 2.4, Heart Rate Monitor).

In the present study, we used training loads to quantify performance measures. External and internal load based measurements were obtained. Mean training load was calculated for the following intervals: PS-W1, W1-W4, W4-W8, and W8-W12.

External load measures were collected throughout the study using wearable devices (MinimaxX S4, Catapult Innovations, Australia) equipped with a 10Hz GPS, gyroscope, and 100Hz triaxial accelerometers, or highly sensitive motion sensors that record acceleration of body movement in 3 dimensions. Both accumulated (sum) and mean distance (DST), duration (DUR), max velocity (Vel_{max}), velocity load (VL) and sprint efforts (SP_E) were obtained using GPS while Player Load™ (PL) and $PL \cdot min^{-1}$ was obtained via accelerometry. PL is derived from the triaxial accelerometers utilizing the following formula:

$$Player\ Load = \sqrt{(fwd_{t=i+1}-fwd_{t=i})^2 + (side_{t=i+1}-side_{t=i})^2 + (up_{t=i+1}-up_{t=i})^2}$$

Where fwd = forward acceleration, side = sideways acceleration, up = upward acceleration and t = time.

Internal load measurements for HR were collected using the Polar Team2 System (Polar Electro Oy, Finland) HR monitors. Both mean and sum HR were collected, while only mean time spent between 85%HRR and 95%HRR (HRR85to95), time spent above 95%HRR (HRR95+), and training impulse (TRIMP) were collected throughout the study. The TRIMP method developed by Banister et. al was determined using the following formula:

$$\text{TRIMP Load} = D(\Delta\text{HR ratio})\cdot(\text{Duration})\cdot e^{b(\Delta\text{HR ratio})},$$

Where D = duration of training session, b = 1.67 for females and 1.92 for males and ((ΔHR ratio) is determined using the following equation:

$$\Delta HR\ ratio = (HR_{ex} - HR_{rest})/(HR_{max} - HR_{rest})$$

Where HR_{rest} = the average heart rate during rest and HR_{ex} = the average HR during exercise. HR_{max} was determined either during a pre-season VO_2max test or Yo-Yo Intermittent Recovery Test (YYIRT) while resting HR_{rest} values were collected in the morning upon arrival to the laboratory in a quiet and dark room while in the supine position. HR_{rest} values were collected over a period of 10-minutes. HR data during training was measured at each formal practice and match and HR_{ex} values were utilized to determine the TRIMP load.

Statistical Analysis

A factor analysis was performed to determine the extent to which measurement overlap exist among a set of variables. A 2-tailed Pearson's Correlation was used to evaluate the strength of relationships between performance measures and amino acids. A stepwise regression was used to determine the amount of variance (of the performance measures) explained by predictors (amino acids). A two-way analysis of variance (ANOVA), with group and time as the two between group factors was conducted to understand their interaction on amino acids. All data are expressed as mean ± standard error of the mean (SEM) unless otherwise stated. A probability level of <0.05 was adopted throughout to determine statistical significance.

Chapter 3

Results

Two-Way ANOVA

For aim 1 a two-way ANOVA was conducted that examined the effect of groups and time (2x5) on amino acids profiles. The two groups examined were starters and non-starters, while the five time points were PS, W1, W4, W8, W12. Of the 34 amino acids examined only Sarcosine showed statistical significance interaction between the effects of groups on amino acids F (1,2) = 289.000, p = .003 (Tab 3.33). In contrast 29 of the 34 amino acids showed statistically significant interaction between the effects of time on amino acids (Tab 3.34), while no amino acids showed a significant interaction between the effects of group and time on amino acids.

Table 3.34 - Amino Acid Two-Way ANOVA

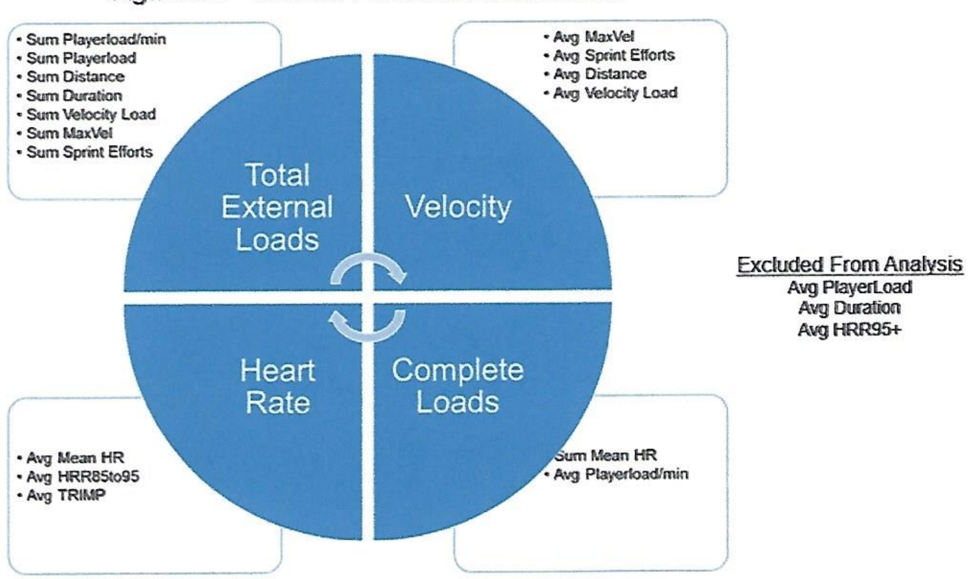
Essential Amino Acids	Group	Time	Group*Time	Others (μmol/L)	Group	Time	Group*Time
Phenylalanine	.966	.000*	.272	Hydroxyproline	.193	.000*	.076
Valine	.270	.116	.501	Alpha-Amino Adipic Acid	1.000	.029*	.900
Threonine	.500	.001*	.790	Sarcosine	.003*	.038*	.093
Tryptophan	.706	.000*	.634	Beta-Alanine	.757	.000*	.658
Isoleucine	.720	.293	.858	Taurine	.177	.019*	.254
Methionine	.715	.011*	.319	Citrulline	.930	.000*	.207
Leucine	.680	.004*	.725	1-Methylhistidine	.813	.524	.418
Lysine	.242	.000*	.622	Gamma-Amino Butyric Acid	.880	.000*	.880
Histidine	.056	.000*	.228	3-Methylhistidine	.186	.000*	.805
Non-Essential Amino Acids	**Group**	**Time**	**Group*Time**	Beta-Amino Isobutryic Acid	.743	.000*	.905
Alanine	.816	.009*	.128	Ethanolamine	1.000	.000*	.995
Glutamine	.868	.000*	.275	Alpha-Amino Butryic Acid	.666	.000*	.326
Glycine	.616	.002*	.087	Cystathionine			
Glutamic Acid	.791	.038*	.965	Homocysteine			
Arginine	.549	.000*	.500	Ornithine	.108	.000*	.876
Tyrosine	.516	.000*	.717	*Computed using alpha = .05			
Serine	.627	.000*	.142	Group - Starters vs. Non-Starters			
Asparagine	.487	.000*	.575	Time - PS to W12			
Aspartic Acid	.265	.000*	.099				
Proline	.359	.000*	.247				

Factor Analysis

For aim 2 a principle components analysis (PCA) was conducted to identify if any underlying factors exists among 19 performance measures. Four criteria were used to determine the appropriate number of components to retain: eigenvalue, variance, scree plot, and residuals: 1) Eigen value - Components with eigenvalues greater than 1 should be retained. These criteria are reliable when the number of variables is <30 and communalities are >.70, or the number of individuals is >250 and the mean communality is >.60. 2) Variance - Retain components that account for at least 70% total variability. 3) Scree Plot - Retain all components with the sharp descent, before eigenvalues level off. These criteria are reliable when the number of individuals is >250 and communalities are >.30. 4) Residuals - Retain the components generated by the model if only a few residuals exceed .05. if several reproduced correlations differ, you may want to include more components.[59]

A Principle component analysis was conducted for the 19 performance measures utilizing a varimax rotation.

Figure 3.1 – Athletic Performance Measures



The initial analysis (FA1) retained four factors that explained 45%, 21%, 9%, and 7% of the variance respectively. Factors 5 to 19 all had eigen values <1.000 (Tab. 3.1). All 19 items correlated >.7 with at least one other item, suggesting reasonable factorability (Tab. 3.3). The Kaiser-Meyer-Olkin measure of sampling adequacy was 0.650 barely above the recommended value of 0.600, and Bartlett's test of sphericity was significant χ^2 (171) = 2617.615, p<.05 (Tab. 3.2). The reproduced correlation matrix showed that 36 (21.0%) non-redundant residuals had values >.05. Inclusion of additional components increase the model fit as it decreased the number of residuals >.05 (Tab. 3.6). Finally, the communalities were all above .7 except for avg of dur, and avg HRR95+ (Tab. 3.4). Criteria from this analysis suggest other analysis should be investigated. Thus, two more PCA were conducted 1) to retain six components 2) to exclude avg dur, and avg HRR95+

The six-factor solution (FA2) explained 91% of the variance an 8% increase from FA1. Factors five and six had eigen values <1.000 (Tab. 3.7). Communality (Tab. 3.9) values improved (all values > 0.827), and non-redundant residuals with values greater than 0.05 was 22 (12%) a decrease of 9% from FA1(Tab 3.12).

The exclusion solution (FA3) which excluded avg dur and avg hrr95+ retained four factors that explained 88% of the variance an 5% increase from FA1 (Tab. 3.13). Communality (Tab. 3.16) values improved (all values > 0.791) from FA1, and non-redundant residuals with values greater than 0.05 was 26 (19%) a decrease of 2% from FA1(Tab 3.18). The Kaiser-Meyer-Olkin measure increased from barely above the recommended to .759, and Bartlett's test of sphericity was significant χ^2 (136) = 2396.497, p<.05 (Tab. 3.14).

After rotation, the first factor accounted for 49.94%, the second for 22.93%, the third 8.53%, and the third for 7.09%. Factor 1 had seven variables all with positive

Table 3.19 - Factor Loading

	Loading		Loading
Factor 1: Total External Load		**Factor 3: Heart Rate**	
sum playerload/min	0.970	avg mean HR	0.898
sum playerload	0.963	avg HRR85to95	0.866
sum distance	0.889	avg TRIMP	0.815
sum duration	0.872		
sum velocity load	0.837	**Factor 4: Complete Load**	
sum max velocity	0.791	sum mean HR	-0.770
sum sprint efforts	0.734	avg playerload/min	0.738
Factor 2: Velocity			
avg max velocity	0.897		
avg spring efforts	0.847		
avg distance	0.788		
avg velocity load	0.764		

loadings and addressed total external load. Factor 2: velocity, and factor 3: heart rate loaded four and three variable respectively. Factor 4 addressed complete load and was the only factor to include items with both negative and positive loading. The positively loaded variable was avg playerload/min, and the negatively loaded variable was sum meanHR (Tab 3.19).

Stepwise Regression

After the factor analysis was complete, the performance measures with the greatest correlation to each athletic performance factor was used to conduct a stepwise multiple regression to examine the relationships of various potential predictors (amino acids). Sum playerload/min showed a correlation of 0.970 with total external load, avg maxvel showed a correlation of 0.897 with velocity, and avg meanHR showed a

correlation of 0.898 with heart rate. Of the 19 regressions conducted phe resulted in the most significant predictor six times, and tau was next with four (Tab. 3.35) suggesting further research is needed to identify the mechanism relating these amino acids so strongly to performance.

Table 3.35 - Stepwise Regression

	Predictors	R	R^2	Most Sig Predictor	$\%R^2$		Predictors	R	R^2	Most Sig Predictor	$\%R^2$
Avg Distance	4	.543	.295	Taurine	.137 46.4%	Avg VelocityLoad	6	.542	.294	Taurine	.105 35.7%
Sum Distance	3	.524	.275	Phenylalanine	.140 50.9%	Sum VelocityLoad	3	.528	.278	Phenylalanine	.127 45.7%
Avg of Duration	4	.625	.391	Hydroxyproline	.167 42.7%	Average PlayerLoad	4	.649	.421	Taurine	.207 49.2%
Sum of Duration	10	.792	.627	Phenylalanine	.152 24.2%	Sum PlayerLoad	4	.612	.375	Phenylalanine	.214 57.1%
Avg MaxVel	5	.707	.500	Arginine	.179 35.8%	Avg Player Load min	5	.649	.421	Proline	.107 25.4%
Sum MaxVel	4	.645	.416	GABA	.203 48.8%	Sum Player Load min	3	.588	.346	Phenylalanine	.232 67.1%
Avg MeanHR	5	.633	.401	Ornithine	.170 42.4%	Avg TRIMP	3	.555	.308	Ornithine	.181 58.8%
Sum MeanHR	6	.739	.546	Asparagine	.218 39.9%	Avg HRR95+	1	.323	.105	Glutamine	.105 100.0%
Avg SprintEfforts	3	.473	.224	Taurine	.082 36.6%	Avg HRR85t095	3	.454	.206	Ornithine	.097 47.1%
Sum SprintEfforts	3	.535	.287	Phenylalanine	.127 44.3%						

There were three amino acids (phe, eta, and baba) that entered the regression equation and was significantly related to sum playerload/min, $F(3,73) = 12.847$, $p < .001$ (Tab. 3.21). The multiple correlation coefficient was .588, indicating approximately 34.6% of the variance of sum playerload/min could be accounted for by the three amino acids (Tab. 3.20). The regression equation for predicting sum playerload/min was:

Sum playerload/min = 179.528 – (phe x 1.795) + (eta x 4.161) + (baba x 16.568)

There were five amino acids (arg, aaa, tau, asp, his) that entered the regression equation and was significantly related to avg maxvel, $F(5,70) = 14.013$, $p < .001$ (Tab 3.25). The multiple correlation coefficient was .707, indicating approximately 50.0% of the variance of avg maxvel could be accounted for by the five amino acids (Tab. 3.24). The regression equation for predicting avg maxvel was:

$$\textbf{Avg maxvel} = 7.000 + (arg \times 0.012) - (aaa \times 0.561) - (tau \times 0.019) + (asp \times 0.046) - (his \times 0.023)$$

There were five amino acids (orn, thr, gly, his, arg) that entered the regression equation and was significantly related to avg meanHR, $F(5,70) = 9.379$, $p < .001$ (Tab 3.28). The multiple correlation coefficient was .633, indicating approximately 40.1% of the variance of avg meanHR could be accounted for by the five amino acids (Tab 3.27). The regression equation for predicting avg meanHR was:

$$\textbf{Avg meanHR} = 170.327 - (orn \times 0.153) + (thr \times 0.129) - (gly \times 0.114) - (his \times 0.282) + (arg \times 0.189)$$

Chapter 4

Discussion

There are several reasons why monitoring training loads have become a scientific approach to understanding athletic performance and determining which athletes are ready for the demands of competition. Monitoring these loads may provide an explanation for decreases in game performance minimizing the degree of uncertainty associated with changes in performance. However, training loads do not provide a measure of the intensity of fatigue, and due to the variability between sports what may be classified as a relevant training load in one sport may not hold true for the next. An additional benefit of monitoring training loads is to reduce the risk of injury and illness.

Not all sports teams participate in athlete monitoring, for some programs/organizations insufficient resources can be a primary reason for not developing a system to monitor training loads. Resources can be in the form of money, time, or equipment; however, it is commonly a lack of knowledge or experience with monitoring training loads that results in an inability to develop a practical and sustainable system to monitor performance. Understanding why the monitoring is taking place, how often the monitoring will occur, and how the data will be interpreted and presented is critical to develop a serviceable system, Additionally, ensuring the appropriate training loads are being monitored will depend on the sport being monitored. Training loads critical to monitoring football players such as force generation may be irrelevant when monitoring the athletic performance of a soccer player.

The major findings of this study for aim 1 was that sarcosine was the only amino acids to show a significant difference between starters and non-starters. Of the previous studies to examine amino acid profiles to our knowledge no other studies have examined >10 amino acid, while also examining the differences between two non-diseased groups. However, a separate analysis of this same study examined differences between training loads of starters and non-starters and showed multiple significant differences. This anomaly provides an intriguing question for future researchers to try and identify. Why would two items with a moderate/high correlation have two completely different outcomes when analyzed separately.

For aim 2 the major findings were that total external load (sum PL/min), velocity (avg VEL_{max}), heart rate (avg meanHR), and complete load (sum meanHR) were retained and explained 88% of the variance. Bartlett's test of sphericity was significant χ^2 (136) = 2396.497, p<.05, KMO .759, and communalities >.791. To our knowledge there were no previous studies to also examine >15 performance measures, and utilize a factor analysis to identify the best factors. By eliminating insignificant factors this will allow future researchers to quickly identify which performance measures will provide meaningful feedback as well as identify performance measures providing similar data.

For aim 3 the major findings were sum PL/min: F (3,73) = 12.847, p <.001, r = .588, r^2 = .346 with three amino acids (phe, eta, and baba) significantly related. The analysis also generated a prediction equation: Sum playerload/min = 179.528 – (phe x 1.795) + (eta x 4.161) + (baba x 16.568). The prediction equation that resulted from the stepwise regression is meaningless however without a quantitative point for training loads to be identified as being fatigued. Areces[60] study of triathletes didn't compare

amino acid profiles against performance measures to confirm decreases in athletic performance. However, they did analyze pre and post amino acids levels for athletes during a triathlon. Both studies showed the same negative effect for phe. For our study phe increased as performance levels decreased, and for the triathletes phe levels increased from pre to post race confirming our findings that phe may have a negative relationship with performance measures. What we did for our study was take that relationship one step further and report the variance, or how much of the performance measure is explained by the amino acid. If studies continue to show a significant variance explained by the same amino acid the next step should be analyzing the effect supplementing with that amino acid has on performance measures.

Campos-Ferraz study tested supplementation of Leucine because of it's characteristic to stimulate mTOR, inhibiting catabolic reactions. For aim 4, phe resulted in the most significant predictor of a performance measure for six of the 19 regressions conducted, and tau was next with four suggesting further research is needed to identify the mechanism causing these amino acids to predict training loads so often. A possible control for future studies should examine the effect supplementing with these amino acids have on performance measures.

Limitation

Although this study was carefully prepared there were still limitations of the study. Since amino acids can be consumed through our diet or supplemented, lack of available and/or reliable data regarding what the athletes did or otherwise didn't consume could potentially impact amino acid profiles. When left to individuals to explain their dietary

consumption, described values are primarily lower than the actual amount consumed. Additionally, self-reported data is limited by the fact that it rarely can be independently verified. Asking an athlete what they ate for dinner is just as arbitrary as asking them how fatigued they feel. The only way to ensure diet does not play a role in a study is to establish dietary consumption and restriction for the athletes.

The second limitation of this study was the performance measure Heart Rate Reserve. HRR is based on resting HR, as an athlete's fitness level changes so will their resting HR. Unless resting HR is remeasured throughout the season the changes in HRR utilization may not be accurately captured.

The last limitation involves the assumption that decreases in training loads resulted in unsuccessful athletic performances. Neither in our study nor any other study we found was performance examined. In most sports success and failure is measured in wins and losses. If we aim to increase athletic performance, we should first confirm that what we are measuring has an effect on the outcomes of these sporting events.

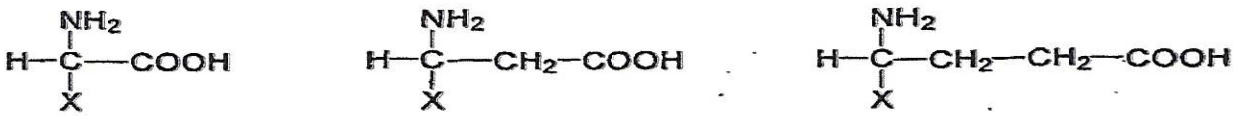
Conclusion

This study confirms a relationship between amino acids and performance measures. Professional and collegiate sports teams as well as forthcoming research should incorporate the given finding when selecting training loads to monitor for soccer players. Analyzing biochemical markers post match as well as following the rest day after a match may provide insight into the time it takes if at all, for an athlete to recover to pre match levels. Recovery time needed to return to pre match levels should be examined to see if a relationship exist with performance measures. Lastly, future

studies should include examining the effect supplementing with the amino acids, shown to have the strongest relationships, have on performance measures.

Despite both the increasing amounts of research and the popularity of load monitoring in high-performance sports, a single definitive tool that is accurate and reliable is not available. Indeed, the nature of the monitoring is likely to be very different depending on the sport and more than one monitoring tool is often utilized. The hope is that one day we can identify specific biochemical relationships to training loads and determine how to delay or eliminate decreases in athletic performances and ultimately enhance not only an athletic performances but the entire sporting community and provide more competitive and entertaining sporting events.

Appendix A



alpha-amino acid | beta-amino acid | gamma-amino acid

Fig. 1.1 Amino acid structure: α-alpha, β-beta, γ-gamma

Amino Acids	
Essential	**Non-Essential**
Phenyl-Alanine ***	Alanine *
Tryptophan ***	Glutamine *
Methionine *	Glycine *
Lysine **	Glutamic Acid *
Leucine #**	Arginine *
Isoleucine #***	Tyrosine ***
Valine #*	Serine *
Threonine ***	Asparagine *
Histidine *	Aspartic Acid *
	Proline *
	Cysteine *

Branched-Chain Amino Acids

*Glucogenic - can be converted to clucose upon metabolism

**Ketogenic - generates ACoA upon metabolism

***Glucogenic & Ketogenic

Fig. 1.2 Amino acids classification

Table 1.1 Previous Studies

Author / Year Journal	Amino Acids Tested	Exercise / Sport Conducted	Results
Campos-Ferraz 2013 [29] Nutrition	BCAA mix, Leucine, Placebo	Swimming (mice)	- Leucine supplementation enhanced resistance to exhaustion compared to BCAA mix ($P < 0.001$) - Muscle & Liver glycogen were significantly spared in the Leucine and BCAA mix ($P < 0.01$) compared to the Pacebo
Areces 2105 [10] PLOS-One	Essential & Non-Essential	Half- Ironman Triathlon	- Essential (-27.1 ±13.0%; $P < 0.001$) and Non-Essential (-24.4 ±13.1%; $P < 0.001$) Amino Acids concentration were significantly reduced after the race - Tryptophan/BCAA ratios increased by 42.7 ±12.7% after the race - Pre and Post changes to amino acids did not correlate with muscle performance variables - Pre and Post changes to amino acids did not correlate with post race creatine kinase concentration
Corsetti 2016 [48] Amino Acids	Essential, Non-Essential [Asparagine, Tryptophan, Aspartic Acid excluded], Misc (Urinary)	Cycling	- Taurine and Carnosine were significantly correlated with the muscle activity markers (Lactate Dehydrogenase, Aspartate Aminotransferase, Creatine Kinase) and indicies of effort
Horvath 2016 [48] Amino Acids	Taurine & β-Alanine	Ex Vivo	- Taurine reduced body and muscle mass, enhanced fatigue resistance, and enhanced force recovery - β-Alanine enhanced fatigue resistance

Appendix B

↑ Uptake of BCAA by the muscle ↓ Plasma level of BCAA ↑ Release of Free Fatty Acids ↑ Plasma free Tryptophan ↑ Transport of Tryptophan into the brain ↑ Synthesis and release of serotonin	=	↑ Tryptophan/BCAA ratio (Causes fatigue)

Fig. 1.3 A theory to explain the cause of central fatigue during exercise

Peripheral fatigue	Depletion of phosphocreatine Accumulation of lactate (protons) Depletion of glycogen Failure of neuromuscular transmission
Central fatigue	Low level of blood glucose Changes in the plasma concentration of amino acids

Fig. 1.4 Possible causes of fatigue during exercise

Appendix C

Table 3.1 - Total Variance Explained

Component	Initial Eigenvalues Total	Initial Eigenvalues % of Variance	Initial Eigenvalues Cumulative %	Loadings Total	Loadings % of Variance	Loadings Cumulative %	Rotation Sums of Squared Loadings Total	Rotation Sums of Squared Loadings % of Variance	Rotation Sums of Squared Loadings Cumulative %
1	8.585	45.186	45.186	8.585	45.186	45.186	5.901	31.056	31.056
2	4.126	21.714	66.900	4.126	21.714	66.900	4.908	25.830	56.885
3	1.759	9.257	76.157	1.759	9.257	76.157	2.743	14.439	71.325
4	1.348	7.096	83.253	1.348	7.096	83.253	2.266	11.928	83.253
5	0.829	4.362	87.615						
6	0.665	3.501	91.116						
7	0.501	2.636	93.751						
8	0.347	1.827	95.578						
9	0.253	1.330	96.908						
10	0.210	1.103	98.011						
11	0.141	0.742	98.752						
12	0.102	0.536	99.288						
13	0.052	0.273	99.561						
14	0.040	0.213	99.774						
15	0.024	0.124	99.898						
16	0.009	0.050	99.948						
17	0.006	0.032	99.980						
18	0.003	0.015	99.994						
19	0.001	0.006	100.000						

Extraction Method: Principal Component Analysis.

Table 3.2 - KMO and Bartlett's Test

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		0.650
Bartlett's Test of Sphericity	Approx. Chi-Square	2617.615
	df	171
	Sig.	0.000

Table 3.3 - Rotated Component Matrix[a]

	Component 1	2	3	4
Sum_PlayerLoad_min	0.974			
Sum_PlayerLoad	0.952			
Sum_Distance	0.871	0.412		
Sum_of_Duration	0.845			0.444
Sum_VelocityLoad	0.838	0.444		
Sum_MaxVel	0.774			0.537
Sum_SprintEfforts	0.740	0.505		
Avg_Distance	0.319	0.880		
Avg_VelocityLoad		0.863	0.310	
Avg_MaxVel		0.835		0.363
Avg_SprintEfforts		0.835		
Average_PlayerLoad		0.717	0.357	
Avg_MeanHR			0.807	
Avg_HRR85t095		0.377	0.795	
Avg_TRIMP		0.523	0.737	
Avg_HRR95+			0.655	
Sum_Mean_HR2	0.466			0.766
Average_of_Duration				0.725
Avg_PlayerLoad_min	0.324	0.380	0.375	-0.622

Extraction Method: Principal Component Analysis.
a. Rotation converged in 8 iterations.

Table 3.4 - Communalities

	Initial	Extraction
Average_of_Duration	1.000	0.605
Sum_of_Duration	1.000	0.952
Average_PlayerLoad	1.000	0.768
Sum_PlayerLoad	1.000	0.968
Avg_PlayerLoad_min	1.000	0.777
Sum_PlayerLoad_min	1.000	0.982
Avg_TRIMP	1.000	0.847
Avg_Distance	1.000	0.928
Sum_Distance	1.000	0.950
Avg_MaxVel	1.000	0.839
Sum_MaxVel	1.000	0.910
Avg_VelocityLoad	1.000	0.908
Sum_VelocityLoad	1.000	0.913
Avg_MeanHR	1.000	0.747
Sum_Mean_HR2	1.000	0.825
Avg_HRR95+	1.000	0.520
Avg_HRR85t095	1.000	0.783
Avg_SprintEfforts	1.000	0.788
Sum_SprintEfforts	1.000	0.809

Extraction Method: Principal Component Analysis.

Table 3.5 – Scree Plot

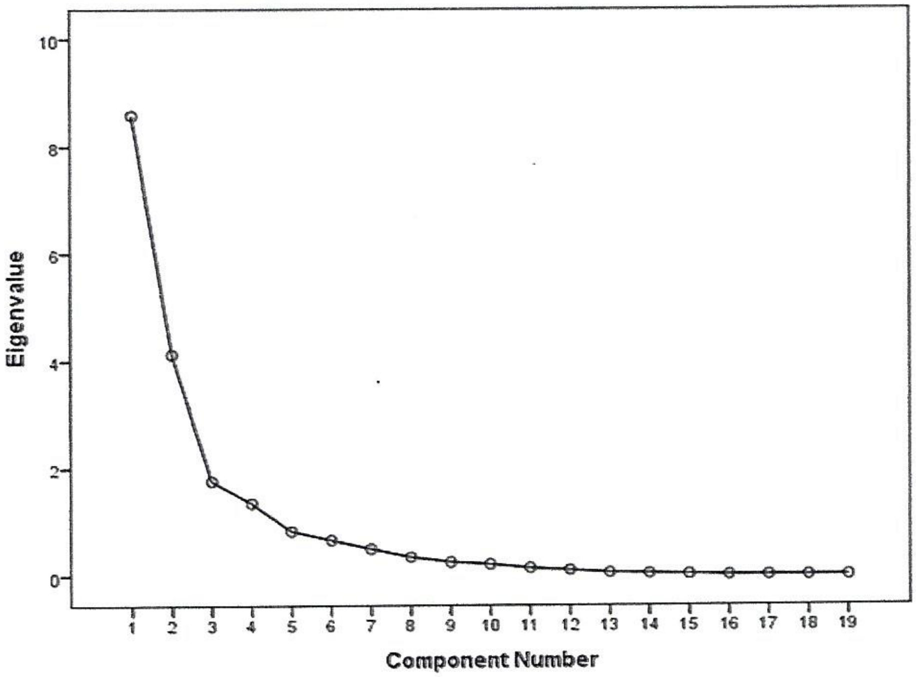


Table 3.6 - Reproduced Correlations

		Average_of_Duration	Sum_of_Duration	Average_PlayerLoad	Sum_PlayerLoad	Avg_PlayerLoad_min	Sum_PlayerLoad_min	Avg_TRIMP	Avg_Distance	Sum_Distance	Avg_MaxVel	Sum_MaxVel	Avg_VelocityLoad	Sum_VelocityLoad	Avg_MeanHR	Sum_Mean_HR2	Avg_HRR95+	Avg_HRR85to95	Avg_SprintEfforts	Sum_SprintEfforts
Reproduced Correlation	Average_of_Duration	.605[a]	0.366	0.048	0.170	-0.333	0.028	0.208	0.221	0.280	0.498	0.486	0.177	0.127	-0.062	0.553	-0.121	0.114	0.286	0.249
	Sum_of_Duration	0.366	.952[a]	0.012	0.800	-0.105	0.759	-0.014	0.132	0.765	0.171	0.891	0.031	0.621	-0.268	0.731	-0.341	-0.093	0.135	0.614
	Average_PlayerLoad	0.048	0.012	.768[a]	0.438	0.633	0.401	0.656	0.810	0.507	0.540	0.179	0.818	0.600	0.504	-0.103	0.058	0.570	0.712	0.557
	Sum_PlayerLoad	0.170	0.800	0.438	.968[a]	0.379	0.956	0.284	0.520	0.936	0.307	0.795	0.441	0.904	0.045	0.450	-0.264	0.190	0.444	0.831
	Avg_PlayerLoad_min	-0.333	-0.105	0.633	0.379	.777[a]	0.442	0.458	0.551	0.348	0.115	-0.052	0.580	0.518	0.490	-0.348	0.138	0.446	0.419	0.387
	Sum_PlayerLoad_min	0.028	0.759	0.401	0.956	0.442	.982[a]	0.204	0.443	0.891	0.167	0.712	0.369	0.888	0.026	0.347	-0.256	0.132	0.353	0.784
	Avg_TRIMP	0.208	-0.014	0.656	0.284	0.458	0.204	.847[a]	0.663	0.351	0.475	0.197	0.705	0.407	0.714	0.130	0.355	0.799	0.597	0.388
	Avg_Distance	0.221	0.132	0.810	0.520	0.551	0.443	0.663	.928[a]	0.633	0.743	0.331	0.910	0.681	0.410	0.014	-0.078	0.532	0.846	0.680
	Sum_Distance	0.280	0.765	0.507	0.936	0.348	0.891	0.351	0.633	.950[a]	0.477	0.809	0.550	0.902	0.056	0.463	-0.295	0.229	0.567	0.864
	Avg_MaxVel	0.498	0.171	0.540	0.307	0.115	0.167	0.475	0.743	0.477	.839[a]	0.384	0.705	0.420	0.151	0.214	-0.192	0.323	0.740	0.519
	Sum_MaxVel	0.486	0.891	0.179	0.795	-0.052	0.712	0.197	0.331	0.809	0.384	.910[a]	0.239	0.666	-0.116	0.750	-0.282	0.091	0.332	0.685
	Avg_VelocityLoad	0.177	0.031	0.818	0.441	0.580	0.369	0.705	0.910	0.550	0.705	0.239	.908[a]	0.618	0.488	-0.047	0.006	0.589	0.824	0.611
	Sum_VelocityLoad	0.127	0.621	0.600	0.904	0.518	0.888	0.407	0.681	0.902	0.420	0.666	0.618	.913[a]	0.172	0.283	-0.221	0.302	0.587	0.840
	Avg_MeanHR	-0.062	-0.268	0.504	0.045	0.490	0.026	0.714	0.410	0.056	0.151	-0.116	0.488	0.172	.747[a]	-0.111	0.501	0.734	0.336	0.110
	Sum_Mean_HR2	0.553	0.731	-0.103	0.450	-0.348	0.347	0.130	0.014	0.463	0.214	0.750	-0.047	0.283	-0.111	.825[a]	-0.092	0.074	0.068	0.342
	Avg_HRR95+	-0.121	-0.341	0.058	-0.264	0.138	-0.256	0.355	-0.078	-0.295	-0.192	-0.282	0.006	-0.221	0.501	-0.092	.520[a]	0.436	-0.097	-0.262
	Avg_HRR85to95	0.114	-0.093	0.570	0.190	0.446	0.132	0.799	0.532	0.229	0.323	0.091	0.589	0.302	0.734	0.074	0.436	.783[a]	0.466	0.265
	Avg_SprintEfforts	0.286	0.135	0.712	0.444	0.419	0.353	0.597	0.846	0.567	0.740	0.332	0.824	0.587	0.336	0.068	-0.097	0.466	.788[a]	0.612
	Sum_SprintEfforts	0.249	0.614	0.557	0.831	0.387	0.784	0.388	0.680	0.864	0.519	0.685	0.611	0.840	0.110	0.342	-0.262	0.265	0.612	.809[a]
Residual[b]	Average_of_Duration		0.029	0.125	0.048	0.145	0.026	0.004	-0.016	0.011	-0.116	-0.101	0.001	0.023	-0.010	-0.092	-0.019	-0.059	-0.119	-0.096
	Sum_of_Duration	0.029		0.010	0.017	-0.037	0.022	-0.002	-0.012	-0.003	0.001	-0.036	0.018	-0.001	-0.007	-0.053	0.018	0.018	-0.003	-0.018
	Average_PlayerLoad	0.125	0.010		0.062	0.064	0.019	0.033	0.038	0.011	-0.043	-0.041	-0.020	-0.080	-0.018	0.048	-0.039	-0.086	-0.121	-0.127
	Sum_PlayerLoad	0.048	0.017	0.062		-0.007	0.014	0.036	0.013	0.018	-0.013	-0.036	-0.014	-0.028	-0.018	-0.020	-0.007	-0.017	-0.041	-0.038
	Avg_PlayerLoad_min	0.145	-0.037	0.064	-0.007		-0.001	-0.050	0.019	-0.027	0.015	0.037	-0.014	-0.019	0.012	0.082	0.031	-0.116	-0.067	-0.070
	Sum_PlayerLoad_min	0.026	0.022	0.019	0.014	-0.001		0.002	-0.003	-0.003	0.014	-0.019	0.003	-0.005	-0.004	-0.020	0.007	0.000	-0.020	-0.028
	Avg_TRIMP	0.004	-0.002	0.033	0.036	-0.050	0.002		0.025	0.050	-0.029	-0.035	-0.064	-0.039	-0.043	-0.046	-0.067	-0.004	-0.039	-0.014
	Avg_Distance	-0.016	-0.012	0.038	0.013	0.019	-0.003	0.025		0.033	0.013	0.010	-0.011	-0.033	-0.006	0.038	0.022	-0.066	-0.059	-0.055
	Sum_Distance	0.011	-0.003	0.011	0.018	-0.027	-0.003	0.050	0.033		-0.003	-0.021	-0.029	-0.016	-0.007	-0.028	0.006	-0.024	-0.036	-0.009
	Avg_MaxVel	-0.116	0.001	-0.043	-0.013	0.015	0.014	-0.029	0.013	-0.003		0.066	-0.020	-0.003	0.026	0.010	0.089	-0.046	-0.034	-0.017
	Sum_MaxVel	-0.101	-0.036	-0.041	-0.036	0.037	-0.019	-0.035	0.010	-0.021	0.066		-0.004	0.010	0.021	0.055	0.028	-0.024	0.011	0.012
	Avg_VelocityLoad	0.001	0.018	-0.020	-0.014	-0.014	0.003	-0.064	-0.011	-0.029	-0.020	-0.004		0.045	-0.014	0.013	0.023	0.034	0.006	-0.024
	Sum_VelocityLoad	0.023	-0.001	-0.080	-0.028	-0.019	-0.005	-0.039	-0.033	-0.016	-0.003	0.010	0.045		0.006	-0.048	0.007	0.056	0.009	0.020
	Avg_MeanHR	-0.010	-0.007	-0.018	-0.018	0.012	-0.004	-0.043	-0.006	-0.007	0.026	0.021	-0.014	0.006		0.040	-0.218	-0.020	-0.049	-0.038
	Sum_Mean_HR2	-0.092	-0.053	0.048	-0.020	0.082	-0.020	-0.046	0.038	-0.028	0.010	0.055	0.013	-0.048	0.040		-0.044	-0.059	0.001	-0.032
	Avg_HRR95+	-0.019	0.018	-0.039	-0.007	0.031	0.007	-0.067	0.022	0.006	0.089	0.028	0.023	0.007	-0.218	-0.044		-0.133	0.087	0.088
	Avg_HRR85to95	-0.059	0.018	-0.086	-0.017	-0.116	0.000	-0.004	-0.066	-0.024	-0.046	-0.024	0.034	0.056	-0.020	-0.059	-0.133		0.064	0.050
	Avg_SprintEfforts	-0.119	-0.003	-0.121	-0.041	-0.067	-0.020	-0.039	-0.059	-0.036	-0.034	0.011	0.006	0.009	-0.049	0.001	0.087	0.064		0.174
	Sum_SprintEfforts	-0.096	-0.018	-0.127	-0.038	-0.070	-0.028	-0.014	-0.055	-0.009	-0.017	0.012	-0.024	0.020	-0.038	-0.032	0.088	0.050	0.174	

Extraction Method: Principal Component Analysis.

a. Reproduced communalities

b. Residuals are computed between observed and reproduced correlations. There are 36 (21.0%) nonredundant residuals with absolute values greater than 0.05.

Table 3.7 - Total Variance Explained									
	Initial Eigenvalues			Loadings			Rotation Sums of Squared Loadings		
Component	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	8.585	45.186	45.186	8.585	45.186	45.186	5.903	31.070	31.070
2	4.126	21.714	66.900	4.126	21.714	66.900	3.763	19.808	50.877
3	1.759	9.257	76.157	1.759	9.257	76.157	2.789	14.679	65.556
4	1.348	7.096	83.253	1.348	7.096	83.253	2.216	11.661	77.217
5	0.829	4.362	87.615	0.829	4.362	87.615	1.638	8.620	85.836
6	0.665	3.501	91.116	0.665	3.501	91.116	1.003	5.279	91.116
7	0.501	2.636	93.751						
8	0.347	1.827	95.578						
9	0.253	1.330	96.908						
10	0.210	1.103	98.011						
11	0.141	0.742	98.752						
12	0.102	0.536	99.288						
13	0.052	0.273	99.561						
14	0.040	0.213	99.774						
15	0.024	0.124	99.898						
16	0.009	0.050	99.948						
17	0.006	0.032	99.980						
18	0.003	0.015	99.994						
19	0.001	0.006	100.000						

Extraction Method: Principal Component Analysis.

Table 3.8 - KMO and Bartlett's Test		
Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		0.650
Bartlett's Test of Sphericity	Approx. Chi-Square	2617.615
	df	171
	Sig.	0.000

Table 3.9 - Rotated Component Matrix[a]	Component					
	1	2	3	4	5	6
Sum_PlayerLoad_min	0.933			0.307		
Sum_PlayerLoad	0.925					
Sum_of_Duration	0.899					
Sum_Distance	0.862	0.348				
Sum_MaxVel	0.846				0.337	
Sum_VelocityLoad	0.807	0.383				
Sum_SprintEfforts	0.760	0.573				
Avg_SprintEfforts		0.881				
Avg_MaxVel		0.846			0.336	
Avg_VelocityLoad		0.747	0.395	0.394		
Avg_Distance		0.735		0.471		
Avg_MeanHR			0.883			
Avg_HRR85t095		0.305	0.879			
Avg_TRIMP		0.378	0.753			
Avg_PlayerLoad_min				0.811		
Average_PlayerLoad		0.452	0.377	0.711		
Average_of_Duration					0.884	
Sum_Mean_HR2	0.566			-0.402	0.575	
Avg_HRR95+						0.938

Extraction Method: Principal Component Analysis.

a. Rotation converged in 7 iterations.

Table 3.10 Communalities		
	Initial	Extraction
Average_of_Duration	1.000	0.833
Sum_of_Duration	1.000	0.952
Average_PlayerLoad	1.000	0.908
Sum_PlayerLoad	1.000	0.980
Avg_PlayerLoad_min	1.000	0.873
Sum_PlayerLoad_min	1.000	0.984
Avg_TRIMP	1.000	0.856
Avg_Distance	1.000	0.940
Sum_Distance	1.000	0.952
Avg_MaxVel	1.000	0.856
Sum_MaxVel	1.000	0.920
Avg_VelocityLoad	1.000	0.909
Sum_VelocityLoad	1.000	0.920
Avg_MeanHR	1.000	0.861
Sum_Mean_HR2	1.000	0.827
Avg_HRR95+	1.000	0.990
Avg_HRR85t095	1.000	0.895
Avg_SprintEfforts	1.000	0.925
Sum_SprintEfforts	1.000	0.930

Extraction Method: Principal Component Analysis.

Table 3.11 – Scree Plot

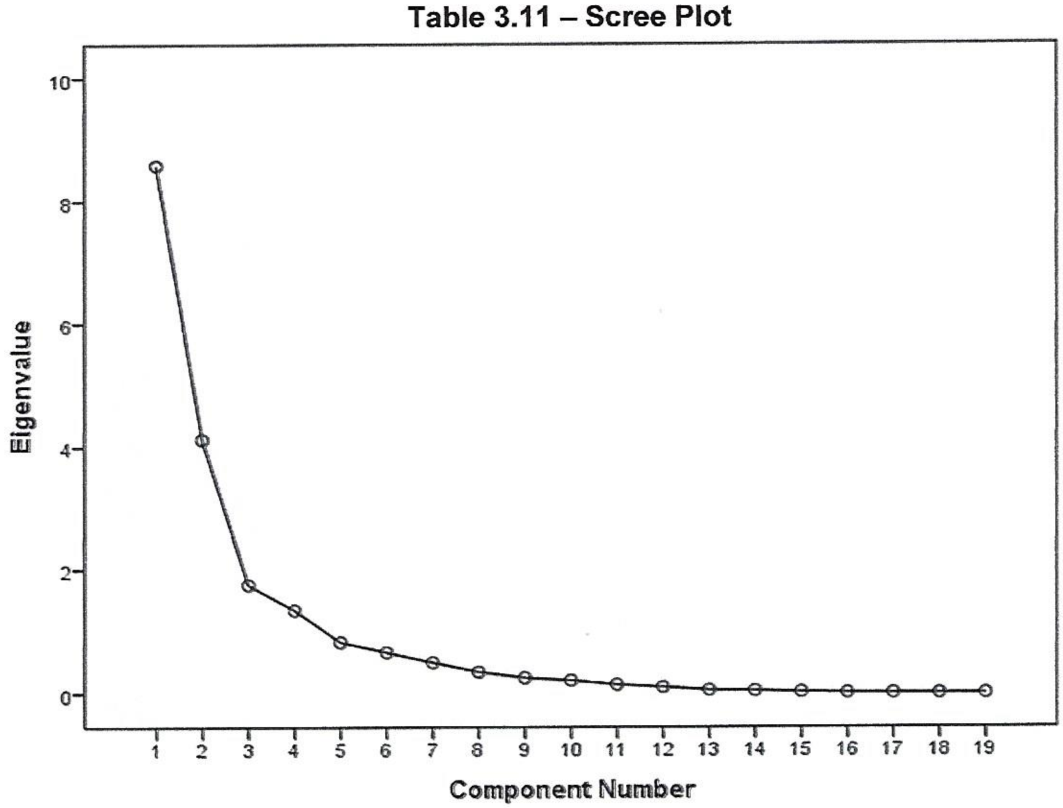


Table 3.12 - Reproduced Correlations

		Average_of_Duration	Sum_of_Duration	Average_PlayerLoad	Sum_PlayerLoad	Avg_PlayerLoad_min	Sum_PlayerLoad_min	Avg_TRIMP	Avg_Distance	Sum_Distance	Avg_MaxVel	Sum_MaxVel	Avg_VelocityLoad	Sum_VelocityLoad	Avg_MeanHR	Sum_Mean_HR2	Avg_HRR95+	Avg_HRR85to95	Avg_SprintEfforts	Sum_SprintEfforts
Reproduced Correlation	Average_of_Duration	.833[a]	0.372	0.226	0.223	-0.193	0.050	0.233	0.271	0.302	0.467	0.444	0.162	0.085	-0.053	0.568	-0.144	-0.009	0.119	0.093
	Sum_of_Duration	0.372	.952[a]	0.018	0.801	-0.100	0.759	-0.014	0.133	0.766	0.171	0.890	0.030	0.620	-0.270	0.731	-0.336	-0.099	0.131	0.610
	Average_PlayerLoad	0.226	0.018	.908[a]	0.480	0.740	0.419	0.678	0.848	0.524	0.513	0.145	0.805	0.568	0.521	-0.091	0.021	0.480	0.578	0.432
	Sum_PlayerLoad	0.223	0.801	0.480	.980[a]	0.411	0.961	0.291	0.531	0.941	0.299	0.784	0.437	0.894	0.049	0.453	-0.274	0.163	0.404	0.794
	Avg_PlayerLoad_min	-0.193	-0.100	0.740	0.411	.873[a]	0.456	0.466	0.585	0.361	0.108	-0.073	0.572	0.491	0.462	-0.343	0.191	0.349	0.327	0.303
	Sum_PlayerLoad_min	0.050	0.759	0.419	0.961	0.456	.984[a]	0.206	0.448	0.893	0.165	0.708	0.367	0.884	0.025	0.348	-0.255	0.119	0.337	0.769
	Avg_TRIMP	0.233	-0.014	0.678	0.291	0.466	0.206	.856[a]	0.666	0.353	0.463	0.189	0.702	0.404	0.741	0.134	0.298	0.802	0.569	0.362
	Avg_Distance	0.271	0.133	0.848	0.531	0.585	0.448	0.666	.940[a]	0.638	0.741	0.323	0.907	0.672	0.399	0.016	-0.057	0.497	0.815	0.651
	Sum_Distance	0.302	0.766	0.524	0.941	0.361	0.893	0.353	0.638	.952[a]	0.475	0.805	0.549	0.899	0.055	0.464	-0.295	0.216	0.552	0.850
	Avg_MaxVel	0.467	0.171	0.513	0.299	0.108	0.165	0.463	0.741	0.475	.856[a]	0.395	0.709	0.425	0.111	0.208	-0.111	0.315	0.776	0.553
	Sum_MaxVel	0.444	0.890	0.145	0.784	-0.073	0.708	0.189	0.323	0.805	0.395	.920[a]	0.242	0.673	-0.133	0.746	-0.246	0.104	0.368	0.719
	Avg_VelocityLoad	0.162	0.030	0.805	0.437	0.572	0.367	0.702	0.907	0.549	0.709	0.242	.909[a]	0.621	0.481	-0.049	0.019	0.594	0.838	0.623
	Sum_VelocityLoad	0.085	0.620	0.568	0.894	0.491	0.884	0.404	0.672	0.899	0.425	0.673	0.621	.920[a]	0.174	0.280	-0.224	0.327	0.616	0.867
	Avg_MeanHR	-0.053	-0.270	0.521	0.049	0.462	0.025	0.741	0.399	0.055	0.111	-0.133	0.481	0.174	.861[a]	-0.099	0.269	0.801	0.289	0.064
	Sum_Mean_HR2	0.568	0.731	-0.091	0.453	-0.343	0.348	0.134	0.016	0.464	0.208	0.746	-0.049	0.280	-0.099	.827[a]	-0.116	0.073	0.054	0.328
	Avg_HRR95+	-0.144	-0.336	0.021	-0.274	0.191	-0.255	0.298	-0.057	-0.295	-0.111	-0.246	0.019	-0.224	0.269	-0.116	.990[a]	0.302	0.001	-0.166
	Avg_HRR85to95	-0.009	-0.099	0.480	0.163	0.349	0.119	0.802	0.497	0.216	0.315	0.104	0.594	0.327	0.801	0.073	0.302	.895[a]	0.531	0.324
	Avg_SprintEfforts	0.119	0.131	0.578	0.404	0.327	0.337	0.569	0.815	0.552	0.776	0.368	0.838	0.616	0.289	0.054	0.001	0.531	.925[a]	0.740
	Sum_SprintEfforts	0.093	0.610	0.432	0.794	0.303	0.769	0.362	0.651	0.850	0.553	0.719	0.623	0.867	0.064	0.328	-0.166	0.324	0.740	.930[a]
Residual[b]	Average_of_Duration		0.022	-0.053	-0.005	0.005	0.004	-0.022	-0.065	-0.011	-0.086	-0.059	0.017	0.065	-0.018	-0.107	0.004	0.065	0.049	0.060
	Sum_of_Duration	0.022		0.005	0.016	-0.042	0.021	-0.002	-0.014	-0.004	0.001	-0.035	0.019	0.000	-0.005	-0.053	0.013	0.024	0.001	-0.014
	Average_PlayerLoad	-0.053	0.005		0.020	-0.043	0.001	0.011	0.001	-0.006	-0.016	-0.007	-0.007	-0.048	-0.035	0.036	-0.002	0.005	0.013	-0.002
	Sum_PlayerLoad	-0.005	0.016	0.020		-0.039	0.009	0.030	0.002	0.013	-0.005	-0.026	-0.010	-0.018	-0.022	-0.023	0.004	0.010	-0.001	-0.001
	Avg_PlayerLoad_min	0.005	-0.042	-0.043	-0.039		-0.016	-0.058	-0.015	-0.041	0.022	0.058	-0.006	0.008	0.040	0.077	-0.022	-0.019	0.025	0.014
	Sum_PlayerLoad_min	0.004	0.021	0.001	0.009	-0.016		0.000	-0.008	-0.005	0.016	-0.015	0.005	-0.001	-0.003	-0.022	0.005	0.013	-0.004	-0.013
	Avg_TRIMP	-0.022	-0.002	0.011	0.030	-0.058	0.000		0.022	0.048	-0.016	-0.026	-0.061	-0.035	-0.070	-0.050	-0.010	-0.007	-0.011	0.013
	Avg_Distance	-0.065	-0.014	0.001	0.002	-0.015	-0.008	0.022		0.028	0.016	0.017	-0.008	-0.024	0.005	0.036	0.001	-0.031	-0.027	-0.026
	Sum_Distance	-0.011	-0.004	-0.006	0.013	-0.041	-0.005	0.048	0.028		0.000	-0.017	-0.028	-0.012	-0.007	-0.029	0.006	-0.011	-0.021	0.005
	Avg_MaxVel	-0.086	0.001	-0.016	-0.005	0.022	0.016	-0.016	0.016	0.000		0.056	-0.024	-0.007	0.066	0.016	0.008	-0.038	-0.069	-0.052
	Sum_MaxVel	-0.059	-0.035	-0.007	-0.026	0.058	-0.015	-0.026	0.017	-0.017	0.056		-0.008	0.003	0.039	0.059	-0.008	-0.037	-0.025	-0.022
	Avg_VelocityLoad	0.017	0.019	-0.007	-0.010	-0.006	0.005	-0.061	-0.008	-0.028	-0.024	-0.008		0.043	-0.008	0.015	0.010	0.029	-0.007	-0.037
	Sum_VelocityLoad	0.065	0.000	-0.048	-0.018	0.008	-0.001	-0.035	-0.024	-0.012	-0.007	0.003	0.043		0.004	-0.046	0.010	0.031	-0.021	-0.007
	Avg_MeanHR	-0.018	-0.005	-0.035	-0.022	0.040	-0.003	-0.070	0.005	-0.007	0.066	0.039	-0.008	0.004		0.028	0.013	-0.088	-0.003	0.007
	Sum_Mean_HR2	-0.107	-0.053	0.036	-0.023	0.077	-0.022	-0.050	0.036	-0.029	0.016	0.059	0.015	-0.046	0.028		-0.020	-0.058	0.016	-0.019
	Avg_HRR95+	0.004	0.013	-0.002	0.004	-0.022	0.005	-0.010	0.001	0.006	0.008	-0.008	0.010	0.010	0.013	-0.020		0.001	-0.011	-0.008
	Avg_HRR85to95	0.065	0.024	0.005	0.010	-0.019	0.013	-0.007	-0.031	-0.011	-0.038	-0.037	0.029	0.031	-0.088	-0.058	0.001		-0.001	-0.009
	Avg_SprintEfforts	0.049	0.001	0.013	-0.001	0.025	-0.004	-0.011	-0.027	-0.021	-0.069	-0.025	-0.007	-0.021	-0.003	0.016	-0.011	-0.001		0.046
	Sum_SprintEfforts	0.060	-0.014	-0.002	-0.001	0.014	-0.013	0.013	-0.026	0.005	-0.052	-0.022	-0.037	-0.007	0.007	-0.019	-0.008	-0.009	0.046	

Extraction Method: Principal Component Analysis.

a. Reproduced communalities

b. Residuals are computed between observed and reproduced correlations. There are 22 (12.0%) nonredundant residuals with absolute values greater than 0.05.

Table 3.13 - Total Variance Explained

	Initial Eigenvalues			Loadings			Rotation Sums of Squared Loadings		
Component	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	8.490	49.940	49.940	8.490	49.940	49.940	5.970	35.116	35.116
2	3.897	22.925	72.865	3.897	22.925	72.865	3.948	23.224	58.340
3	1.449	8.525	81.389	1.449	8.525	81.389	3.129	18.408	76.748
4	1.205	7.086	88.475	1.205	7.086	88.475	1.994	11.727	88.475
5	0.645	3.794	92.269						
6	0.400	2.356	94.625						
7	0.266	1.565	96.190						
8	0.225	1.325	97.515						
9	0.157	0.921	98.436						
10	0.104	0.614	99.051						
11	0.056	0.330	99.381						
12	0.047	0.274	99.655						
13	0.024	0.140	99.794						
14	0.018	0.106	99.900						
15	0.009	0.052	99.952						
16	0.006	0.036	99.989						
17	0.002	0.011	100.000						

Extraction Method: Principal Component Analysis.

Table 3.14 - KMO and Bartlett's Test

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		0.759
Bartlett's Test of Sphericity	Bartlett's Test of	2396.497
		136
		0.000

Table 3.15 - Rotated Component Matrix[a]

	Component 1	2	3	4
Sum_PlayerLoad_min	0.970			
Sum_PlayerLoad	0.963			
Sum_Distance	0.889	0.383		
Sum_of_Duration	0.872			-0.401
Sum_VelocityLoad	0.837	0.376		
Sum_MaxVel	0.791			-0.501
Sum_SprintEfforts	0.734	0.535		
Avg_MaxVel		0.897		
Avg_SprintEfforts		0.847		
Avg_Distance	0.334	0.788	0.364	
Avg_VelocityLoad		0.764	0.438	
Average_PlayerLoad		0.529	0.489	0.428
Avg_MeanHR			0.898	
Avg_HRR85t095			0.866	
Avg_TRIMP		0.404	0.815	
Sum_Mean_HR2	0.485			-0.770
Avg_PlayerLoad_min	0.305		0.409	0.738

Extraction Method: Principal Component Analysis.

a. Rotation converged in 7 iterations.

Table 3.16 - Communalities

	Initial	Extraction
Sum_of_Duration	1.000	0.944
Average_PlayerLoad	1.000	0.791
Sum_PlayerLoad	1.000	0.976
Avg_PlayerLoad_min	1.000	0.833
Sum_PlayerLoad_min	1.000	0.979
Avg_TRIMP	1.000	0.844
Avg_Distance	1.000	0.922
Sum_Distance	1.000	0.948
Avg_MaxVel	1.000	0.871
Sum_MaxVel	1.000	0.934
Avg_VelocityLoad	1.000	0.908
Sum_VelocityLoad	1.000	0.907
Avg_MeanHR	1.000	0.844
Sum_Mean_HR2	1.000	0.838
Avg_HRR85t095	1.000	0.822
Avg_SprintEfforts	1.000	0.847
Sum_SprintEfforts	1.000	0.832

Extraction Method: Principal Component Analysis.

Table 3.17 – Scree Plot

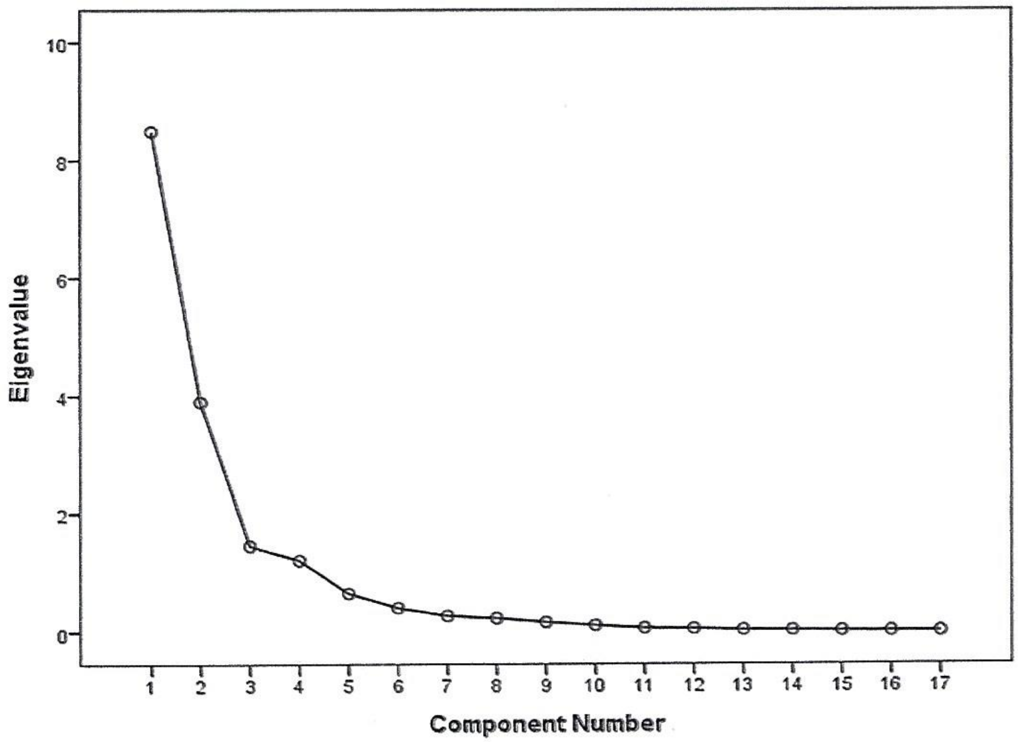


Table 3.18 - Reproduced Correlations

		Duration	PlayerLoa	erLoad	erLoad_m	erLoad_m	MP	ance	ance	Vel	Vel	cityLoad	cityLoad	nHR	n_HR2	85t095	tEfforts	ntEfforts
Reproduced Correlation	Sum_of_Duration	.944ᵃ	0.004	0.801	-0.095	0.769	-0.023	0.129	0.764	0.162	0.887	0.028	0.626	-0.269	0.718	-0.095	0.138	0.623
	Average_PlayerLoad	0.004	.791ᵃ	0.457	0.695	0.432	0.656	0.796	0.506	0.475	0.146	0.806	0.613	0.527	-0.143	0.570	0.672	0.539
	Sum_PlayerLoad	0.801	0.457	.976ᵃ	0.404	0.964	0.287	0.519	0.937	0.289	0.782	0.438	0.904	0.046	0.440	0.181	0.423	0.816
	Avg_PlayerLoad_min	-0.095	0.695	0.404	.833ᵃ	0.458	0.449	0.558	0.359	0.081	-0.089	0.580	0.523	0.476	-0.383	0.400	0.365	0.342
	Sum_PlayerLoad_min	0.769	0.432	0.964	0.458	.979ᵃ	0.211	0.449	0.895	0.166	0.703	0.371	0.883	0.018	0.349	0.114	0.329	0.759
	Avg_TRIMP	-0.023	0.656	0.287	0.449	0.211	.844ᵃ	0.661	0.353	0.459	0.191	0.701	0.413	0.756	0.127	0.818	0.586	0.380
	Avg_Distance	0.129	0.796	0.519	0.558	0.449	0.661	.922ᵃ	0.630	0.738	0.334	0.907	0.685	0.402	0.009	0.535	0.857	0.696
	Sum_Distance	0.764	0.506	0.937	0.359	0.895	0.353	0.630	.948ᵃ	0.471	0.807	0.549	0.903	0.052	0.458	0.227	0.566	0.867
	Avg_MaxVel	0.162	0.475	0.289	0.081	0.166	0.459	0.738	0.471	.871ᵃ	0.415	0.706	0.426	0.119	0.221	0.334	0.805	0.582
	Sum_MaxVel	0.887	0.146	0.782	-0.089	0.703	0.191	0.334	0.807	0.415	.934ᵃ	0.241	0.664	-0.123	0.768	0.101	0.367	0.713
	Avg_VelocityLoad	0.028	0.806	0.438	0.580	0.371	0.701	0.907	0.549	0.706	0.241	.908ᵃ	0.620	0.478	-0.052	0.589	0.835	0.623
	Sum_VelocityLoad	0.626	0.613	0.904	0.523	0.883	0.413	0.685	0.903	0.426	0.664	0.620	.907ᵃ	0.161	0.287	0.292	0.580	0.830
	Avg_MeanHR	-0.269	0.527	0.046	0.476	0.018	0.756	0.402	0.052	0.119	-0.123	0.478	0.161	.844ᵃ	-0.077	0.793	0.291	0.062
	Sum_Mean_HR2	0.718	-0.143	0.440	-0.383	0.349	0.127	0.009	0.458	0.221	0.768	-0.052	0.287	-0.077	.838ᵃ	0.110	0.094	0.368
	Avg_HRR85t095	-0.095	0.570	0.181	0.400	0.114	0.818	0.535	0.227	0.334	0.101	0.589	0.292	0.793	0.110	.822ᵃ	0.459	0.246
	Avg_SprintEfforts	0.138	0.672	0.423	0.365	0.329	0.586	0.857	0.566	0.805	0.367	0.835	0.580	0.291	0.094	0.459	.847ᵃ	0.653
	Sum_SprintEfforts	0.623	0.539	0.816	0.342	0.759	0.380	0.696	0.867	0.582	0.713	0.623	0.830	0.062	0.368	0.246	0.653	.832ᵃ
Residualᵇ	Sum_of_Duration		0.018	0.016	-0.046	0.012	0.007	-0.009	-0.002	0.009	-0.033	0.021	-0.006	-0.006	-0.040	0.021	-0.006	-0.027
	Average_PlayerLoad	0.018		0.043	0.002	-0.012	0.034	0.052	0.011	0.021	-0.007	-0.008	-0.093	-0.041	0.088	-0.086	-0.081	-0.109
	Sum_PlayerLoad	0.016	0.043		-0.032	0.006	0.033	0.014	0.017	0.005	-0.024	-0.011	-0.028	-0.020	-0.009	-0.008	-0.020	-0.023
	Avg_PlayerLoad_min	-0.046	0.002	-0.032		-0.017	-0.041	0.012	-0.039	0.049	0.075	-0.015	-0.025	0.026	0.118	-0.070	-0.012	-0.025
	Sum_PlayerLoad_min	0.012	-0.012	0.006	-0.017		-0.005	-0.009	-0.007	0.015	-0.011	0.001	0.001	0.005	-0.022	0.018	0.003	-0.003
	Avg_TRIMP	0.007	0.034	0.033	-0.041	-0.005		0.027	0.048	-0.012	-0.029	-0.060	-0.045	-0.085	-0.043	-0.023	-0.028	-0.006
	Avg_Distance	-0.009	0.052	0.014	0.012	-0.009	0.027		0.036	0.019	0.007	-0.009	-0.037	0.002	0.044	-0.069	-0.070	-0.071
	Sum_Distance	-0.002	0.011	0.017	-0.039	-0.007	0.048	0.036		0.004	-0.020	-0.027	-0.016	-0.003	-0.024	-0.023	-0.035	-0.011
	Avg_MaxVel	0.009	0.021	0.005	0.049	0.015	-0.012	0.019	0.004		0.035	-0.022	-0.008	0.058	0.003	-0.057	-0.098	-0.081
	Sum_MaxVel	-0.033	-0.007	-0.024	0.075	-0.011	-0.029	0.007	-0.020	0.035		-0.006	0.012	0.028	0.036	-0.034	-0.024	-0.017
	Avg_VelocityLoad	0.021	-0.008	-0.011	-0.015	0.001	-0.060	-0.009	-0.027	-0.022	-0.006		0.043	-0.004	0.018	0.034	-0.005	-0.037
	Sum_VelocityLoad	-0.006	-0.093	-0.028	-0.025	0.001	-0.045	-0.037	-0.016	-0.008	0.012	0.043		0.017	-0.052	0.066	0.016	0.030
	Avg_MeanHR	-0.006	-0.041	-0.020	0.026	0.005	-0.085	0.002	-0.003	0.058	0.028	-0.004	0.017		0.006	-0.079	-0.004	0.009
	Sum_Mean_HR2	-0.040	0.088	-0.009	0.118	-0.022	-0.043	0.044	-0.024	0.003	0.036	0.018	-0.052	0.006		-0.094	-0.024	-0.059
	Avg_HRR85t095	0.021	-0.086	-0.008	-0.070	0.018	-0.023	-0.069	-0.023	-0.057	-0.034	0.034	0.066	-0.079	-0.094		0.071	0.069
	Avg_SprintEfforts	-0.006	-0.081	-0.020	-0.012	0.003	-0.028	-0.070	-0.035	-0.098	-0.024	-0.005	0.016	-0.004	-0.024	0.071		0.133
	Sum_SprintEfforts	-0.027	-0.109	-0.023	-0.025	-0.003	-0.006	-0.071	-0.011	-0.081	-0.017	-0.037	0.030	0.009	-0.059	0.069	0.133	

Extraction Method: Principal Component Analysis.

a. Reproduced communalities

b. Residuals are computed between observed and reproduced correlations. There are 26 (19.0%) nonredundant residuals with absolute values greater than 0.05.

Appendix D

Table 3.20 - Model Summary[d]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics: R Square Change	F Change	df1	df2	Sig. F Change	Durbin-Watson
1	.482[a]	0.232	0.222	34.80437	0.232	22.646	1	75	0.000	
2	.549[b]	0.301	0.282	33.41783	0.069	7.353	1	74	0.008	
3	.588[c]	0.346	0.319	32.56459	0.044	4.929	1	73	0.030	1.906

a. Predictors: (Constant), Phenylalanine
b. Predictors: (Constant), Phenylalanine, Ethanolamine
c. Predictors: (Constant), Phenylalanine, Ethanolamine, Beta_Amino_Butyric_Acid
d. Dependent Variable: Sum_PlayerLoad_min

Table 3.21 - ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	27432.381	1	27432.381	22.646	.000[b]
	Residual	90850.796	75	1211.344		
	Total	118283.177	76			
2	Regression	35643.559	2	17821.779	15.959	.000[c]
	Residual	82639.618	74	1116.752		
	Total	118283.177	76			
3	Regression	40870.121	3	13623.374	12.847	.000[d]
	Residual	77413.056	73	1060.453		
	Total	118283.177	76			

a. Dependent Variable: Sum_PlayerLoad_min
b. Predictors: (Constant), Phenylalanine
c. Predictors: (Constant), Phenylalanine, Ethanolamine
d. Predictors: (Constant), Phenylalanine, Ethanolamine, Beta_Amino_Butyric_Acid

Table 3.22 – Scatterplot

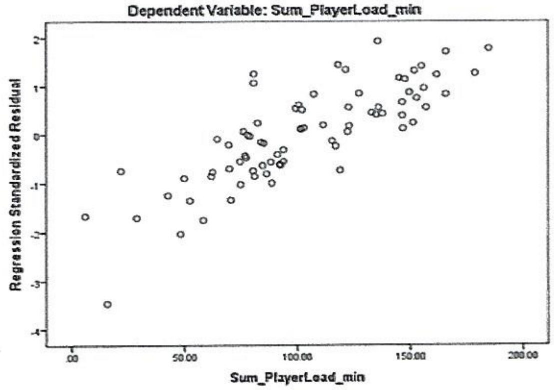


Table 3.23 - Coefficients[a]

Model		Unstandardized Coefficients B	Std. Error	Standardized Coefficients Beta	t	Sig.	95.0% Confidence Interval for B Lower Bound	Upper Bound	Correlations Zero-order	Partial	Part
1	(Constant)	226.378	26.320		8.601	0.000	173.946	278.810			
	Phenylalanine	-1.609	0.338	-0.482	-4.759	0.000	-2.283	-0.936	-0.482	-0.482	-0.482
2	(Constant)	199.106	27.199		7.320	0.000	144.910	253.302			
	Phenylalanine	-1.613	0.325	-0.483	-4.966	0.000	-2.260	-0.966	-0.482	-0.500	-0.483
	Ethanolamine	4.659	1.718	0.263	2.712	0.008	1.235	8.082	0.262	0.301	0.263
3	(Constant)	179.528	27.933		6.427	0.000	123.857	235.199			
	Phenylalanine	-1.795	0.327	-0.537	-5.491	0.000	-2.447	-1.144	-0.482	-0.541	-0.520
	Ethanolamine	4.161	1.689	0.235	2.463	0.016	0.794	7.527	0.262	0.277	0.233
	Beta_Amino_Butyric_Acid	16.568	7.463	0.219	2.220	0.030	1.694	31.442	0.115	0.251	0.210

a. Dependent Variable: Sum_PlayerLoad_min

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	R Square Change	F Change	df1	df2	Sig. F Change	Durbin-Watson
					Change Statistics					
1	.424[a]	0.179	0.168	0.80358	0.179	16.182	1	74	0.000	
2	.557[b]	0.310	0.291	0.74188	0.131	13.821	1	73	0.000	
3	.627[c]	0.394	0.368	0.70037	0.083	9.909	1	72	0.002	
4	.675[d]	0.456	0.425	0.66801	0.062	8.145	1	71	0.006	
5	.707[e]	0.500	0.465	0.64480	0.044	6.203	1	70	0.015	1.707

Table 3.24 - Model Summary[f]

a. Predictors: (Constant), Arginine
b. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid
c. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid, Taurine
d. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid, Taurine, Asparagine
e. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid, Taurine, Asparagine, Histidine
f. Dependent Variable: Avg_MaxVel

Table 3.25 - ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	10.449	1	10.449	16.182	.000[b]
	Residual	47.785	74	0.646		
	Total	58.234	75			
2	Regression	18.056	2	9.028	16.403	.000[c]
	Residual	40.178	73	0.550		
	Total	58.234	75			
3	Regression	22.917	3	7.639	15.573	.000[d]
	Residual	35.317	72	0.491		
	Total	58.234	75			
4	Regression	26.551	4	6.638	14.875	.000[e]
	Residual	31.683	71	0.446		
	Total	58.234	75			
5	Regression	29.130	5	5.826	14.013	.000[f]
	Residual	29.104	70	0.416		
	Total	58.234	75			

a. Dependent Variable: Avg_MaxVel
b. Predictors: (Constant), Arginine
c. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid
d. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid, Taurine
e. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid, Taurine, Asparagine
f. Predictors: (Constant), Arginine, Alpha_Amino_Adipic_Acid, Taurine, Asparagine,

Table 3.26 – Scatterplot

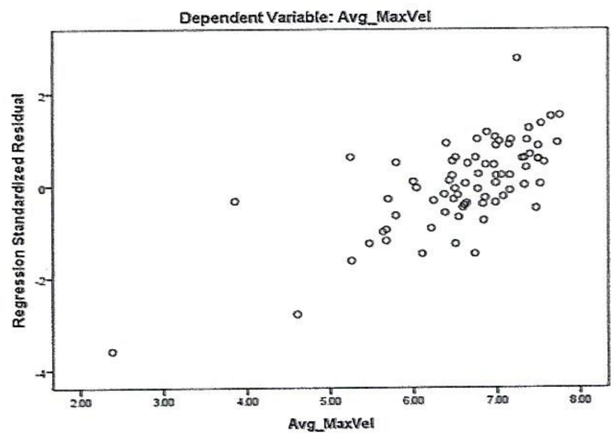


Table 3.27 - Coefficients[a]

Model		Unstandardized Coefficients B	Unstandardized Coefficients Std. Error	Standardized Coefficients Beta	t	Sig.	95.0% Confidence Interval for B Lower Bound	95.0% Confidence Interval for B Upper Bound	Correlations Zero-order	Correlations Partial	Correlations Part
1	(Constant)	4.999	0.406		12.322	0.000	4.191	5.808			
	Arginine	0.020	0.005	0.424	4.023	0.000	0.010	0.030	0.424	0.424	0.424
2	(Constant)	5.933	0.451		13.157	0.000	5.034	6.831			
	Arginine	0.019	0.005	0.405	4.157	0.000	0.010	0.028	0.424	0.438	0.404
	Alpha_Amino_Adipic_Acid	-0.791	0.213	-0.362	-3.718	0.000	-1.215	-0.367	-0.383	-0.399	-0.361
3	(Constant)	6.826	0.512		13.342	0.000	5.806	7.846			
	Arginine	0.016	0.004	0.334	3.527	0.001	0.007	0.025	0.424	0.384	0.324
	Alpha_Amino_Adipic_Acid	-0.820	0.201	-0.375	-4.081	0.000	-1.221	-0.420	-0.383	-0.433	-0.375
	Taurine	-0.018	0.006	-0.298	-3.148	0.002	-0.029	-0.007	-0.364	-0.348	-0.289
4	(Constant)	5.884	0.589		9.990	0.000	4.710	7.059			
	Arginine	0.011	0.005	0.241	2.514	0.014	0.002	0.021	0.424	0.286	0.220
	Alpha_Amino_Adipic_Acid	-0.654	0.200	-0.299	-3.261	0.002	-1.053	-0.254	-0.383	-0.361	-0.285
	Taurine	-0.021	0.006	-0.349	-3.796	0.000	-0.032	-0.010	-0.364	-0.411	-0.332
	Asparagin	0.030	0.011	0.280	2.854	0.006	0.009	0.051	0.401	0.321	0.250
5	(Constant)	7.000	0.724		9.671	0.000	5.556	8.443			
	Arginine	0.012	0.004	0.248	2.681	0.009	0.003	0.021	0.424	0.305	0.227
	Alpha_Amino_Adipic_Acid	-0.561	0.197	-0.257	-2.847	0.006	-0.954	-0.168	-0.383	-0.322	-0.241
	Taurine	-0.019	0.005	-0.313	-3.479	0.001	-0.030	-0.008	-0.364	-0.384	-0.294
	Asparagin	0.046	0.012	0.428	3.829	0.000	0.022	0.070	0.401	0.416	0.324
	Histidine	-0.023	0.009	-0.259	-2.491	0.015	-0.042	-0.005	-0.043	-0.285	-0.210

a. Dependent Variable: Avg_MaxVel

Table 3.28 - Model Summary[f]

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate	Change Statistics: R Square Change	F Change	df1	df2	Sig. F Change	Durbin-Watson
1	.412[a]	0.170	0.158	9.448	0.170	15.115	1	74	0.000	
2	.481[b]	0.232	0.211	9.151	0.062	5.888	1	73	0.018	
3	.538[c]	0.289	0.260	8.860	0.058	5.862	1	72	0.018	
4	.587[d]	0.345	0.308	8.569	0.055	5.988	1	71	0.017	
5	.633[e]	0.401	0.358	8.249	0.056	6.601	1	70	0.012	2.007

a. Predictors: (Constant), Ornithine

b. Predictors: (Constant), Ornithine, Threonine

c. Predictors: (Constant), Ornithine, Threonine, Glycine

d. Predictors: (Constant), Ornithine, Threonine, Glycine, Histidine

e. Predictors: (Constant), Ornithine, Threonine, Glycine, Histidine, Arginine

f. Dependent Variable: Avg_MeanHR

Table 3.29 - ANOVA[a]

Model		Sum of Squares	df	Mean Square	F	Sig.
1	Regression	1349.278	1	1349.278	15.115	.000[b]
	Residual	6605.710	74	89.266		
	Total	7954.988	75			
2	Regression	1842.293	2	921.146	11.001	.000[c]
	Residual	6112.695	73	83.736		
	Total	7954.988	75			
3	Regression	2302.466	3	767.489	9.776	.000[d]
	Residual	5652.521	72	78.507		
	Total	7954.988	75			
4	Regression	2742.136	4	685.534	9.337	.000[e]
	Residual	5212.852	71	73.420		
	Total	7954.988	75			
5	Regression	3191.346	5	638.269	9.379	.000[f]
	Residual	4763.642	70	68.052		
	Total	7954.988	75			

a. Dependent Variable: Avg_MeanHR

b. Predictors: (Constant), Ornithine

c. Predictors: (Constant), Ornithine, Threonine

d. Predictors: (Constant), Ornithine, Threonine, Glycine

e. Predictors: (Constant), Ornithine, Threonine, Glycine, Histidine

f. Predictors: (Constant), Ornithine, Threonine, Glycine, Histidine, Arginine

Table 3.30 – Scatterplot

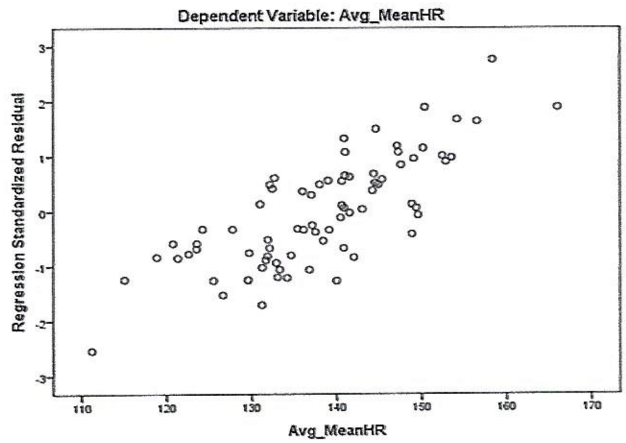


Table 3.31 - Coefficients[a]

Model		Unstandardized Coefficients: B	Std. Error	Standardized Coefficients: Beta	t	Sig.	95.0% Confidence Interval for B: Lower Bound	Upper Bound	Correlations: Zero-order	Partial	Part
1	(Constant)	155.058	4.449		34.855	0.000	146.194	163.922			
	Ornithine	-0.227	0.058	-0.412	-3.888	0.000	-0.344	-0.111	-0.412	-0.412	-0.412
2	(Constant)	142.163	6.841		20.780	0.000	128.528	155.798			
	Ornithine	-0.265	0.059	-0.480	-4.510	0.000	-0.381	-0.148	-0.412	-0.467	-0.463
	Threonine	0.122	0.050	0.258	2.426	0.018	0.022	0.222	0.132	0.273	0.249
3	(Constant)	155.843	8.707		17.899	0.000	138.486	173.200			
	Ornithine	-0.258	0.057	-0.467	-4.528	0.000	-0.371	-0.144	-0.412	-0.471	-0.450
	Threonine	0.161	0.051	0.340	3.136	0.002	0.058	0.263	0.132	0.347	0.312
	Glycine	-0.077	0.032	-0.255	-2.421	0.018	-0.141	-0.014	-0.205	-0.274	-0.241
4	(Constant)	173.040	10.967		15.778	0.000	151.172	194.908			
	Ornithine	-0.247	0.055	-0.449	-4.488	0.000	-0.357	-0.138	-0.412	-0.470	-0.431
	Threonine	0.202	0.052	0.427	3.859	0.000	0.098	0.306	0.132	0.416	0.371
	Glycine	-0.082	0.031	-0.269	-2.635	0.010	-0.143	-0.020	-0.205	-0.298	-0.253
	Histidine	-0.261	0.107	-0.252	-2.447	0.017	-0.474	-0.048	-0.192	-0.279	-0.235
5	(Constant)	170.327	10.611		16.051	0.000	149.163	191.491			
	Ornithine	-0.153	0.065	-0.277	-2.367	0.021	-0.282	-0.024	-0.412	-0.272	-0.219
	Threonine	0.129	0.058	0.273	2.229	0.029	0.014	0.244	0.132	0.257	0.206
	Glycine	-0.114	0.032	-0.377	-3.525	0.001	-0.179	-0.050	-0.205	-0.388	-0.326
	Histidine	-0.282	0.103	-0.271	-2.730	0.008	-0.487	-0.076	-0.192	-0.310	-0.252
	Arginine	0.189	0.073	0.339	2.569	0.012	0.042	0.335	0.358	0.294	0.238

a. Dependent Variable: Avg_MeanHR

Appendix E

Table 3.32 - Descriptive Statistics

	Mean	Std. Deviation	N
PS_SARCOSINE_S	5.67	4.509	3
W1_SARCOSINE_S	7.67	2.082	3
W4_SARCOSINE_S	9.67	4.041	3
W8_SARCOSINE_S	1.00	0.000	3
W12_SARCOSINE_S	9.67	4.041	3
PS_SARCOSINE_NS	13.67	4.041	3
W1_SARCOSINE_NS	7.33	5.132	3
W4_SARCOSINE_NS	9.00	1.732	3
W8_SARCOSINE_NS	1.00	0.000	3
W12_SARCOSINE_NS	8.33	1.528	3

Table 3.33 - Sarcosine Tests of Within-Subjects Effects

Measure:

Source		Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power[a]
group	Sphericity Assumed	9.633	1	9.633	289.000	0.003	0.993	289.000	1.000
	Greenhouse-Geisser	9.633	1.000	9.633	289.000	0.003	0.993	289.000	1.000
	Huynh-Feldt	9.633	1.000	9.633	289.000	0.003	0.993	289.000	1.000
	Lower-bound	9.633	1.000	9.633	289.000	0.003	0.993	289.000	1.000
Error(group)	Sphericity Assumed	0.067	2	0.033					
	Greenhouse-Geisser	0.067	2.000	0.033					
	Huynh-Feldt	0.067	2.000	0.033					
	Lower-bound	0.067	2.000	0.033					
time	Sphericity Assumed	314.133	4	78.533	4.301	0.038	0.683	17.205	0.702
	Greenhouse-Geisser	314.133	1.146	274.115	4.301	0.160	0.683	4.929	0.257
	Huynh-Feldt	314.133	1.684	186.564	4.301	0.119	0.683	7.242	0.358
	Lower-bound	314.133	1.000	314.133	4.301	0.174	0.683	4.301	0.230
Error(time)	Sphericity Assumed	146.067	8	18.258					
	Greenhouse-Geisser	146.067	2.292	63.729					
	Huynh-Feldt	146.067	3.368	43.374					
	Lower-bound	146.067	2.000	73.033					
group * time	Sphericity Assumed	89.867	4	22.467	2.902	0.093	0.592	11.608	0.517
	Greenhouse-Geisser	89.867	1.844	48.736	2.902	0.175	0.592	5.351	0.280
	Huynh-Feldt	89.867	4.000	22.467	2.902	0.093	0.592	11.608	0.517
	Lower-bound	89.867	1.000	89.867	2.902	0.231	0.592	2.902	0.175
Error(group*time)	Sphericity Assumed	61.933	8	7.742					
	Greenhouse-Geisser	61.933	3.688	16.794					
	Huynh-Feldt	61.933	8.000	7.742					
	Lower-bound	61.933	2.000	30.967					

a. Computed using alpha = .05

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