

**OVER-TRAINING SYNDROME A STUDY TO  
DETERMINE THE CORRELATION BETWEEN  
THE PHYSIOLOGICAL SYMPTOMS AND THE  
PSYCHOLOGICAL SIGNS IN COLLEGE  
WRESTLERS**

**By**

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## **CHAPTER I**

### **INTRODUCTION**

As social animals, humans feel the need to be part of society. Social acceptance is a condition in today's world. "Looking good" and "succeeding" may be perceived by some individuals as those attributes which may garner that social acceptance and self-respect (Berger, Pargman & Weinberg, 2002). Society is filled with individuals who may share advice and attempt to influence others on how to look good. They may also tout the best pathways to achieve these attributes. Advice may range from eating at the fast food restaurant "Subway®" so the American public can lose weight like the ubiquitous Jared, whose diet consisted of Subway® sandwiches and an exercise program of walking, to the kind of exercise facility at which to work out, and how long the workouts should be. Being fit and trim is important not only for appearance reasons, but for health reasons as well (Quinn, 2004).

For athletes, the need to be in good condition achieves a level of importance that cannot be matched by non-athletes. The nature of their sports and the amount of physical conditioning that is necessary in order to compete at the desired level requires exceptional motivation. Athletes by their very nature are extremely motivated if they wish to succeed in the elite arena of competition. Participation in athletics promises many rewards. Those rewards include financial stability, fame, and credibility. In order to

achieve good conditioning and prepare for competition, athletes train very hard. However, the relationship between the amount of training necessary and the amount of training undertaken is often out of balance, and is most often misunderstood. Signs of stress and needed recovery time may be ignored. This break in recognition or denial of the required-undertaken connection of rest and recovery results in over-training (Hines, 2001). While training is good, over-training and not allowing the body to recover is bad. In some sports, the consequences of over-training can be devastating and can lead to permanent injuries or death (Viscardi, 2004; Luttermoser, Gochenour, & Shaughnessy, 1999).

Wrestling is one such sport that conditions the athletes to work hard and ignore signs of stress and lack of recovery. The athletes who yield to these signs are often labeled as weak or unable to muster the strength and resolve to train hard. The combination of skills required to be competitive in wrestling is manifest in the type of training and motivation to train at the level of competition. The confounding requirements to achieve the right balance of speed, flexibility, suppleness, and strength add to the difficulty in identifying optimal training methods. Wrestling is not financially rewarding in terms of a career choice. Receiving accolades and glory at the collegiate and international level may be the highest level of achievement experienced by the athlete. Wrestlers typically struggle to maintain a balance between their weight and the amount of training required to compete effectively. They often believe that they will maintain an advantage of superior ability and strength if they are able to compete at a lower weight. This belief invariably leads to over-training by failing to providing the body the necessary elements to allow complete recovery between workouts (Hines, 2001).

Unfortunately, the amount of literature relating to college wrestling resulting from clinical trials and other scientific research is sparse. Training in college wrestling and other sports combines or exclusively uses endurance, speed, or strength to place physical physiological, biochemical and psychological stresses on the body. Wrestlers and those who train for long periods of time expend a lot of physical energy. Surprisingly, these athletes are more susceptible to illnesses that stress the immune system (Mackinnon, 1999).

The breakdown in immunity resulting in associated susceptibility to upper respiratory tract infections will be discussed in detail as part of the physiological and immunological signs of over-training syndrome. A comprehensive literature review will be presented discussing different response aspects of over-training. Cortisol, an immunosuppressant that is secreted by the adrenal glands in response to physical stresses, plays a major role in the over-training syndrome. Testing for the chemical cortisol is one type of a physiological marker commonly utilized in physiological research.

A variety of tests exists which help point to psychological markers which identify the underpinnings of behavior. These tests include the Eating Attitudes Test (EAT) (Garner & Garfinkel, 1979), Eating Disorder Inventory (EDI) (Garner, 1991), Profile of Mood States (POMS) (McNair, Lorr & Droppleman, 1971), Recovery-Stress Questionnaire for Athletes (RESTQ-Sport) Kellman & Kallus, ), Positive Affect-Negative Affect Scale (PANAS) (Watson, Clark & Tellegen, 1985), and Exercise-Induced Feeling Inventory (EFI) (Gauvin & Rejeski, 1993). Each of these tests can help establish psychological markers, and thereby identify behavioral underpinnings that are associated with over-training in athletes.

### ***Statement of the Problem***

The problem was to establish a correlation between the mind and body, (i.e., cortisol levels, and changes possible in RESTQ-Sport score). There has been a tremendous amount of research completed regarding over-training syndrome. The population samples have typically included distance athletes, (e.g., marathoners, triathletes, swimmers), with some investigations focusing on populations of individuals involved in team sports, primarily basketball and soccer (Hoffman, Epstein, Yarom, Zigel & Einbinder, 1999; Lehmann, Schnee, Scheu, Stockhausen, & Bachl, 1992).

In order to compete in wrestling, the athlete must compete within weight class restrictions. To achieve the weight for competition, the athlete restricts caloric and fluid intake in combination with training sessions designed to burn calories and dehydrate the body in order to maintain the weight requirement preparing for competition. These weight restrictions may not allow the athlete to adequately recover from workout and competition.

Research has attempted to demonstrate over-training syndrome in athletes who train for long periods of time, and in athletes who compete for long durations. While athletes who compete in wrestling train for extended periods of time, their actual duration of competition time is relatively short by comparison. Athletes, coaches, athletic trainers, and other allied health professionals need to understand warning signs of over-training in order to help prevent the over-training syndrome. Further research in this arena specifically addressing the training regimens of wrestlers is needed.

### ***Purpose of the Study***

The primary purpose of this study was to determine if there was a correlation between the physiological signs of over-training and the psychological signs of over-training in collegiate wrestlers.

The investigation would help to explore the relationship between the physiological marker, cortisol, and the psychological marker, rest-recovery score from the RESTQ-Sport Questionnaire. This information could help the athletes, coaches, athletic trainers, and other allied health professionals understand the warning signs of over-training and help the athlete understand the need for proper intervention to possibly prevent over-training.

The present study specifically evaluated differences in cortisol levels and rest-recovery scores by weight class, and evaluated the relationship between the physiological marker being tested (cortisol) and the psychological marker (rest-recovery score) being measured using the RESTQ-Sport psychological test. The focus of this study was to evaluate the belief that a correlation existed between the physiological signs and psychological signs of over-training/under-recovery in male collegiate wrestlers.

### ***Significance of the Problem***

The assessment of the physiological and psychological signs of over-training syndrome is a process that greatly benefited athletes who primarily train for, and compete in, longer duration distance sporting events such as marathons, triathlons, bicycle road racing, and others. The process of this investigation has served to link the need for adequate training with adequate recovery for short competitive duration.

While distance athletes and individuals involved in team sports have been studied extensively, little research has been completed with wrestlers. The current research provided a mechanism to document expectations and planning for a preferred training and recovery for coaches (wrestling and other), athletes (wrestlers and others), and those who assist (allied health professionals, strength coaches, nutritionist, sports psychologists and others) these athletes to compete.

### ***Research Questions***

The following research questions were tested:

1. What is the relationship between salivary cortisol and recovery-stress for all wrestlers combined?
2. What is the relationship between salivary cortisol and recovery-stress for each wrestler, individually, as they represented different weight classes?
3. Did salivary cortisol and recovery-stress scores significantly change during the course of the 12-week training?

### ***Delimitations***

This study had the following delimitations:

1. Subjects were male collegiate wrestlers between the ages of 18 and 25.
2. Subjects had successfully competed for and attained a varsity position on the college or university wrestling team.
3. Subjects were healthy individuals with no underlying health (i.e. chronic illness, injury etc.) concerns.



### ***Limitations***

The research may have been limited by:

1. The subjects were asked to self-report (respond to questionnaire) their levels of recovery.
2. The subjects were asked to self-report any illnesses or injuries.
3. The limited sample size.
4. The convenience of the sample.

### ***Assumptions***

The following assumptions were made:

1. The subjects made an honest effort to comply with the protocol of the study.
2. The subjects made an honest effort to answer questions in the RESTQ-Sport questionnaire.
3. The subjects made an honest effort to reveal any information about illness and injury.
4. The subjects fully participated in the sampling time including pre-season, season, and post-season data collection.
5. Except for following NCAA guidelines and the procedures for attaining a varsity position there was no attempt to account for variability due to existing physical status or subjects.

### ***Definition of Terms***

AMENORRHEA: absence or abnormal stoppage of the menses (Doorland's, 2003).

AMINO ACID: any organic compound containing an amino (-NH<sub>2</sub>) and a carboxyl (-COOH) group (Doorland's, 2003).

ANOREXIA: lack or loss of the appetite for food (Doorland's, 2003).

AUTONOMIC: self-controlling: functionally independent (Doorland's, 2003).

BRANCH-CHAIN AMINO ACID: leucine, isoleucine, and valine; they are incorporated into proteins or catabolized for energy (Doorland's, 2003).

CORTISOL: the major natural glucocorticoid synthesized in the zona fasciculata of the adrenal cortex; it affects the metabolism of glucose, protein, and fats and has appreciable mineralocorticoid activity. It also regulates the immune system and affects many other functions (Doorland's, 2003).

CYTOKINE: a generic term for non-antibody proteins released by one cell population on contact with specific antigens, which act as intercellular mediators, as in the generation of an immune response (Doorland's, 2003).

CYTOTOXIC: pertaining to or exhibiting cytotoxicity (Doorland's, 2003).

CYTOTOXICITY: the degree to which an agent possesses a specific destructive action on certain cells or the possession of such action (Doorland's, 2003).

DALTON: an arbitrary unit of mass, being 1/12 the mass of the nuclide of carbon-12, also called atomic mass unit (Doorland's, 2003).

ENDORPHIN: any of three neuropeptides, amino acid residues of  $\beta$ -lipotropin; they bind to opioid receptors in the brain and have potent analgesic activity (Doorland's, 2003).

EPINEPHRINE: a catecholamine hormone secreted by the adrenal medulla and neurotransmitter, released by certain neurons and active in the central nervous system (Doorland's, 2003).

**ESSENTIAL AMINO ACIDS:** the nine  $\alpha$ -amino acids required for protein synthesis that cannot be synthesized by humans and must be obtained in the diet: histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine (Doorland's, 2003).

**EXCITATORY AMINO ACIDS:** a group of non essential amino acids that act as excitatory neurotransmitters in the central nervous system, including glutamic acid or L-glutamine, aspartic acid or L-aspartate, and the excitotoxins (Doorland's, 2003).

**GLUCONEOGENESIS:** the formation of glucose from molecules that are not themselves carbohydrates, as amino acids, lactates, and the glycerol portion of fats (Doorland's, 2003).

**IMMUNOLOGY:** a branch of biomedical science concerned with the response of the organism to antigenic challenge. It encompasses the study of the structure and function of the immune system (Doorland's, 2003).

**LEUKOCYTE:** a colorless blood cell capable of amoeboid movement; there are several types, classified into the two large groups: granular and nongranular (Doorland's, 2003).

**MICRONUTRIENT:** any essential dietary element required only in small quantities, e.g., trace minerals (Doorland's, 2003).

**NATURAL KILLER CELL:** cells capable of mediating cytotoxic reactions without prior sensitization against the target (Doorland's, 2003).

**NONESSENTIAL AMINO ACID:** the eleven  $\alpha$ -amino acids required for protein synthesis that are synthesized by humans and are not specifically required in the diet: alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, proline, serine, and tyrosine (Doorland's, 2003).

**OSMOLALITY:** the concentration of osmotically active particles in solution expressed in terms of osmoles of solute per kilogram of solvent (Doorland's, 2003).

**OVERTRAINING:** an accumulation of training or non-training stress resulting in long-term decrement in performance capacity with or without related physiological and psychological signs and symptoms (Kreider, Frye & O'Toole, 1998).

**PARASYMPATHETIC:** of or pertaining to that division of the autonomic nervous system made up of the ocular, bulbar, and sacral divisions (Doorland's, 2003).

**PHYSIOLOGY:** the science which treats of the functions of the living organism and its parts, and of the physical and chemical factors and processes involved (Doorland's, 2003).

**PSYCHOLOGY:** that branch of science which deals with the mind and mental process, especially in relation to human and animal behavior (Doorland's, 2003).

**RECOVERY-STRESS QUESTIONNAIRE FOR ATHLETES (RESTQ-Sport):** an instrument that systematically reveals the recovery-stress states of athletes (Kellman & Kallus, 1996).

**RHABDOMYOLYSIS:** disintegration or dissolution of muscle, associated with excretion of myoglobin in the urine (Doorland's, 2003).

**SYMPATHETIC:** pertaining to, caused by, or exhibiting sympathy (Doorland's, 2003).

## CHAPTER II

### REVIEW OF LITERATURE

#### *Introduction*

Wrestling is an excellent example of a sport that conditions the athletes to work hard and ignore signs of stress and lack of recovery. The athletes that give into these signs are often times labeled as weak and unwilling to train hard. The combination of skills required to be competitive are manifest in the type of training and motivation to train at the level of competition. The confounding requirements to achieve the right balance of speed, flexibility, suppleness, and strength add to the lack of knowledge in how best to train. Wrestling is not financially rewarding in terms of a career choice. This means that the highest level of achievement could be a tribute to glory at the collegiate and international level. Typically, wrestlers struggle to maintain a balance between their weight and the amount of training required to compete effectively. They often believe that they will have an advantage of superior ability and strength if they are able to compete at a lower weight. This invariably leads to over-training by not providing the body the necessary elements to allow complete recovery between workouts.

This study wove together various factors of over-training as they related to loss in weight and increase in strength. Unfortunately, the amount of literature relating to college wrestling from clinical trials and other scientific research was less than adequate (Hines,

2000). Studies of sports whose training regimen rival that of college wrestling, not in type but in duration and intensity, was used to create an idea of how over-training can be addressed. Training in college wrestling and other sports, combined or exclusively used, endurance, speed or strength to place physical stresses on the body. Those stresses indicated were physical, physiological, biochemical and psychological. Wrestlers and those who trained for long periods of time expended a lot of physical energy, and surprisingly were more susceptible to illnesses that stress the immune system (Mackinnon, 1999).

### **Over-training to Decrease Weight**

#### ***Dehydration***

Albert (2000) indicated that dehydration could cause severe problems leading to the breakdown of the functioning of vital organs and can even result in death. She stated that dehydration reduced muscle endurance. But a small loss of water, between one and two percent, did not reduce muscle strength. She believed that while this loss of water might help in very short-lived high impact sports, any duration training under conditions of dehydration would lead to dire consequences. Albert (2000) indicated that loss of water from muscles was likely to work only for sprint events at world-class levels.

Dehydration was one of the problems that arose from over-training. Dehydration resulted in metabolic drift, the gradual increase in oxygen uptake and subsequent increase in cardiac output required to perform the same external workload which may increase heart rate through increase of  $VO_2$  and central sympathetic stimulation which is primarily intended to augment fatiguing skeletal muscle. She continued by stating that even in

cases of small levels of dehydration, an athlete cannot perform to his or her potential. This performance deficit occurred because a one or two percent dehydration reduced cardiac output. This is never truer than in the sport of wrestling, which combines speed, strength and endurance for relatively long periods of time (Albert 2000).

With dehydration and fasting, and when combined with over-training, athletes and wrestlers make the mistake of loading up on nutrition through binge eating and drinking. This practice can be counterproductive. This is because rehydration should occur first. Proper rehydration involves the intake of large amount of liquids. Eating without adequate liquids often results in diarrhea, vomiting and gastrointestinal pains. This indicates that rehydration is not complete since water is key in restoring a system's gastric and other digestive juices. Most processes in the human body occur in aqueous solutions. Any enzyme action would be nullified if it were forced to occur in the absence of adequate amounts of water. Albert (2000) also suggested that liquids should be consumed first and in moderate quantities. Nutrients could be supplied from the liquid through the use of sports beverages. She further suggested that the fluid be a few degrees cooler than the normal body temperature. Cooler liquids enabled the water to be absorbed into the body more easily. Carbohydrates, especially in the liquid form, were the best because they can be assimilated into the system quickly (Albert, 2000).

The measurement of osmolality, or concentration of particles in urine, is one way to measure dehydration. A test set of 29 athletes that trained under conditions of extreme heat were able to maintain their hydration levels to around normal based on feedback from osmolality of their urine (Shirreffs & Maughan, 1998).

These researchers utilized eleven subjects in a trial. The subjects' first urine sample of the day was collected for five consecutive days. The osmolality was determined and then compared to osmolalities from the urine of a control cohort. With athletes who over trained with the intent of losing weight, and who accompanied the training without the uptake of fluid, the osmolality of urine was approximately three hundred mosmols per kilograms greater than those for the control. Using these results, the researchers established a set of guidelines that provided feedback to athletes (Shirreffs & Maughan, 1998).

In college wrestling, the year 1997 can be considered a year of tragedy (Viscardi, 2004). Three college wrestlers, William Billy Saylor, a nineteen year-old student attending Campbell University in North Carolina, Joseph LaRosa, a twenty year-old student attending the University of Wisconsin at La Crosse, and Jeffery Reese, a twenty-one year-old student attending the University of Michigan died from over-training. All three wrestlers were victims of their attempts to leverage what they thought would be superior strength in a lower weight category by dehydrating themselves, working out in elevated temperatures, and using clothing to increase body insulation to increase body heat. Jeffrey Reese attempted to lose seventeen pounds in one exercise session so that he could wrestle in the 150 pound category. He worked out in a 92-degree room wearing a rubber suit. Sources are not clear whether this was the first time he attempted this method of weight loss. He died of kidney and heart malfunction. The official death certificate called it "death from rhabdomyolysis." His heart and kidneys stopped working because of a "cellular breakdown of skeletal muscle due to heavy exercise." Reese also refused to hydrate himself while exercising. This cost him his life. Joe LaRosa was also wearing a



rubber suit. He was in steam-filled shower room. According to Dan Gable, the wrestling coach at the University of Iowa during 1997, LaRosa was trying to be under a preferred weight before the weigh-in which was going to happen in the next twenty-four hours. He collapsed and died while riding a stationary bike. This is also what took Saylor's life. He was in a predawn workout when he suffered a heart attack. All three wrestlers, according to researchers Luttermoser, Gochenour, & Shaughnessy (1999), were attempting to lose, or had previously lost, 15% of their body weight. The athletes were trying to lose between 30 to 40 pounds of their pre-season weight.

After the deaths of LaRosa, Saylor and Reese, the National Collegiate Athletic Association (NCAA) created immediate guidelines so that rapid weight losses and gains could be avoided (NCAA, 2007). This meant that the use of rubber suits or any non-permeable clothing was banned. Additionally, to prevent athletes from binge eating and drinking after pre-competition weigh-ins in an effort to remove and regain weight while maintaining strength, weigh-ins were to be conducted two hours prior to competition as opposed to twenty-four hours. Also, a seven-pound (3.1 kg) weight allowance was instituted. This meant that a wrestler could compete in a weight class seven pounds lower than his or her weight during competition (NCAA, 2007).

For a brief period, after these cases, there was an increase in effort to identify the causes of the need for weight loss. At the department of Kinesiology, Arizona State University, Alderman et al., (2004) tested more than twenty-six hundred college level competitive wrestlers. Their pre-season weights and post-season weights were determined. Also, some of the athletes were randomly weighed just prior to the beginning of a wrestling bout. The following two parameters were measured: rapid weight gain, and

rapid weight loss. The researchers discovered that rapid weight gains between weigh-in and competition averaged at 3.4 kilograms. In some cases, weight gains never occurred. In other cases, weight gains of up to seventeen kilograms were not uncommon (Alderman, Landers, Carlson & Scott, 2004). Over-training by means of running using sweat inducing clothing was a common weight loss method. The cyclical weight gain and weight loss was more efficient at making weight among the more successful athletes. This possibly created the notion that a wrestler could drastically lose weight before a weigh in and then easily regain it in time for competition with no decrease in performance (Alderman et al., 2004).

Perriello et al., (1995) provided a breakdown of different methods used to lose weight. The most common method indicated was fasting and the restriction of fluid intake. Not far behind were running and over use of exercise devices. About a third of those tested used rubber suits. A smaller number of subjects perspired in a sauna, induced vomiting or used diuretics. These investigators published a table which described the different methods used to lose weight; running was used 73% of the time, fasting was used 75% of the time, and restricting fluids was used 73% of the time. These discrepancies in percentage calculations suggested that most of the athletes and wrestlers tested used two or more methods of weight reduction.

Davis (2004) stated that damage done by frequent purging was starkly portrayed in the documentary on HBO on horse jockeys, titled "Jockey." The documentary followed the travails of three jockeys, one of who became dangerously ill because of his methods for weight loss. This jockey spent his life savings on his hospital bills. His fellow jockeys planned a musical program to help pay for medical bills. These athletes

are under great pressure to maintain a certain weight at the risk of losing their livelihood. According to the documentary, this practice was not only tolerated but also encouraged. In the jockey dressing rooms, the row of toilets contained one toilet which was designed differently and made purging more convenient (Davis, 2004).

The constant purging also resulted in another negative effect on the body. Partially digested food with the concentrated stomach acids damaged the esophagus during the regurgitation. The acid also wore away the lining of the teeth and most jockeys had to wear bridges early into their careers (King & Mezey, 1987). College and high school wrestlers faced very similar predicaments, since the methods used to maintain weight were predominately the same.

Albert (2000) asserted that most wrestlers were under the mistaken assumption that if the weigh-in occurred twenty-four hours prior to the event, they could regain their strength by overloading on high-energy content food so that they did not feel the effects of the weakness from having lost a large amount of weight in a short period of time. He added that the rate of assimilation of food needed to rebuild the lost nutrients and water required a lot longer. So in effect, the wrestlers move was counterproductive.

A more comprehensive clinical trial also indicated that loss of weight did not really give an athlete an advantage (Roemmich & Sinning, 1997). The clinical trial was conducted using two groups of test subjects. The entire respondent cohort consisted of boys with a mean age of fifteen and a half years. A control group was given a regular diet in typically prescribed combinations of carbohydrates, proteins and fats. The test group consisted of wrestlers. The control group was described as “recreationally active.” The wrestlers were fed a diet that was high in carbohydrates and low in fat. The diet was

designed to reduced weight. No protein or energy supplements were given during the wrestling season. The results indicated that although the wrestlers had lost weight in terms of fat as well as muscle mass, they did not possess the same strength in the arms and legs prior to the season. After the wrestling season was over, and upon resuming energy intake, the wrestlers regained their strength (Roemmich & Sinning, 1997).

Oppliger et al., (1995) maintained that some of these ill-conceived notions regarding effective weight loss actually found their origins in the high school environment. A new program was instituted for Wisconsin high school. Based on their body mass indices, weight classes were established. This constituted a new direction in which weight was established over empirical body weight measurements rather than on preconceived demarcations. More importantly, no wrestler was allowed to lose more than three pounds a week. Weekly weight measurements were enacted for all the test subjects. The researchers reported that since the implementation of this program, instances of weight cutting, over-training and fasting prior to a weigh-in had significantly reduced. This trial involved two hundred test subjects (Oppliger et al., 1995). Based on a comprehensive survey, the Center for Disease Control, (CDC) (2004), determined that the need to decrease weight was also prevalent among non-athletic college students. Without taking into account possible confounding factors such as college students who were athletes versus those who were recreational athletes and did not perform exercise, the survey made apparent that the need to reduce weight reached epidemic proportions because of societal misconceptions as to what an individual's ideal weight should be (CDC, 2004).

Weyer et al., (1998) reported a study in which 4700 students participated. Twenty-percent of men and women were over-weight. Forty-two percent of the women and less than half of the men had dieted in the thirty days prior to the study. Forty-percent of the men had exercised, and less than two-thirds of the women had exercised thirty days prior to the test. Smaller numbers, four percent of the women and less than one percent of the men, had vomited to lose weight. Seven percent of the women and one percent of the men had taken diet pills to lose weight. The risky behaviors that became apparent as a result of the study were anorexia and bulimia. The disparity in numbers among sexes indicated the pressure exerted on women to fit an ideal weight could be more pronounced. The likelihood is strong that when career and fame are at stake, wrestlers would face a lot of pressure to meet weight restrictions (Weyer et al., 1998).

### **Over-Training to Build Strength**

#### ***Over-Training***

The preceding sections titled Over-Training to Decrease Weight included several examples of over-training and other modes used to decrease weight. Over-training to build strength, however, could result its own set of consequences

Although incidences of death are few, symptoms of over-training could cause significant physical and psychological damage. The effects of this type of training could be long lasting. One such example was training with resistance. The primary aim of resistance training was to increase size, endurance, bulk, strength, power and velocity of relevant movement (ACSM, 1995). Resistance training involved a combination of training volume and intensity. Most athletes combined both volume and intensity. There

were different combinations of the volume and intensity parameters. Most athletes involved what is called “periodization” (ACSM, 2002). This means that an athlete would use one set of exercises and repeat this at periodic intervals. One of the markers which indicated that the person training might be over-doing exercise became apparent when physical impairment occurred. This became more serious if recovery times increased and time required to return to the exercises in question were prolonged. Sometimes impairment resulted in a decrease in willingness to exercise, a psychological marker (Fry et al., 1996a). Over-training in resistance increased if the resistance was too high or the exercise volume was too much; that is, too many repetitions occurred at a weight. Ideal resistance training involved an optimal level. If the resistance was high, the repetitions or sets should be reduced. Low resistance weights should be combined with higher repetitions (ACSM, 1995). From a resistance volume perspective, the ratio of two hormones, testosterone and cortisol, played an important role (Marinelli et al., 1994). While studies have not identified a direct correlation with the levels of this ratio, there is evidence to show that the testosterone to cortisol ratio decreased as the training volume increased. The ratio of testosterone to cortisol is thought to regulate anabolic process in recovery, so a change in this ratio is considered an important indicator and perhaps a cause, of overtraining (Kuipers& Keizer, 1988). Testosterone is a hormone that has a unique effect on a man's total body. It is produced in the testes and in the adrenal glands. It is to males what estrogen is to females. Testosterone helps to build protein and is essential for normal sexual behavior and producing erections. It also affects many metabolic activities such as production of blood cells in the bone marrow, bone formation, lipid metabolism, carbohydrate metabolism, liver function, and prostate gland

growth (Dorland, 2003). In the context of this work, testosterone was useful for increasing lean muscle mass and muscle strength. Cortisol is a hormone produced by the adrenal glands. This will be discussed at several points in this literature review. Cortisol is used in metabolizing nutrients in the system. It also helps in maintaining the immune system, and aids the body in fighting against physical and emotional stresses (Dorland, 2003). Essentially, cortisol energizes the body in the fight or flight response to stress. Excessive release of cortisol results in the correspondingly excessive break down of nutrients. This occurs in response to vigorous exercise. The decreased ratio thus hampers recovery times (Fry, 2001).

The above examples of over-training were the detriments of excess volume training. With increased resistance however, the mechanisms of involved problems are different. The testosterone/cortisol ratio does not increase with increases in the resistance aspects of training. However, catecholamine levels are increased with increases in resistance. Typical catecholamines are epinephrine and norepinephrine secretions that are implicated in symptoms of depression due to either decreased secretions or increased uptakes. Since catecholamine levels are secreted in the nervous system, the nervous system attempts to compensate for loss of muscle strength by increasing catecholamine secretions. Fry (2001) asserts that this produces an artificial work-out high, “runners’ high,” a term that is described as a “feel-good” sensation after a particularly strenuous work out. Clinicians believe that seeking this high might result in people working out at levels that are dangerous to their bodies. One of the theories advanced was catecholamine release (Lehman et al., 1992). This was associated not only with runners’ high but also with the second wind. The depressed release of this chemical might cause symptoms of

depression (Mengshoel, Saugen, Forre, & Vollestad, 1995). Another popular ideation is the beta-endorphin theory. Like catecholamine, beta-endorphin secretions have been implicated in the feeling of runner's high (Colt, Wardlaw, & Frantz, 1981). These endorphins are also the first respondents in the study of pain. They are also implicated in helping with mood disorders and in the response to stress.

Sometimes, the problems with over-training do not directly vary with decreased muscle strength or muscle fatigue. Over-training also affects the joints. Since long recoveries and the reticence to begin training again are signs of over-training, joint overuse results in some of the symptoms of over-training (Fry et al., 1996b). Psychological factors might be associated with the decreased desire to rejoin an exercise regimen.

Hines (2001), writing in the Oklahoma Sports Science and Orthopedics website provided a formalism for overtraining. He called it "overtraining syndrome," and has conducted significant research from surveys and from written reports. As a researcher, he bemoaned the fact that comprehensive research had not been conducted on such a health-issue, and asserted that there are several causes of the symptoms of "overtraining syndrome." In addition to the symptoms that have been identified earlier, he found that the immune system was compromised such that wrestlers suffered from ringworm and herpes outbreaks, conditions that one would not typically associate with wrestling related injuries. Hines (2001) also believed that most of the problems from over-training in college wrestlers primarily came from a lack of understanding of how much exercise was really necessary, and what constituted optimal training conditions for the wrestlers. He criticized the fact that there was too much pressure on athletes and coaches to perform.



This coupled with little knowledge of nutrition facts made for difficult progress in the right direction. The collegiate wrestling season lasts the year around, and he expressed disapproval that there were not enough provisions to allow wrestlers to get adequate and rejuvenating rest.

Herzog et al., (1985) stated that yo-yo dieting or constant recycling of weight during a wrestling season actually disrupted the typical metabolic processes to such an extent that the body learned to use efficiently whatever energy it could get from the disrupted nutrition. Thus, further loss of weight in the weight-cycling process did not occur as much as when the cycle was first begun. Metabolic efficiency increased fourfold when nutrients were used (Herzog et al., 1985).

Hines (2001) stated that overtraining syndrome reflected an accumulation of training and non-training stresses that resulted in a long-term decrease in performance in which restoration of performance may take weeks to months to resolve. These problems arose because the stressors, physical and emotional, constantly impacted the athlete without time to recover. He also maintained that the fine line between under-training and over-training should be demarcated. Optimal training levels occurred when the person undergoing training realized that progressive overloading would enhance fitness. In the same progression, the body responded positively because the body was trained to adapt. When the body responded negatively, acutely or chronically, to an applied stress, it was a sign that the athlete or wrestler was over-training (Hines, 2001).

### ***Symptoms of Over Training***

To provide a comprehensive summary of the problems associated with over-training, the following symptoms can be considered: the decreased ability to perform in terms of strength, power, muscle endurance, cardiovascular endurance, and flexibility (i.e., related to the ability to use muscles rapidly in response to a stressor). These symptoms are some of the characteristics of over-training (Kreider, Fry & O'Toole, 1998). Muscle coordination decreases. Also importantly, there is a decrease in tolerance to training, and recovery times are significantly prolonged (Kreider, Fry & O'Toole, 1998). Physiological markers of interest, are parasympathetic (i.e. low resting heart rate, rapid heart rate recovery post exercise, decreased catecholamine levels, unusual digestive disturbance) and sympathetic responses (i.e. restlessness, excitation, increased resting heart rate, increased blood pressure, increased metabolic rate) (Lehman et al., 1992; Fry & Kraemer, 1997). These are physical and motor characteristics. Physiologically, overtraining causes altered resting heart rate (Kreider, Fry & O'Toole, 1998). Heart murmurs occur. Body fat decreases, although this might not necessarily be a negative. Body weight also decreases. Oxygen volume and heart rates also increases even when a workout is not taking place (Lehmann et al., 1998). The lactic acid response from exercise often decreases. Basal metabolic rates decrease. Chronic fatigue sets in, although this might not be associated with the more formalized Chronic Fatigue Syndrome (Nijs, Vanherberghen, Duquet, & De Meirleir, 2004). Over-training causes metabolic changes, which can impair sleep. It can also cause involuntary eating disorders which are not borne out of a need to rapidly lose weight. Headaches are one of the frequent causes of over training. Gastrointestinal disorders are rife (Nijs et al., 2004).

In the past twenty years, the number of women participating in strenuous sports has increased dramatically. Many questions and concerns have arisen with the changing role of the female athlete. For women, there can be significant disruptions in menstrual cycles (Dueck et al., 1996a). Estrogen, the female hormone, acts as an inhibitor in terms of the process of building muscle strength. However, estrogen does tend to increase endurance (Dueck, Manore & Matt, 1996b). Especially among women athletes, a triad develops when they over-train and lose extensive amounts of weight. This triad involves disordered eating, amenorrhea (i.e., disrupted menstrual cycles), and osteoporosis (Nattiv et al., 1994). Used as a method to decrease weight, over-training and disordered eating results in disruptions of menstrual cycle. Because of the connection between hormonal levels and bone formation, bone density is reduced. Researchers state that bone density increases when menstruations revert to normal. Typically, however, female athletes who experience disordered eating and amenorrhea reflect bone densities of someone twice as old (Brooks-Gunn, Warren, & Hamilton, 1987; Garner & Garfinkel, 1980). If this triad is a part of a female athlete's life, over-training can give rise to bone disorder which might be permanent. The problems of scoliosis and stress fractures are not far behind. This triad, if brought on by over-training can result in hampering of any athletic efforts (Myburgh et al., 1990; Warren et al., 1991).

Over-training also affects muscles. They tend to be sore, and short and long term damage may occur. Over-training can cause restriction of joint movement with the wearing away of the sinusoids and ligament elasticity (Kibler et al., 1989). Some of the problems affecting the bones and joints can eventually give rise to problems of chronic osteo-arthritis (Hoaglund, Yau, & Wong, 1973). Over-training and weight-reducing

strategies such as fasting or other ways of limiting nutrition intake can reduce muscle endurance. These reducing strategies tend to reduce glycogen stores from the liver and the muscles (Sherman et al., 1981). This often results in lightheadedness and lean tissues also show stunted growth. These reductions in performance metrics are observed even after an athlete is on a weight loss diet for three days (Helge, Richter & Kiens, 1996).

While working-out can give rise to an increased response of “feel good” neurosecretions, they can also create an artificial feeling of strength (Morgan & Goldston, 1987). When this “high” wears away, especially if it is associated with chronic pain, psychological results can create the exact opposite effect and depression can set in (Gross, 1994). Apathy towards society, normal life, and the need to exercise is quite commonplace. The increase in self-esteem that generally results from exercising can result in an emotional move in the opposite direction (Raglin, 1990). People who experience problems with over-training experience problems with concentration, and are often overtly sensitive to stress (Steptoe, Kearsley & Walters, 1993). This brings us back to the ideation of the hormonal imbalances. While testosterone, cortisol and catecholamines play an important role in the development of muscles, they also have a myriad of other functions (Orth, Kovacs & Debold, 1992). If these secretions are affected based on over-training, it is not difficult to see that other metabolic aspects of life that depend on these secretions can also be affected.

Later in this literature review, the problems with decrease in the strength of the immune system will be explored in detail. An introduction at this point will suffice. With over-training comes an increased occurrence of illnesses or infection, which may not be directly related to the muscles, bones or joints (Roberts, 1986). Even in these non-

exercise related illnesses, much like the healing of muscles, recovery times are higher (Fitzgerald, 1991). The levels of the mainstays of the immune system, neutrophils lymphocytes, and agents that trigger non-sexual cell division and replication such as mitogens and eosinophils, are decreased (Mackinnon, 1999). This means that the body becomes more susceptible to illnesses. Additionally, from a biochemical standpoint, the hypothalamus does not function effectively. The hypothalamus, a tiny region in the frontal portion of the brain, is known as the “maintainer of the status quo,” regulates involuntary function such as breathing, heart rate and blood pressure (Dorland, 2003).

The sex hormone binding globulins (SHBG), which maintain normal levels of androgen and estrogen in the body, are also affected. Muscle glycogen is decreased. Iron, serum hemoglobin and ferritin, the protein that binds iron in the blood, also decreased. Nitrogen levels in the blood are also depleted (Fry, 2001).

Amino acids are called the building blocks of protein, which are the primary constituents of muscles (Butterfield, 1991). Central Fatigue is a condition associated with over-training (Davis, 1995). From a biochemical perspective, it is associated with a build up of a ringed amino acid called tryptophan. Excessive levels of tryptophan result in excess secretions of the neurochemical serotonin (Davis et al., 1992). Serotonin secretions are responsible for many of the problems with over-training syndrome. Excess serotonin induces a fatigue- like state (Davis et al., 1993). It also causes drowsiness, changes the endocrine system, and impairs the autonomic system. Additionally, it suppresses the appetite and also changes “neuromotor excitability” (Hines, 2001). Depending on its levels, serotonin, which is either a mood enhancing or depressing neurochemical, also causes the psychological perception of fatigue (Davis et al., 1993).

The perception of fatigue is not necessarily negative. Serotonin is perhaps the ultimate mechanism to inform the body that it is being stressed to levels that are not sustaining. The increase in tryptophan and the onset of Central Fatigue coincides with the breakdown of other building amino acids (Davis, 1995).

Branch chain amino acids such as leucine, valine and isoleucine are broken down due to over-training (Davis, 1995). Glutamine is also broken down (Kargotich et al., 1992). There is a general depletion of amino acids in the muscles. This causes muscle mass to break down to supply the rapidly diminishing caches of amino acids. This also causes the muscle's glucose and glycogen supplies to decrease (Kreider, Fry & O'Toole, 1998). Additionally, loss of amino acid causes a reduction in muscle mass and the loss of energy-giving glucose causes fatigue (Kreider, Fry & O'Toole, 1998).

### ***Injuries from Over-Training***

It is necessary to recognize the kind of injuries that are typical to wrestling. Pasque and Hewett (2000) performed a comprehensive study, using 458 wrestlers from fourteen schools. Their injuries were monitored over an entire college-wrestling season. Roughly half the athletes suffered injuries or illness. The most common injuries were associated with either typical wrestling moves such as "takedowns," or with the parts of the body most used by wrestlers, such as shoulders and knees. If all the workouts and wrestling matches were taken into account, the injuries occurred one out of every 167 such instances. Takedown injuries were most common with 68% of the injuries occurring during this maneuver. When focusing on specific injuries to body parts, shoulder injuries were the most common accounting for 25% of all injuries. The next highest injuries

occurred at the knee, which accounted for 17% of the injuries. The preceding summary demonstrates that every aspect of the human body is needed for normal functioning may be adversely affected by over-training (Pasque & Hewett, 2000).

### ***Physiological Markers***

One of the ways of understanding the amount of training necessary to be competitive, and whether the amount is adequate, can be judged by a subjective scale called the Borg Ratings Perception of Exertion (RPE) (Borg, 1982). This test is more subjective, and Borg (1982) demonstrated that the RPE is significantly correlated with heart rate, oxygen consumption, pulmonary ventilation, and lactate concentration to validate exercise intensity. The RPE relies on the physical activity in which the person is engaged. This test depends on levels of physical fatigue and parameters such as heart rate, a score would be timed by 10 to equal heart rate. Borg's RPE ranges from a score of six, which involves no exertion, to twenty, which reflects maximum exertion. A Borg RPE of between 12 and 14 is often sufficient as a level of moderate exercise activity. To reiterate, Borg's RPE is unique because it involves self-monitoring while performing the activity rather than arriving at fixed guidelines based on repetitions and maximal weights (Borg, 1982).

The target and estimated maximal heart rate is also a good measure to determine if an athlete is over-training. The estimated maximal heart rate is obtained by subtracting a person's age from 220. For moderate exercise, it is recommended that a person perform between 64% to 76% of the maximal heart rate. For vigorous exercise this number rises to 77% to 93% of the maximum heart rate (ACSM, 2006).

Researchers at the University of Wisconsin in Lacrosse conducted a clinical trial to recognize the efficacy of Borg's RPE (Day et al., 2004). They divided the test into three parts. The first part involved exercising to 90% of the one repetition maximum, but with four to five repetitions. The second test was to perform 10 repetitions at 70% of the one repetition maximum. The last set was to perform 15 repetitions at 50% of the maximum. The idea for balancing the volume and the resistance was to ensure that the amount of stressors on the body remained the same. Each exercise consisted of only one set. The exercises involved curls, bench press, back squat, overhead press and triceps pushdowns. RPE were measured immediately after each set and thirty minutes after the exercise was completed. The results showed that RPE was higher for the test set that contained higher resistance and fewer repetitions. This subjective test is very useful when measuring perceived exertion. It also illustrates that higher resistance at fewer repetitions places greater strain on the body (Day et al., 2004).

CDC (2004) provides yet another way to determine levels of activity. It is called the Metabolic Equivalent Level (MET) (CDC, 2004; ACSM, 1995). Using a table prescribed by the CDC (2004), activity levels and METs for those activities can be used as a guide. Essentially, if the MET falls between three and six, the activity is deemed moderate. A MET score greater than six denotes vigorous activity. From the perspective of a reference and scalability of MET, the simple activity of lying back and reading a book is the 1 MET. Strictly defined, a MET is the energy oxygen used by the body (CDC, 2004; ACSM, 1995).



### ***Guidelines to Avoid Over Training***

Prevention of over-training must begin with the need to structure the individual athlete's training programs in such a way as to allow an adequate balance of training stress and recovery (Lehman, Foster & Keul, 1993). Too often the recovery element is overlooked as an essential aspect of any training regime (Fry, 2001).

The inherent dangers in over-training, from the perspective of losing weight rapidly as well as for gaining strength, have caused certain guidelines to be initiated to prevent mishaps and even deaths (NCAA, 2007).

It has long been recognized that individual athletes can tolerate different levels of training and competition stress, and require differing lengths of recovery periods (Budgett, 1994). Therefore the need for individualized training programs remains paramount. Varied training is also essential to avoid the problem of monotonous over-training, which has been identified particularly among swimmers (Kuipers & Keizer, 1988)

During periods of high extraneous stress, whether environmental, occupational, educational, or social, training volumes and intensities may need to be modified and scaled down. Together, the combination of stresses may exceed the individual's capacity to adapt (Selye, 1957). It is entirely possible for over-training to develop when a normally tolerable volume and intensity of training is undertaken. However, over-training generally occurs in combination with other stresses from environment, profession, or private circumstances (Kreider, Fry & O'Toole, 1998).

Periodization is a variation in training volume and intensity over a specified period. The goal is to prevent staleness while peaking physiologically for competition

(Kraemer & Ratamess, 2000). To avoid over-training, periodization has been suggested because it tends to reduce the monotony and adds variety to the training. Care should be taken that training volume and training intensity are not only varied but also balance each other out. High volume should be counter-balanced with low intensity, and low volume should be counter-balanced with high intensity. Both volume and intensity should not be carried out for long periods of time (Kraemer & Ratamess, 2000). Muscles and joints that are overly fatigued should be rested. Circuit training provides a balanced workout and does not overtly stress any one group of muscles or joints. The effect of training should not include focus on muscles and joints alone, cardiovascular factors should also be taken into account as well (Fry, 2001).

In terms of weight measurement, researchers, physicians, and coaches have suggested that based on preseason optimal weights and the demands of a wrestling competitive season, a minimum weight has to be assigned to a wrestler (Horswill, 1992). This represents the lowest weight at which the wrestler can be allowed to wrestle. Additionally, weight loss should be constantly measured and not allowed to drop below minimum levels. Horswill (1992) stated that coaches, athletes and sports administrators should be made aware of the new guidelines so that an internal support structure for struggling athletes might be formed.

Benson (2004) proposed added suggestions on how to prepare for competition within a sound strategy for strengthening and nutrition. He stated that during workouts, fat is burned and the reduction of carbohydrate reserves is actually stymied. This was a positive occurrence. He also suggested that continuous nourishment was provided so that the vital substances lost from the body could be continually replenished. Before a

marathon, people often eat a large pasta meal. This carbohydrate “hyper-loading” resulted in excess energy that did not result from depleted muscle glycogen. This attention to a “pre exercise” diet might result in increased performance. Benson (2004) also stated that taking care of cardiovascular factors is as important as muscle strengthening. The rate of fat being burned during exercise depends upon how much fat is accessed through the blood. Better cardiovascular fitness will enhance circulation and will burn fat more easily. He added that over-training and rapid weight loss regimens actually depleted cardiovascular performance due to lower heart rates and blood volume turnovers.

Hines (2001) also provided more specific information on how to nutritiously support the body during training, especially during the process of over-loading. Over-loading tests the body’s response to progressively higher stresses through the process of strengthening the body. He cautioned that over-loading should be distinguished from over-training. Hines (2001) also suggested that wrestlers should maintain a high carbohydrate diet in order to support the intense exercise regimens. He maintained that a pre-exercise diet four to five hours before an event or exercise should hyper-load carbohydrates with excess stores of energy. If the exercise was to be prolonged, a small carbohydrate-laden meal should be consumed a short time before the actual workout. He also suggested that an electrolyte drink should be used to nourish cardiovascular exercises which lasted over one hour. Additionally, wrestlers should consume a meal two to three hours after working out. This meal should contain carbohydrates and proteins in the ration of 3:1. Hines (2001) continued by suggesting that the athlete consume glutamate supplements about two hours before strenuous exercise.

He maintained that the loss of immune resistance was due to the depletion of glutamine reserves in the muscles, and that the catabolic (i.e., breakdown) activity of muscles from over-training far outweighed the anabolic (i.e., building) mechanism.

Quinn (2004) described a study which indicated that glutamine supplements do not prevent muscle loss from over-training. The study was conducted with thirty-six college wrestlers. The cohort was divided into a test group and a control group. Both groups ingested a high protein diet. The control subgroup was administered a placebo, while the test subgroup was administered a glutamine supplement. Results showed that weight loss for all wrestlers came from fat and muscle, however, the loss of muscle mass was not restored with any statistical significance among the glutamine-receiving subgroup (Quinn, 2004).

Despite the above guidelines, carbohydrates and proteins in the ration of 3:1, and consume glutamate supplements about two hours before strenuous exercise, there are some confounding characteristics. A mere recommendation of increasing carbohydrates from a simplistic standpoint means that the sugar levels will be easily absorbed and serve as energy boosts (Finn, Dolgener, & Williams, 2004). Simple sugars provide a quick fix, while complex starchy sugars obtained from rice and pasta provide a sustained energy boost. In addition to the potential physical “boost,” athletes and coaches also need to understand whether possible psychological changes are due to carbohydrate administrations. Finn, Dolgener, & Williams (2004) studied fifteen college wrestlers who became part of an investigation designed to test the physiological and psychological effects of carbohydrates after a workout. The moods of the participants were judged prior to a weigh-in which was subsequently followed by a workout. The subjects’ moods were

again judged after the weigh-in, and also after the administration of a high carbohydrate substance. This cohort also consisted of a control group. This control group was given a placebo instead of the carbohydrate concoction. Blood lactate responses were measured. The results showed that blood lactate responses were significantly different for the carbohydrate group, and the psychological parameters measured in terms of positive or negative mood measures failed to show a change between the test and the control subjects (Finn, Dolgener, & Williams, 2004).

Another study authored by Langfort et al., (2004) also resulted in mixed results when eight, non-athletic male subjects were administered low carbohydrate nutritional supplements. A confounding factor occurred in the study due to the testing of the non-athletic participants, and it is possible that over-training thresholds for untrained participants were lower and resulted in different responses than trained athletes such as wrestlers. In this study, physiological and biochemical characteristics were measured. The test consisted of performing exercises with progressive loading until failure. The exercises were performed after the subjects were administered a mixed diet. Prior to a repeat of the test, the participants were fed a low carbohydrate diet. After exercising, several parameters were measured. These included the concentration of lactates, ammonia, epinephrine and norepinephrine in the blood. The researchers found that the responses for each of the parameters were different, adding to the confounding. These results were especially revealing since this trial was conducted to understand muscle fatigue and relative rates of catabolism. The test results indicated that epinephrine levels remained unchanged. Norepinephrine levels decreased indicating that they favored a lower workout, as did ammonia levels. Lactate levels changed favoring a higher

workload. The results suggest that with a low carbohydrate diet the breakdown of proteins producing ammonia were rapid, while the breakdown of lactates were much slower (Langfort et al., 2004).

Selsby, DiSilvestro, & Devor (2004) maintained that weight training is an essential component of training for wrestling, and that chromium and chromium chelates (i.e., inorganic compounds formed from multiple covalent-coordinated bonds) in concert with magnesium ions have been implicated in increasing muscle strength. While this clinical trial was not conducted on wrestlers, it included males who were involved in weight training. The results were very good in terms of muscle strength for a one repetition maximum and other weight training exercises at seventy percent of the one repetition maximum when compared to a placebo, which was multidextran. The chromium was administered in small quantities, and using chelates did not show improved or enhanced strength. The results of this study indicated that replenishing broken-down muscles might have more than one mechanism (Selsby, DiSilvestro, & Devor, 2004).

Sometimes, over-training a certain set of muscles will impact other aspects of training. As has been stated before, wrestling is a sport that involves physical strength, flexibility and speed. Scott and Docherty (2004) tested subjects who were not wrestlers but were males of average age twenty-five. These subjects participated in weight training programs. Each participant in the study was assigned a random sequence of a horizontal jump, vertical jump and a five-repetition-maximum set of back squats. Every participant participated in all three parts of the program. A maximum weight was used as a parameter so that the researchers could identify how preloading made an impact on other

exercises and other parts of the body. The study consisted of participants warming up, performing horizontal and vertical jumps, resting, performing progressive back squats until a five-repetition maximum was attained, resting and then performing the jumps again. The results showed that back squats did not significantly enhance or diminish the participants jumping ability. This study however indicates that more testing on different muscle groups correlated with different wrestling type activity would be important to understand the consequences of over-training not only on health but as a counter productive measured to diminishing performance (Scott & Docherty, 2004).

In another study from Yokohama Japan, Miyama and Nosaka (2004) initiated a study relevant to wrestling, but not focused on wrestlers per se. Researchers asked subjects to perform several sets of consecutive drop jumps from a height of sixty centimeters. Half the test subjects performed the jumps on wood, the other half on sand. The results from measurements of maximum isometric force and serum creatinine kinase levels performed immediately before and after the exercise, and several times in the hours after the exercise, showed that muscle damage was severe when the jumps were performed on wood as opposed to when they were performed on sand. The researchers indicated that in some developing countries most wrestling activity took place on sand, at least at the beginning. Although current competitions are conducted on foam, it is important to be cognizant of any underlying surface. Studies show that injuries are more frequent on artificial grass, which is carpet placed over concrete, versus on natural grass (Miyama & Nosaka, 2004).

### *Psychological Markers*

In attempting to understand the psychological factors associated with over-training, athletes must consider a notion known as obligatory running (Garner, Olmsted, Bohr, & Garfinkel, 1982; Holloway & Baechle, 1990). Simply put, this can be described as an addiction to running. And this is not necessarily to attain the “runner’s high.” This concept of “obligatory runner” is seen in marathoners. While this moniker is not assigned to wrestlers, it is conceivable that projections can be made given the need to over-train. Obligatory running goes beyond the mere need to train. It is classified as a behavioral disorder, not unlike anorexia nervosa or bulimia (Garner et al., 1982; Holloway & Baechle, 1990). The need to exercise is the same as the need to binge and purge. Obligatory running is often described as the male equivalent of anorexia (Yates, Shisslak, Allender, Crago, & Leehey, 1992). This comparison appears on all fronts: physiology, psychology and social. The only parameter that every discipline agrees on is that behavior disorder is due to a need for perfectionism. The point though is perhaps like comparing apples and oranges. Researchers have to delve deeper to find the emotional yearnings that cause obligatory running and anorexia (Blumenthal, O'Toole, & Chang, 1984). Naturally, a runner does not have preoccupation with food as an anorexic does and will answer in the negative when questions such as these are asked. Blumenthal et al., (1984) found that personality profiles of marathon runners were different from those of anorexics.

From an assessment of the psychological profiles of college athletes, results are once again conflicting. One study showed that the amount of time spent running was not correlated with eating disorders (Warren, Stanton, & Blessing, 1990). Richert and



Hummers (1986) study showed exactly the opposite results. Unfortunately, these correlations were not associated with wrestling. But the parallels between the need to run, the need to over-train, and specific aspects of wrestling were unmistakable.

Two different tests were used to identify the results from the above two studies. The Eating Disorder Inventory (EDI) is an inventory that lends to self-report. It queries the respondent of this inventory to identify symptoms related to anorexia and bulimia. This 20 minute test contains sixty-four questions with twenty-seven questions added in the second version. The questions focus on eleven separate parameters: “Thinness, Bulimia, Body Dissatisfaction, Ineffectiveness, Perfectionism, Interpersonal Distrust, Interoceptive Awareness, Maturity Fears, Asceticism (provisional), Impulse Regulation (provisional), Social Insecurity (provisional)” (EDI-2, 2004). The Eating Attitudes Test (EAT) is also a comprehensive test that is used more often (EAT, 2004). This test starts out with determining simple questions of age, sex, height, and weight reported at current, lowest, and highest. For medical reasons, it identifies the demographics concerns of the subject. It also queries the training level of the subject. The next part of the questionnaire can be scored based on a scale of “Always, Usually, Often, Sometimes, Rarely and Never.” Questions can ask a tester whether he is or she is terrified of gaining weight, preoccupation and obsession with food, perceptions of self and what others perceive the person to be. In addition there are five questions that ask for Yes/No type answers. These questions range from whether the subject has deliberately purged, taken laxatives or other weight loss pills, and even whether he or she has attempted suicide (EAT, 2004).

Coen and Ogles (1993) conducted further research on obligatory runners’ psychological dispositions. The research used a control group of non-obligatory runners.

The psychosocial parameters measured were “identity diffusion, anger, anxiety and perfectionism.” The results showed that obligatory runners did not express anger or identify diffusion. The report showed, however, that obligatory runners tested higher on anxiety, and they move towards perfection ideals. If this were measured on the EDI questionnaire, it would appear as a “drive for thinness” (Coen & Ogles, 1993). In another study consisting of almost four hundred obligatory and non-obligatory runners, with more than a third of them male, results showed that psychological problems manifested greatly in terms of responses to EAT questionnaires (Slay, Hayaki, Napolitano, & Brownell, 1998). Unfortunately, the motivation to run was mostly negative. It was reported that this motivation surmounts even the necessity to stop on account of injuries. In this study, the problems manifested significantly more in women than in males ( Slay et al., 1998).

The notions that lack of eating and over-training are cyclical and self-perpetuating were advanced by Epling et al., (1983). Based on results obtained in more than one study, the researches indicated that lack of food causes the need to train more. More training in turn forces the person to eat less, hence the cyclical aspect of these behaviors. The authors also indicate that it is possible, though it does not come through in the reports that since this behavior are psychological, not every one suffers from it. The study’s conclusions were derived from studies on rats. There is also an indication that youth are more susceptible than the older generations. In the experiment, rats’ access to ninety percent of adolescent rats and seventy percent of their older counterparts increased their uses of the wheel, inversely commensurate with the decrease in food intake. Some of the rats tested used the running wheel, according to the authors, “literally running themselves to death” (Epling, Pierce, & Stefan, 1983). Davis et al., (1993) stated that anorexia was

associated with participation in sports and the need to over-train, based on their interviews of anorexics who had been hospitalized. When compared to age-matched controls, more than three-fourths of the anorexic patients interviewed indicated that they had participated in competitive sports (Davis, Durnin, Gurevich, Le Maire, & Dionne, 1993). This even included competitive dancing. The need to exercise hampers recovery and pushes patients to relapse at greater rates (Strober, Freeman, & Morrell, 1997).

From a psychological perspective, there is another effect that is mostly prevalent in men, especially college wrestlers. The perception of anorexics is that they are over weight. All efforts, including over training and starvation, are then focused on decreasing this weight (Pope, Katz, & Hudson, 1993). In reverse anorexia, the perception is that they are smaller and less muscular. The reality is that the afflicted person is normal or even more muscular than usual. This false perception leads to the abuse of anabolic steroids. The authors found that the extra strength achieved from these banned substances allowed a person to train more than is normal in order to achieve that seemingly unattainable bulk. While anorexia is a problem even among males, it would seem that reverse anorexia is a premise that is restricted to males.. The emotional factors come into play no matter what the eating or perceived weight disorder. (Pope, Katz, & Hudson, 1993).

The Profile of Mood States is an important psychological marker that can be used to create an emotional profile of an athlete. This is an often utilized questionnaire that is extensible to different surroundings. The original questionnaire contained 65 questions. Later the questions were reduced to 30 items. Test subjects often have to rate different psychological parameters on a scale of one to five, one being “no influence” five being

“completely affected.” The questionnaire is subdivided into the following six subheadings: (a) Tension-anxiety, (b) Depression-dejection, (c) Anger-hostility, (d) Vigor-activity (physical or mental strength), (e) Fatigue-inertia (Remaining inactive), and (f) Confusion-bewilderment (Cockerill, Nevill, & Lyons, 1991). In order to avoid confusion with the answers of athletes who might not be completely forthcoming, Morgan and co-workers developed graphical representations of the Profile of Mood States. This is called the Morgan Iceberg profile, and the subsequent graph is often constructed from trial studies where the responses from non-athletes as controls can be used as controls (Morgan et al., 1987).

### ***Over Training and A Weakening Immune System***

Hines (2001) mentioned that, college wrestlers and other athletes that over-trained were highly susceptible to disease of the immune system such as ringworm and hepatitis. Fitzgerald, (1991) asserted that there were several theories as to these observations, leading Nieman, (1994) to maintain that whether physical, physiological or societal, they are all related to the dynamics of over-training. There is no doubt that moderate exercise is recommended for people of all ages and in every physical condition (i.e., after the recommendations of a health care professional). Moderate exercise improves physical stature and self-esteem. Additionally, there is enough research to suggest that moderate exercise also strengthens the immune system, making a person less susceptible to disease (Brenner, Shek, & Shephard, 1994; Nieman, 1994; Peters-Futre, 1997; Pedersen & Hoffman-Goetz, 2000). To confound the issue, researchers have found that marathoners are at greater risk of viral infection, including cancers (Mackinnon, 1999). Researchers

have proposed that there is a J-shaped curve that links amount of exercise and immune strength (Mackinnon, 1999). The characteristics show that as exercise on the x-axis increases, the susceptibility to diseases decreases. After a certain short threshold, the susceptibility increases. This is the J-shape (Mackinnon, 1999).

The nose is the primary passageway for air entering the respiratory system. The air we breathe warms the nasal passages. Bigger particles are trapped in the nose. Smaller particles such as viruses remain suspended until they reach the bronchi. The mucus created in the endothelial layers of the nasal cavity traps the smaller suspended particles drawing them to the back of the throat or in the nasal cavity. These trapped potential pathogens are then thrown out of the body when bigger particles cause enough irritation to cause sneezing. Also coughing and expectorating small particle laden mucosa prevents the pathogens from making inroads into the pulmonary area due to extended residence in the body (Martini, Timmons & Tallitsch, 2006). When exercise levels gets particularly high the need for larger amounts of oxygen to replenish rapidly diminishing supplies and to counter the lactic acid burns, the individual exercising breathes either through the mouth or through a combination of nose and mouth. The depressed breathing through the nose tends to cool and dry the nasal passages this decreasing the amount of mucus created (Peters & Bateman, 1983). Nieman et al., (1990b) asserts that breathing through the mouth prevents the first line of defense that the nose provides, thereby allowing infectious agents access to the bronchi and other susceptible parts without interference. Upper respiratory tract infections have been reported in marathon runners, but not in those individuals running between ten or twenty kilometers. From a logical standpoint the dynamics should be just as same for marathoners and those that run shorter races

(Nieman, Johanssen, Lee, & Arabatzis, 1990a). Other confounding factors also involve the path which a long distance race takes, and the number of runners involved. Typically, marathons are popular affairs with several thousand people running. These are factors that need to be taken into account. Marathoners frequently travel to different destinations. (Peters & Bateman, 1983). The problems with air travel and different environments, which generally require acclimation, results in increased susceptibility to infection. Less controversial however, is an explanation for the J-shaped curve based on biology (Mackinnon, 1999). However, the mechanism is complicated and can involve several interrelated factors.

There are different types of white blood cells in the system. From an immunization perspective, these are divided into macrophages and Natural Killer (NK) cells (Nieman, 1994). Macrophages are large cells that engulf a pathogen in the process of denaturing it into constituent proteins that it then proceeds to destroy. The process of destruction involves presentation of the pathogen to other cytotoxic cells as antigens (Smith, 2000). Cytotoxic cells then produce antibodies specific to the antigen based on a signal from the macrophage. These antibodies are responsible for binding with the antigens rendering them ineffectual. Macrophages do not contain toxins of their own. Natural Killer cells cause lysis of an intruder cell wall, destroying the cell. Cytokines also play an important role in immune function. These are small proteins secreted in response to an infection (Smith, 2000). The behavior of cytokines is complex. There are also several different kinds of cytokines which are based on specific function (Smith, 2000). They work in cascades, which are sequential activations of chains of cytokines. This constitutes a signal. Cytokines often signal the Natural Killer (NK) cells and/or

macrophages to perform their function. Smith (2000) maintains this is why various parameters in the immune system are well-correlated. The levels of NK cells and macrophages measured before, during and after exercise (i.e., in the short and long term) has been the subject of much research. In addition, specific antibodies such as immunoglobulins (e.g., IgA) play an important role in the immune process (Smith, 2000). Lysozymes are responsible for breakdown of the intruding cell. Other white blood cells such as T cells and B cells also contribute to the body's immune system. Types of T cells include CD4 and CD8 cells (Uhaweb, 2001).

It is important to see how the above-mentioned immune supporting factors increase or decrease with exercise. Catecholamine levels are increased with increased exercise resistance. NK cell levels increase in the blood during exercises. Since NK levels have shown an increase if a subject is injected with catecholamines, it indicates that catecholamines, which are released in times of stress, trigger the NK cell response (Smith, 2000). It is possible the catecholamines fool the system into believing that the immune system is under attack during vigorous exercise. There is no evidence that shows that the production of NK cells increase with exercises, just that more of them are pushed into the circulatory system (Nieman et al., 1995). These levels fall to normal level almost as soon as the exercise routines are completed. Occasionally, NK cell-counts dip below normal. But the count value reverts to normal after a few hours. The question that remains: are the few hours during which the NK cell levels dip below normal enough to support an infection (Nieman et al., 1995). On the other hand, moderate exercises tend to increase NK levels even at resting levels. This explains why moderate exercises help bolster the immune system (Shephard, 1997).

Macrophage levels are similar to NK cells. Moderate exercise has no effect on it. Heavy exercises decrease macrophage activities. Macrophages also create a tumor necrosis factor, the substance that destroys tumor cells (Woods et al., 1993). Smith (2004) proposes that this is perhaps one explanation of the incidence of cancers among ultra marathoners and over- trainers. Prostaglandins are chemicals produced in response to trauma. These prostaglandins serve as inhibitors of both macrophages and NK cells. Microtraumas are created in response to vigorous exercises, which release prostaglandins (Smith, 2004). These in turn probably inhibit both macrophages and natural killer cells. T cells are important contributors to the immune system. CD4+ T-cells are responsible for signaling cytokines that activate macrophages and NK cells. One parameter that is considered by experts as important is the ration of T-cells CD4+ to CD8+. This ratio should be 1.5 or higher. There is evidence to suggest that this number is significantly lower following exercise (Shephard, 1997). CD4+ also signals the release of leukocytes. Leukocytes are responsible for defense against invasion of pathogens, and remove toxins, wastes, and abnormal or damaged cells (Mackinnon, 1999). With exercise this proliferation of leukocytes is also reduced. This decrease in leukocyte response to the site of infection can last for several hours. The body is particularly susceptible during this time. The response of these immunity substances also follows the J-curve. This means that most of the levels respond appropriately with moderate exercise. The depression in levels and immune response takes place only when exercise is vigorous. Shephard & Shek (1999) also assert that the mechanism for this behavior is not known. Neutrophils are known for killing bacteria. These are roving bands of white blood cells that often work in concert with macrophages. Neutrophils are also effective since their function is



an oxidative process. Research has shown the neutrophil levels increase with exercise. It is possible though, that they are not as effective with exercise because of general oxygen starvation (Shephard & Shek, 1999).

Immunoglobulins (A and G) are specific in their immune responses (Mackinnon, 1999). With moderate exercises the levels of these cytokines do not change. With vigorous exercise however, the IgA levels decrease with the level of exercises (Nieman et al., 1989). IgA found in the saliva and the nasal epithelium is reduced with increase efficiency. IgA typically suppresses the attachment of viruses to the nasal epithelium, the susceptibility to upper respiratory tract infections increases with exercise (Tharp & Barnes, 1990). The following brief summary provides a glimpse into the problems associated with respiratory problems and the kinds of physical activity. This is important because the kinds of exercises that competitive wrestlers engage in are also exercises that are associated with physical activity. These studies also indicate how the incidence of upper respiratory tract infections correlated with the type of activity and the levels of activity. Some of this activity is recreational and others are competitive activities. Yet some of the activities that will be reviewed herewith are associated with career choices. First, the results of salubrious effects: Activities that involved exercises with a varied bent at seventy percent of peak activity, three times a week at forty minutes per session caused no change in test subjects that were infected with the common cold virus (Weidner et al., 1998). In other trials participants who walked for thirty-seven minutes a day for five consecutive days actually reported a decrease of incidence of respiratory infections (Nieman et al., 1993). Nieman et. al., (1990b) studied walking trials that involved running for forty-five minutes with intensity at 60 percent of the heart reserve

rate. Walking provided an improvement in upper respiratory conditioning with no signs of infections. Karper and Boschen (1993) reported that moderate exercise three days a week reduced the number of infections.

The discussion of the biological implications of exercise as related to immune response did not provide a specific mechanism. Even so, there is no doubt as to the results. Moderate exercise improves immune response and keeps test subjects relatively healthy (Nieman et al., 1990b). A summary of the ill effects of over-training will show that immune system based disease become more rampant based on volume, frequency and resistance of exercise regimens (Douglas & Hanssen, 1978). These investigators studied sixty-one rowers and found that rowers tended to experience respiratory tract infections with degrees of severity and frequency that increased with effort in their sport. Distance running was also shown to increase infections (Shephard, Kavanagh, & Mertens, 1995). The incidences increased especially when the regimen called for running more than fifteen kilometers per week. The researchers reported findings generated from a study of almost seven hundred participants. Their results indicated that if the running distances increase significantly, so did the incidences of respiratory problems. Nieman et al., (1990b) proposed that initial military training involved typical “boot camp” like regimens. In a study of military training conducted for ninety-six Air Force cadets, the results showed that the parameters that have a bearing on the immune system showed that the immune system was generally lower. However, none of the cadets involved in the study reported any incidents of respiratory problems. Special warfare training on approximately five hundred men showed that the incidences of upper respiratory tract infections were particularly high (Nieman et al., 1990b). Even the seemingly recreational

sport that might involve vigorous activity, such as orienteering, might cause problems (Linenger, Flinn, & Thomas, 1993). A study of fifty-five males and twenty-eight women between the ages of nineteen to thirty-four conducted in 1987 showed that individuals involved in orienteering were more susceptible to illnesses than the controls. Orienteering is a sport that is similar in nature to treasure hunting. It involves using maps and other guidance devices in determining certain control points. The terrain where orienteering occurs determines the level of exercise (Peters & Bateman, 1983).

Heath et al. (1991) proposed that sometimes it is the effort that goes into the training that is also very important. Among marathoners the incidence of respiratory problems is twice that of non-marathoners. Even among competitive marathoners, (i.e., those who run faster), the incidences of infections are almost six-fold (Heath et al., 1991). The same statement may be made about ultra marathoners. A survey of those that ran 32 kilometers per week or lower had a lower incidence of symptoms of cold. When the distance over a week increased beyond seventy kilometers, more runners complained of colds (Shephard & Shek, 1999). Those athletes who run faster and expend more energy tend to be more susceptible. It is the duration of the marathon and the expended effort that makes a difference. Studies have shown that athletes who run the marathon are more susceptible than those who merely train for the marathon and utilize an equal training regime to those who actually run it (Nieman, Johanssen, Lee, & Arabatzis, 1990a). In another investigation researching respiratory problems, eighty-seven male college wrestlers and gymnasts were tested over a period of eight weeks. More than eighty-five percent of the test subjects had symptoms of respiratory problems over the period of the study (Strauss, Lanese, & Leizman, 1988).

In addition to physical and biological factors, environmental factors also play an important role in decreasing athletes' immune strengths. (Costa Rosa, Safi & Guimaraes (1996). Costa Rosa et al., (1996) stated that extremes of heat and cold, high and low ambient pressures, inhalation of polluted air and a resultant acute-phase response in the alveolar macrophages, sleep deprivation, time-zone shifts, and exposure to high and low gravitational forces can all influence immune function adversely.

In brief, this literature review thus far has discussed weight loss, over-eating and their associations with nutrition. In addition to physical, biological, psychological and environmental factors, over-training and the consequent loss of immune strength also appears to be dependent on the amount and type of nutrition. This is important in the context of this research. Over-trainers typically forsake nutrition to lose weight, and this holds true especially for college wrestlers. Over-trainers also attempt to load on nutrition in the vain hope that they can regain their weight, energy and strength. What do wrestlers try to gain? Whatever will give them an advantage. Wrestling demands muscle, suppleness and flexibility, and also the immediate burst of energy. Suppleness and flexibility can be honed by training. Muscle and energy can be obtained through nutrition.

### ***Nutrition***

Carbohydrates are the only fuel that can be directly converted into ready energy (Sherman, 1995). Simple sugars are absorbed quickly and provide instant boosts of energy. Complex sugars are useful because they enhance endurance and sustainability during highly intense wrestling events. Sherman (1995) asserts that carbohydrates, for

those that train hard, replace muscle and liver glycogen. He also recommended that those whose exercise regimens demand two hours of workouts a day should have an intake of between eight to ten grams of carbohydrates per kilogram of body weight (Sherman, 1995). Glucose concentrations, when measured during exertion, show that glucose levels remain normal at moderate exercise. These levels then fall as the resistance and volume of exercise increases. This exercise first takes up the muscle and liver glycogen (Hawley, Dennis, Lindsay, & Noakes, 1995). Prolonged vigorous exercise depletes these supplies. The exerciser then depletes blood glucose. During glycolysis, the macrophage levels decrease, as does the process of creation of lymphocytes. Additionally, the process of phagocytosis in engulfing intruding pathogens and presenting them as antigen surfaces for antibody binding consumes a large amount of glucose. Macrophage functionality is significantly decreased in the absence of adequate amounts of glucose. This in turn increases immuno-suppression (Nieman, 1997).

This current research proposes to measure cortisol levels in the saliva of a cohort of college wrestlers and compare them with RESTQ-Sports Questionnaire scores. As mentioned previously, cortisol is secreted by the adrenal glands. This secretion takes place as a response to hypothalamus-pituitary-adrenal triad activation. The path from the triad to cortisol goes via the activation of the adrenocorticotrophic hormone (ACTH). Decrease in blood glucose stimulates this pathway further, increasing the levels of cortisol. Cortisol has a suppressive effect on the purveyors of the immune system. This includes, in addition to the natural killer cells, the immunoglobulin (i.e., IgA, IgG and IgM) cytokines. Cortisol also decreases leukocyte function (Nieman, et al. 1993; Tvede, et al. 1994).

Triglyceride (TG) is the major storage form of lipids (fat) in the body. Triglyceride represents the largest source of potential energy, accounting for approximately 80% of total bodily energy stores (Gollnick & Saltin, 1988). The role of TG is a vast source of energy. TG also provides thermal insulation, serves as a cushioning protection for organs, acts as a carrier for fat-soluble vitamins, and is a structural component of many cell membranes (Gollnick & Saltin, 1988). Fats therefore have an important role to play. Measuring how much fat is in the diet is also a tall order because most foods have fat in them. There is also the struggle in trying to discern whether the fat comes from saturated poly-fatty acids or monounsaturated fatty acids (Devlin & Williams, 1991). In the presence of fat, therefore, glycolysis of glucose and glycogen might be reducing. It appears that this shift of energy source depends on the amount and concentration of fatty acids in the bloodstream (Saltin & Astrand, 1993).

Results from rats fed on a fat rich diet show that such quantities of fat then become the first source of fuel preventing the breakdown of glutamines in the muscles (Costa Rosa et al., 1996). The presence of free fatty acid molecules in the cell and plasma membrane of all cells can help regulate the functions of cells associated with immunization (Osterud et al., 1989). Monocytes are smaller leukocytes, which eventually mature into macrophages. They produce prostaglandins. Prostaglandins serve as immunosuppressors by depressing levels of T-lymphocyte and macrophage IL-2 (Osterud et al., 1989).

Protein represents a substantial source of potential energy, accounting for approximately 17% of bodily energy stores. Skeletal muscle represents the largest source of protein. Protein also has a role in the transport and regulation of the metabolism of

various fuels. Protein's structural and functionally important roles are thought to limit protein use as a primary and limiting substrate of energy production during exercise (Goodman, 1988).

Research has shown that while weight loss occurred as close to two or three days before an event, the typical numbers were approximately ten days (Tipton, 1990). Another extreme example of excessive weight loss was that of athletes, especially college wrestlers, who tried to lose 10 to 20 pounds, sometimes in one week, during the competitive season (Steen, 1989). The average weight loss was about four and one half pounds (ACSM, 1996). The main emphasis was to reduce the weight before the weigh-in. After weight loss occurred, athletes believed that they were free to increase their nutritional uptake in order to facilitate return of strength. (Clarkson, Manore, & Oppliger, 1998). As soon as an event was completed, the need to reduce weight before the next event started another cycle of rapid weight loss efforts. This cyclical process could be repeated more than ten times during a competitive season (ACSM, 1996). Albert (2000) stated that this loss of weight resulted from dehydration, loss of muscle mass, and glycogen reserves, essential components of the body. He stated that the only way to safely reduce body fat was through training.

A comprehensive study by Short and Short, (1983) of college athletes involved in several high physical intensive varsity sports showed that most athletes partook of a protein diet that met the regulations set by the World Health Organization; that athletes should consume approximately 0.8 grams of proteins per kilogram of body weight in a day. The only athletes that did not meet this criterion were wrestlers and gymnasts who were concerned with maintaining competitive weights (Short & Short, 1983). Proteins,

peptides and amino acids are the building blocks of muscles. Branched chain proteins such as leucine, isoleucine, valine, glutamine and tryptophan play an important role (Abidi, 1976). Proteinaceous foods are not difficult to obtain. Most meats, dairy products and eggs contain all the required amino acids. For vegetarians, though not a complete diet, grains and legumes provide a good source of proteins. Long-term deficiencies of protein in the diet can result in the loss of muscle mass, muscle strength and muscle endurance (Shephard, 1997).

Proteins are not extensively metabolized and are often thrown out of the body as waste and urea (Shephard, 1997). Since proteins are not assimilated in the system, the full feeling of having eaten a tasty protein-rich diet without the excess carbohydrates that can be stored as glycogen and then synthesized into fat in the liver does not arise. The most common mistake that most people make is that the itinerant fat with some of these proteinaceous foods can be counterproductive to the positive effects of a protein diet (Butterfield, 1987). The Atkins diet fails to recognize that since carbohydrates are first burned for those with a sedentary lifestyle, the energy sources are soon depleted and muscle glycogen begins to breakdown. This discussion is important because it is not out of the ordinary for wrestlers to partake of Atkins diet to lose weight without recognizing the full impact of the consequences (Barnard, 2004).

From an immunity perspective, glutamine is very important. It is used in the synthesis of RNA and DNA (Newsholme, 1994). It is the most abundant protein in muscle and plasma. Glutamine is used as an energy source during lymphocyte proliferation and also during macrophage function of phagocytosis (Newsholme & Parry-Billings, 1990). Glutamine concentrations are directly correlated with the activity of NK



cells (Newsholme & Parry-Billings, 1990). NK cells are really a type of lymphocyte. They are distinguished from T-cells or B-cells. Their primary function is to kill cells. They receive a signal from cells that have either been infected by a virus or are otherwise cancerous. Experiments in rats have shown that the production of IL-1 and phagocytic activity by macrophages is sustained if the protein in the diet is between twenty and forty percent and impaired if the protein levels fall. Lack of protein in the diet has a detrimental effect on T-cell activity. Kingsbury et al., (1998) demonstrated that glutamine levels were restored in athletes who were given a protein rich diet. The levels prior to the diet were below normal. In another study, test subjects who rode an exercise bicycle until exhaustion were given a dextrose solution or a dextrose solution along with a protein additive. They found that glutamine levels were low by almost twenty percent following the exercise and also a while after the exercise was concluded. This drop in glutamine levels was sustained. The role of glutamine in preventing infections was no more common than in a study of ultra marathoners. The test set was given a mixture of glutamine in water after the race. A control group was given a maltodextrose solution. For a week after the event, the test group did not suffer from any infections when compared to the control group (Kingsbury, Kay, & Hjelm, 1998).

The problems with protein deficiencies have been discussed. There are also problems with over use of proteins. Glutamine levels have to be maintained at sustainable levels (Parry-Billings et al., 1992). Excess imbibing of this amino acid will result in the loss of muscle and plasma levels. This means that the excess is excreted through the renal system, a result as the body tries to balance normal levels. It is also possible that the liver uses the excess glutamine in the conversion of glucose to glycogen (Antonio & Street,

1999). This result was seen based on diet administered to test subjects that was rich in protein, fat, and that contained very low amounts of carbohydrates. Carbohydrates should be the first energy providers. A study found that a low carbohydrate diet resulted in lowering of glutamine levels (Bjorkman & Wahren, 1988).

Vitamins and supplements play an important role in the immune system, especially with reference to training and exercise (Bendich & Chandra, 1990). Vitamin A is otherwise known as retinol. Deficiencies in retinol result in the impairment of IgA production (Bendich & Chandra, 1990). It results in the breakdown in the proliferation of mitoses induced lymphocytes. Lower levels of retinol allow the binding of bacteria to respiratory epithelial cells (Beisel, Edelman, Nauss, & Suskind, 1981). On the other hand, beta-carotene supplements enhance the activity of natural killer cells as they advance the proliferation of lymphocytes. With beta-carotenes, the CD4+ T-cells increase in number. While deficiencies of retinol have created the above-mentioned problems, excess Vitamin A results in liver problems, nausea, bone disorder and joint pain (Colgan, 1986). Vitamin B-12, cyanocobalamin, and Vitamin B (Folic Acid) can be used in the immune system because they contribute to the production of both red and white blood cells (Colgan, 1986). Vitamin C, (Ascorbic acid) helps the immune system in a two-pronged fashion (Nieman et al., 1990a). First, is used in the proliferation of T-cell lymphocytes; second, it can be used to suppress glucocorticoids, which themselves are involved in suppressing immune function. Ascorbic acid is important because it beefs up resistance to pathogens that might cause upper respiratory tract infections. Nieman et al. (1990a) also recommended six hundred milligrams of this vitamin C to prevent cold and other pulmonary diseases, especially for athletes like wrestlers and marathoners. The best

results were obtained for marathoners who were provided with a cocktail of ascorbic acid, tocopherol and beta-carotenes. Colgan (1986) stated that Tocopherol, otherwise known as Vitamin E, is implicated in the maintenance of well-being and vitality. Ascorbic acid is lost in the sweat and urine after a particularly strenuous workout and is also taken up by lymphocytes from the plasma (Colgan, 1986).

Chandra (1991) maintains that minerals play an important role on the general well-being of individuals by enhancing the immune system. Several minerals are well-known. They are called macronutrients or micronutrients depending on the amounts present in the body. Minerals exist and function in the body in only trace amounts. Iron, selenium, iodine, copper, zinc and magnesium have their own roles to play. Iron and zinc are well studied and their roles in helping the immune system have been uniquely identified. Chandra (1991) adds that meat and seafood are rich source of zinc. Zinc is important because it chelates with proteins resulting in metalloenzymes, which have myriad functions. Zinc is a co-enzyme with a protein that is responsible for the growth, maturity and replication of T-cell lymphocytes. Deficiencies of zinc in the diet results lower production of IL-2 and the proliferation of lymphocytes. Chandra (1991) also maintains that zinc is lost during exercise and training. This means that over-training will result in the loss of nutritive and immune value of zinc. The loss of zinc is indicated in athletes whose numbers in almost all trials are significantly lesser than those for the controls, non athletic participants in the studies. Iron depletion is more commonplace among athletes. He adds that statistics have revealed that a large segment of the human population is unknowingly deficient in iron. Like some vitamins and nutrients, iron also

helps lymphocyte proliferation, macrophage production of IL-1 and the activity of natural killer cells (Chandra, 1991).

With over-training comes the loss of iron through sweat. Up to one milligram a day of iron can be lost through exercise. The notion of foot strike occurs in long distance runners (Ogino et al., 1994). Iron breakdown occurs during long distance running and hemoglobin breakdown can occur as well. This is seen through red coloration in the urine. Selenium and copper impact the immune system in myriad and smaller ways, some of which have already been discussed above (Williams, 1999).

### ***Supporting Evidence for Proposal***

IgA is a protein of the family of immunoglobulins. This immune protein antibody attacks and destroys viruses that seek to enter the body through the mouth. It therefore serves as the first line of defense against illnesses of the respiratory track especially in athletes (Tomasi &Plaut, 1985). This has been discussed in detail above. A comprehensive study of twenty-six elite swimmers in Australia, by merely identifying IgA levels in the saliva, showed that there was an inverse correlation (Gleeson et al., 1999). A difficult training regimen, combined with lower levels of IgA, induced a higher susceptibility to disease. In fact, Gleeson et al., (1999) stated that if the IgA levels fell by six percent, there was a greater chance of the athlete complaining of an infection. In developing a model for training and IgA levels, Gleeson et al., (1999) determined that “preseason IgA levels” were not as good as when pre-training IgA levels were determined. Pre-training is the time just before a training regimen. Interestingly, however, IgA levels in the saliva can be used with even greater specificity. This is

especially useful when over training and training schedules are determined (Mackinnon et al., 1989). There is a general consensus that IgA levels are fine tuned during the course of the day. These are akin to other physiological and psychological factors. This means that it is possible to take IgA measurements at different times of the day and determine at what time the IgA is likely to be the lowest and the susceptibility to an infection is the greatest (Dimitriou, Sharp, & Doherty, 2002).

A comprehensive study in Britain among swimmers was conducted to find out if the IgA levels followed a circadian rhythm. Trials were conducted with appropriate warm up and swimming test phases. The usual precautions involved not eating, drinking, smoking prior or participating in sex at certain predefined times prior to the test. The test was conducted at six am and at six pm. Prior to the test, psychological profiles were drawn for the participants. These were used to determine whether they were emotionally better disposed towards testing in the morning or evening. Saliva IgA levels were measured soon after the prescribed swimming regimen. The results indicated that IgA levels were highest in the evening. This indicated that these times were good for swimming training. This meant that the immune system was stronger at this time and less susceptible to infections (Dimitriou, Sharp, & Doherty, 2002). The levels of cortisol were inversely correlated with IgA levels. Cortisol is an immunosuppressant. It also promotes gluconeogenesis. This means that it synthesizes glucose from fats and proteins. Since it stimulates the appetite it performs a dual role. Cortisol levels increased after repetitions of 400-meter swims by the participants. All the participants in this test were competent swimmers who swam at either the local or the national levels. Another factor that was measured in this case was the salivary flow levels. This is the amount of saliva generated.

From the point of view of this test, this determines how the immune properties of IgA are put into action. This salivary flow was highest during the evening test (Dimitriou, Sharp, & Doherty, 2002).

Cortisol is a small lipophilic steroid of molecular weight approximately 362 daltons. It is produced in the adrenal glands when the adrenocorticotrophic hormone is attached to membranes receptors on the adrenal cortex. Following this pathway, cortisol is released into the blood stream. Most of cortisol is bound to proteins and enzymes. A very small amount of cortisol is free flowing. Bodily fluids such as tears, urine, blood and saliva contain most of the free cortisol. A salivette is used to obtain between 0.5 to 1.5 mL of saliva. This saliva can be stored at room temperature for up to a month. However, because of the formation of mold, it is stored in the refrigerator. This storage does not affect cortisol levels. In order for cortisol levels to be true reflections of the amounts, salivation should not be stimulated. The salivette comes equipped with a wad of cotton. Chewing on the cotton provides the requisite sample of saliva in under a minute (appendix H).

Cytokines are of several types: Lymphokines, chemokines, monokines, interleukins. Cytokines work in cascades. They are created de novo, in response to interference. They subsequently bind with membranes through the process of signal transduction acting on a cell. There are different cytokines depending on the types of cells that produce them. For example, IL1 alpha and IL-1 beta are produced by monocytes, macrophages and B cells. Natural killer cells and Th cells produce IL-3. Th1 cells produce IL-2. These are a few among many. Different cells might give rise to one type of cytokine. Different cytokines might also be produced by one cell, depending on

the stimulus. They are associated with the immune system in the proliferation of lymphocytes, enhancing phagocytic action, and the secretion of antibodies. All of these processes are geared towards protection of the body from ailments (Uhaweb, 2001).

### *Summary*

This literature review illustrates the necessity of addressing the practice of over-training among college athletes, especially college wrestlers. College wrestling demands strength, balance, suppleness, and weight management from its proponents. In attempting to meet these demands, an athlete's lack of understanding of the dangers inherent in over-training can be deleterious to his physical and emotional health. It is incumbent, therefore, that further research as outlined in the preamble to this proposal be conducted, and that a set of informational and educational guidelines/requirements be drawn up for everybody involved with the sport.

In the literature it has been shown that athletes will deny themselves adequate nutrition and hydration after workouts in order to make or maintain a certain weight for weight classification competition (Albert, 2000; Shireffs & Maughn, 1998). Over-training is used to decrease weight by dehydrating the body during workouts and also, by not properly rehydrating after the workout ends (Alderman et al., 2004; Luttermoser, Gochenour & Shaughnessy, 1999; Shireffs & Maughn, 1998). Athletes involved in heavy periods of training need to make sure that they are consuming enough carbohydrates and calories so as not to catabolize amino acids from muscle as a fuel source for the exercise (Hines, 2000). Athletes think that they can compensate for the lack of nutrition by binge eating after weight certification for the event has happened, but it has been shown that

this is counter productive (Alderman et al., 2004). The literature has shown that over-training is used to decrease weight by dehydrating the body during workouts and also, by not properly rehydrating after the workout ends. It is this practice that can cause the body to begin the break down of functioning properly (Alderman et al., 2004; Davis, 2004; Perriello et al., 1995).

In order to optimize athletic performance, athletes must be optimally trained. Athletes who under train may not perform to their potential, where as athletes who train too often or too intensely may experience negative training adaptations and decreased performance capacity (Kreider, Fry & O'Toole, 1998; Lehman et al., 1992; Kibler et al., 1989). The symptoms of over-training considered are: the decreased ability to perform in terms of strength, power, muscle endurance, cardiovascular endurance, and flexibility (Fry & Kraemer, 1997; Nijs, Vanhergerghen, Duquet & de Meirleir, 2004). There are also physiological markers of interests, parasympathetic and sympathetic responses to training and under-recovery (Sherman et al., 1981; Helge, Richter & Kiens, 1996; Roberts, 1986; Fitzgerald, 1991). These symptoms are physical and motor characteristics.

There are injuries that are associated with over-training. The response to female athletes that over-train has been demonstrated to preclude them stress fractures, amenorrhea, and eating disorders (Dueck et al., 1996a; Dueck, Manore & Matt, 1996b; Nattiv et al., 1994). These symptoms are associated with training and the result hampers any athletic effort. Injuries to joints and muscles tend to have both short and long term damage (Kibler et al., 1989; Hoaglund, Yau & Wong, 1973). By combining the effects of restricting fluids, nutrition and increased work load the joints can eventually give rise to problem of chronic osteo-arthritis (Sherman et al., 1981; Helge, Richter & Kiens, 1996).



Intense training places a demand on the musculoskeletal system that may lead to over clinical damage as well as functional and biomechanical adaptations that may be detrimental to sport performance (Steptoe, Kearsley & Walters, 1993; Orth, Kovacs & Debold, 1992).

The psychological signs are effects on the nervous system that may have effects on the body systems. The athlete looks at exercise as the way to make themselves feel good (Garner, Olmstead, Bohr & Garfinkel, 1982; Holloway & Baechle, 1990). Like the runners high that is perceived during a particular good workout, the athlete strives to feel that sense of “good feeling” again during each workout and will continue to work until that feeling is attained (Blumenthal, O’Toole & Chang, 1984; Yates et al., 1992). The parameter seems to be agreed that this may be the need for perfectionism.

The weakening of the immune system plays a role in over-training. As the athlete exercises and continues to workout no matter what the condition they continue to compromise their immune system and how it functions (Hines, 2001; Fitzgerald, 1991; Nieman, 1994). As the immune system becomes compromised the incidence of infection arises and the length of time to recover becomes lengthened as well (Mackinnon, 1999; Shephard, 1997; Shephard & Shek, 1999). Athletes, coaches, and athletic trainers want to keep athletes healthy during training and competition. They have long believed that athletes are susceptible to illness, in particular upper respiratory tract infection, during long periods of intense training and after major competition (Tharp & Barnes, 1990; Weidner et al., 1998; Nieman et al., 1993; Nieman et al., 1989).

## **CHAPTER III**

### **METHODOLOGY**

The primary purpose of this study was to determine if there was a correlation between the physiological signs of over-training and the psychological signs of over-training in collegiate wrestlers. Three research questions were developed:

1. What is the relationship between salivary cortisol and recovery-stress for all wrestlers combined?
2. What is the relationship between salivary cortisol and recovery-stress for each wrestler, individually, as they represented different weight classes?
3. Did salivary cortisol and recovery-stress scores significantly change during the course of the 12-week training?

The subsequent sections will discuss the following: a) selection of subjects, b) selection of instruments and markers, c) procedures, and d) research design and statistical analysis.

#### ***Selection of Subjects***

The data were gathered from wrestlers competing at an institution recognized by the National Collegiate Athletic Association (NCAA). These athletes qualified by competing with other wrestlers in each weight class to win the varsity spot within a

particular weight class. The subjects were enrolled in classes in a NCAA institution that participated in NCAA wrestling. The subjects were varsity wrestlers at a NCAA institution that competed against other NCAA institutions, and participated in the adherence to NCAA guidelines, NCAA Wrestling Rules and Interpretations (2007). A total of 10 subjects, one representing each weight, were selected to participate in this investigation based upon status as “varsity”. The subjects’ ages ranged from 18-25 years. One person from each weight class was selected for this study based upon status as “varsity”. Weight classification was determined by the rules of competition in the NCAA (NCAA, (2007), Part 1, Rule 3, Section 2].

### ***Selection of Instrument and Biological Marker***

#### ***Recovery-Stress Questionnaire for Athletes (RESTQ-Sport).***

The Recovery-Stress Questionnaire for Athletes (RESTQ-Sport) (appendix g) is an instrument that systematically reveals the recovery-stress states of athletes. The specific characteristics of the RESTQ-Sport Questionnaire allows systematic and direct measurement of appraised events, states, and activities regarding their frequency while simultaneously considering stress and recovery processes. The RESTQ-Sport Questionnaire contains 4 subcategories that reflect the stress and recovery of each individual. The subcategory General Stress contains; general stress, emotional stress, social stress, conflicts/pressure, fatigue, lack of energy, and physical complaints. The subcategory General Recovery contains; success, social recovery, physical recovery, general well-being, and sleep quality. The subcategory Sports Specific Stress contains; disturbed breaks, emotional exhaustion, and injury. The subcategory Sports Specific

Recovery contains; being in shape, personal accomplishment, self-efficacy, and self-regulation. The recovery-stress state indicates the extent to which persons are physically and/or mentally stressed, whether or not they are capable of using individual strategies for recovery, and which strategies are used to reduce stress (Kellman & Kallus 1996). The RESTQ-Sport Questionnaire was developed to measure the frequency of current stress along with the frequency of recovery-associated activities. The current recovery-stress state depends on preceding stress and recovery activities. Through the simultaneous assessment of stress and recovery, a differentiated picture of the current recovery-stress state can be provided (Kellman & Kallus, 1996). The RESTQ-Sport Questionnaire reflects a test-retest reliability of  $r = .79$ , which implies that intra-individual differences in the recovery-stress states can be well reproduced (Kellman & Kallus, 1996). The RESTQ-Sport Questionnaire has been validated against a broad range of criteria, which cover psychological measures such as mood state, performance, and biological state indicators (Kellman & Kallus, 2001). The instrument was selected as a measurement tool for this study because it is widely used and reliable for understanding the recovery-stress states of athletes.

#### *Cortisol.*

Cortisol is a small lipophilic steroid of molecular weight approximately 362 daltons. It is produced in the adrenal glands when the adrenocorticotrophic hormone is attached to membrane receptors on the adrenal cortex. Cortisol is then released into the blood stream. Most of cortisol is bound to proteins and enzymes. A very small amount of cortisol is free flowing. Bodily fluids such as tears, urine, blood and saliva contain most of the free cortisol. Cortisol stimulates proteins breakdown to amino acids in all cells of

the body except the liver. These liberated amino acids circulate to the liver where they are synthesized to glucose via gluconeogenesis (McArdle, Katch & Katch, 2006). Cortisol secretion also accelerates lipid mobilization for energy. This is especially apparent during starvation and exercise. There is considerable variability in the cortisol response to exercise, depending on such factors as exercise intensity and duration, fitness level, nutritional status, and even circadian rhythm (McArdle, Katch & Katch, 2006).

Cortisol was chosen as the biomarker for this study because it is a marker of distress for both theoretical and empirical reasons. It has been shown that the hypothalamic-pituitary-adrenal (HPA) axis is excessively activated in depressive's states (Kalin & Dawson, 1986), and Morgan et al. (1987) observed that 80% of stale swimmers possess depressions of clinical significance. Accordingly, staleness should be characterized by HPA axis dysregulation which would theoretically include elevations in resting cortisol levels (Morgan et al. 1989).

### ***Procedures***

Upon approval from the Oklahoma State University Institutional Review Board, participants for this study were selected from a convenience sample by rank (i.e., weight class) and status (i.e., varsity vs. reserve) on the varsity wrestling team. The two measuring tools used in this study were the Recovery-Stress Questionnaire for Athletes (RESTQ-Sport) and salivary cortisol testing. The RESTQ-Sport Questionnaire was used to evaluate the subjects' psychological recovery status. The salivary cortisol testing was used to measure the physiological response to training and recovery.

Prior to data collection, participants were required to have qualified to compete for a varsity position on the college or university wrestling team. If consent was given and each participant qualified to compete on the varsity team, he was then eligible to become a subject. The researcher then instructed each participant on the proper technique for answering the questionnaire, and submitting a saliva sample based upon established protocols from Kellman & Kallus authors of the questionnaire and Salimetrics (appendix G) process to measure salivary cortisol testing. Each subject completed the Recovery-Stress Questionnaire for Athletes, and provided a saliva sample. The data collection took 13 weeks to gather the information pertinent for this study. The collection time was conducted at weekly intervals to add sensitivity based upon previous studies and time points. The data was collected in the same room with each subject producing a saliva sample and completing the RESTQ-Sport Questionnaire weekly in the afternoon prior to practice beginning.

The Recovery-Stress Questionnaire for Athletes was administered each time a saliva sample was obtained. The subjects were given the questionnaire and seated at a desk where they responded to questions in the questionnaire.

The saliva collection protocol was followed in accordance with the guidelines prescribed by Salimetrics. Each subject was given a cup of water, and then instructed to thoroughly rinse his mouth by swishing one mouthful of water in the attempt to cleanse the teeth and mouth cavity of unwanted materials. The subjects were provided a 2 ml tube labeled with the subjects' identifying numbers. The subjects slowly expectorated 2 ml of un-stimulated saliva. This process took approximately 10 minutes. The samples were placed upon ice until they were delivered to Cytovance Biologics LLC for testing. The

samples were then placed in a -20 freezer until they were thawed for testing. Once thawed the samples were centrifuge for clarifying, and spun for 10 minutes at 3300 rpm at 4 degrees C. Once centrifuged, the samples were carefully removed from the centrifuge, and the testing protocol established by Salimetrics high sensitivity salivary cortisol enzyme immunoassay kit was strictly followed (appendix H). Cytovance Biologics is a biochemical lab that produces proteins and provides testing services. Cytovance Biologics provided the results of the salivary cortisol levels (appendix F).

### ***Research Design and Statistical Analysis***

The research design for this study was exploratory, descriptive, and correlational. The researcher utilized the Statistical Package for the Social Sciences (SPSS 14.0 for Windows) to complete all statistical analysis. The physiological marker (cortisol) and psychological marker (rest-recovery score) of the college wrestlers served as the variables of interest.

## CHAPTER IV

### RESULTS AND DISCUSSION

#### *Introduction*

This chapter reports the descriptive statistics of the subjects followed by a detailed statistical analysis of the variables of interest in the male college wrestlers included in this study. The first goal of this study was to measure the response of salivary cortisol and stress and recovery scores from the RESTQ-Sport in wrestlers competing in a college wrestling program. The second goal was to compare salivary cortisol and recovery scores week by week within weight class, as well as between weight classes. Subjects were grouped by weight class based upon a standardized competition weight classification (NCAA, 2007). One outlying score appeared when the subjects reported cortisol levels. This score, an extremely high level of cortisol, was investigated in order to determine the cause. Results of the investigation determined that the cause of the outlying score was due to the subject's continued training before sample collection which produced an extremely high level of cortisol. This cortisol score was subsequently eliminated, and an average of all cortisol scores for that specific subject was used in place of the original score.



### ***Descriptive Statistics***

Descriptive information for all wrestlers was collected during physical examinations conducted at University of Central Oklahoma. A total of 10 subjects volunteered for the study. In order to maintain weight during competition, and to meet necessary weight requirements pursuant to qualification for competition, the wrestlers in this study continued to train and compete during the course of the investigation. The mean values and standard deviations for the wrestlers for age (AGE) were  $20.68 \pm 1.29$  years. The mean values and standard deviations for the wrestlers weight (WT), weight to lose to reach competition weight (WTLOSS), body fat percentage (BODYFAT), and height (HEIGHT) were  $185.9 \pm 48.16$  pounds,  $14.90 \pm 3.21$  pounds,  $15.35 \pm 6.80$  percent,  $70.20 \pm 2.15$  inches are indicated in Table 4-1.

**Table 4-1**  
**Descriptive Statistics of Wrestlers**

	N	Mean	Std. Deviation
AGE	10	20.7000	1.2517
WT	10	185.9000	48.1605
WTLOSS	10	14.9000	3.2128
BODYFAT	10	15.3550	6.8012
HEIGHT	10	70.2000	2.1499
Valid N (listwise)	10		

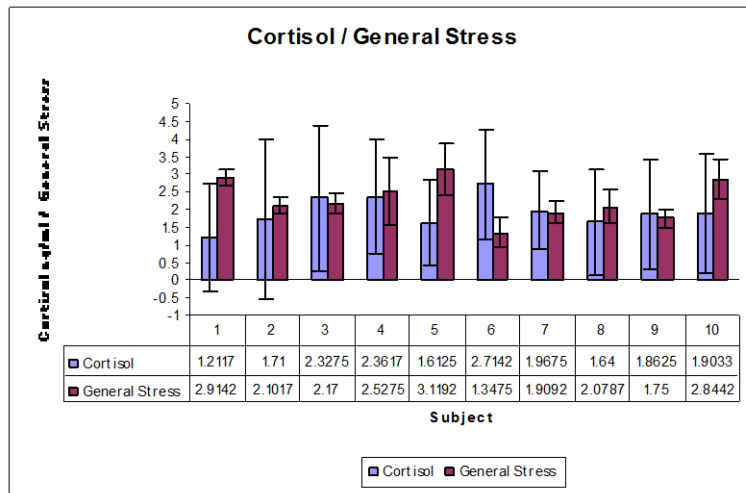
Plots of the descriptive indices for salivary cortisol and each of the stress recovery variables for individual wrestlers are presented next to allow for a direct comparison of scores. These data are presented in graphs 4-1 through 4-4. The graphs show the mean cortisol levels along with standard deviations values, reported in ng/ml. Stress recovery scores were determined based upon the results of the RESTQ-Sport Questionnaire. The 4 subcategories of the RESTQ-Sport Questionnaire used in the current study were as

follows: a) General Stress, b) General Recovery, c) Sports Specific Stress, and d) Sports Specific Recovery. In other words, four categories of stress and recovery were explored in this study, along with cortisol levels, for these wrestlers.

*Salivary Cortisol and General Stress*

The following plot (Graph4-1) shows means and standard deviations for the subcategory general stress and salivary cortisol for all ten wrestlers.

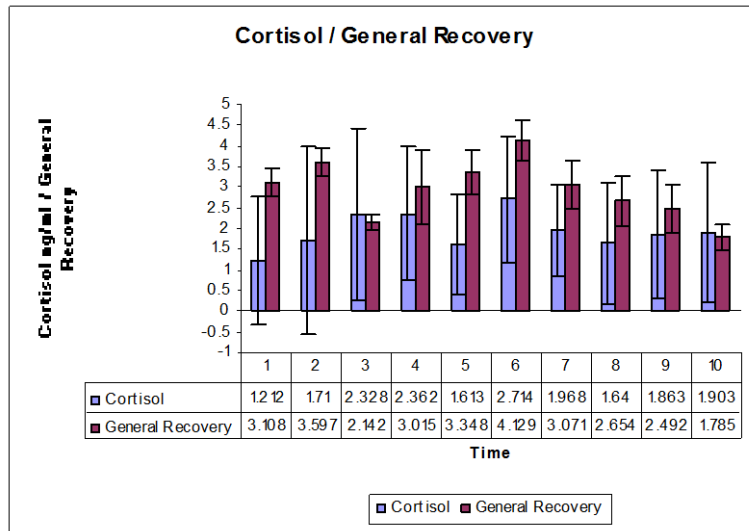
**Graph 4-1**  
**Salivary Cortisol and General Stress for Individual Wrestlers**



*Salivary Cortisol and General Recovery*

Graph 4-2 reflects means, standard deviations for the subcategory general recovery, and salivary cortisol for all ten wrestlers.

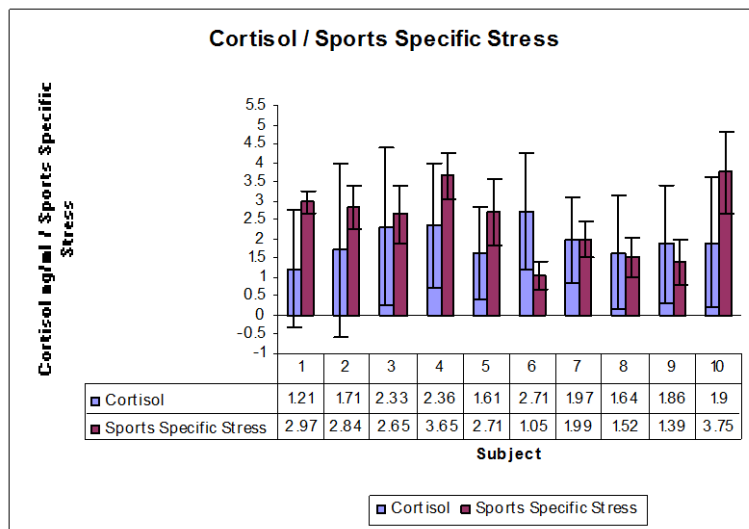
**Graph 4-2**  
**Salivary Cortisol and General Recovery for Individual Wrestlers**



*Salivary Cortisol and Sports Specific Stress*

Shown below is the Graph 4-3 depicting means and standard deviations for the subcategory sports specific stress, and salivary cortisol for all ten wrestlers.

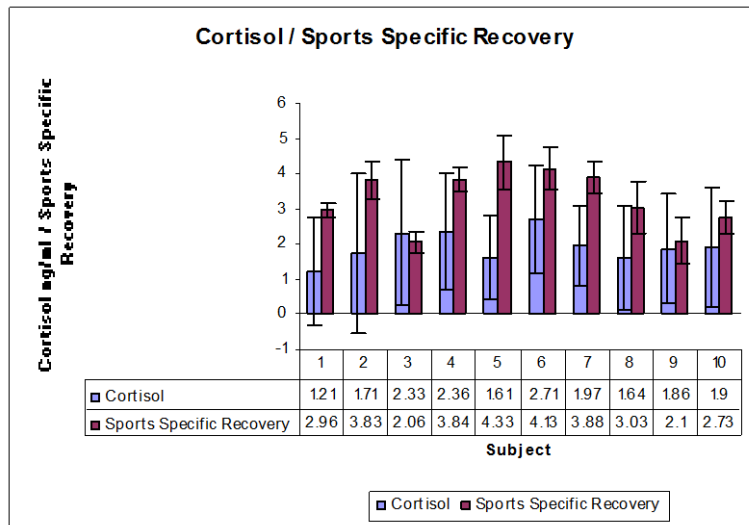
**Graph 4-3**  
**Salivary Cortisol and Sports Specific Stress for Individual Wrestlers**



*Salivary Cortisol and Sports Specific Recovery*

Graph 4-4 presents means and standard deviations for the subcategory sports specific recovery, and salivary cortisol for all ten wrestlers.

**Graph 4-4**  
**Salivary Cortisol and Sports Specific Recovery**



Graphs 4-1 through 4-4 illustrate the wrestlers’ means and standard deviations for salivary cortisol and the individual subcategories of the RESTQ-Sport Questionnaire. These figures provide data useful in comparing the wrestlers, who varied by weight class. These descriptive statistics allow for a visual comparison of the average cortisol and stress recovery scores for this sample of wrestlers.

***Research Question 1***

The following research question was addressed: What is the relationship between salivary cortisol and recovery-stress for all wrestlers combined? The cortisol and stress recovery variables for all wrestlers (N = 10) were explored via Pearson product moment correlations. The calculated coefficient between cortisol and general stress ( $r = 0.248$ ,  $p = .490$ ) revealed a lack of association between these two variables. Further, statistically

non-significant correlations were uncovered between salivary cortisol and general recovery ( $r = 0.159$ ,  $p = .661$ ), between cortisol and sports specific stress ( $r = 0.153$ ,  $p = .673$ ), and between cortisol and sports specific recovery ( $r = -0.119$ ,  $p = .744$ ). Means standard deviations, and the correlational indices are presented in Table 4-2.

**Table 4-2**  
**Salivary Cortisol Stress Recovery Scores for All Wrestlers (N=10)**

	Mean	Std. Deviation	N
Cortisol ng/ml	2.8336	1.4007	10
General Stress	27.3133	6.7621	10
General Recovery	35.2091	8.3075	10
Sports Specific Stress	29.4210	11.1790	10
Sports Specific Recovery	39.4565	9.9287	10

		Cortisol ng/ml	General Stress	General Recovery	Sports Specific Stress	Sports Specific Recovery
Cortisol ng/ml	Pearson Correlation	1.000	.248	.159	.153	-.119
	Sig. (2-tailed)	.	.490	.661	.673	.744
	N	10	10	10	10	10
General Stress	Pearson Correlation	.248	1.000	-.306	.787**	.049
	Sig. (2-tailed)	.490	.	.389	.007	.892
	N	10	10	10	10	10
General Recovery	Pearson Correlation	.159	-.306	1.000	-.363	.791**
	Sig. (2-tailed)	.661	.389	.	.303	.006
	N	10	10	10	10	10
Sports Specific Stress	Pearson Correlation	.153	.787**	-.363	1.000	.008
	Sig. (2-tailed)	.673	.007	.303	.	.982
	N	10	10	10	10	10
Sports Specific Recovery	Pearson Correlation	-.119	.049	.791**	.008	1.000
	Sig. (2-tailed)	.744	.892	.006	.982	.
	N	10	10	10	10	10

\*\* Correlation is significant at the 0.01 level (2-tailed).

### ***Research Question 2***

What is the relationship between salivary cortisol and recovery-stress for each wrestler, individually, as they represented different weight classes?

*Subject 1*

Table 4-3 presents the overall average cortisol and recovery-stress scores for Subject 1 over the 12 week training session.

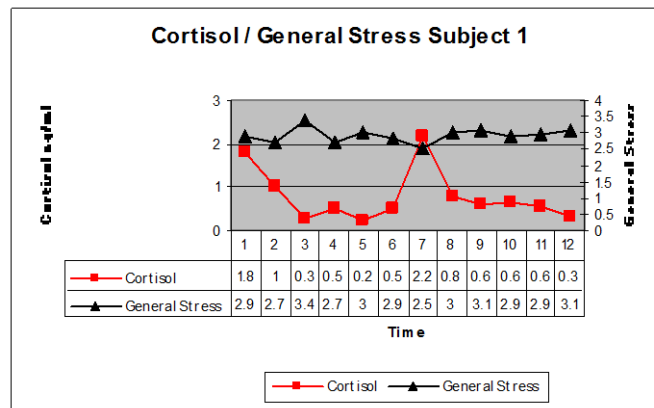
**Table 4-3**  
**Subject 1 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	1.2117	1.5503	12
General Stress	2.9142	.2248	12
General Recovery	3.1083	.3496	12
Sports Specific Stress	2.9717	.2972	12
Sports Specific Recovery	2.9592	.1757	12

The individual's scores, plotted week-by-week across the 12 training sessions, are shown in the graphs that follow. Four graphs are presented (Graphs 4-5 through 4-8), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

Graph 4-5 presents cortisol and general stress for subject 1. The data in this graph

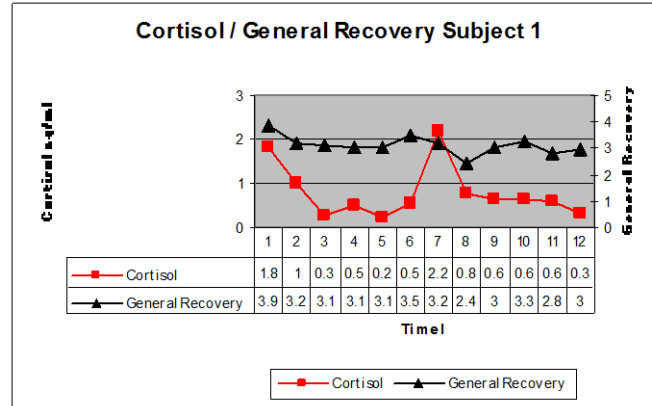
**Graph 4-5**



were correlated, to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = 0.291$ ;  $p = .375$ ) suggested no significant association between these two variables. Correlational analysis of the

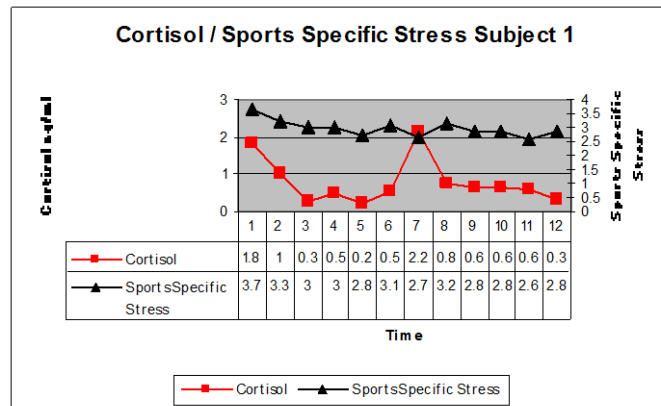
subject's data in graph 4-6 indicated no real relationship existed between salivary cortisol and general recovery ( $r_{(10)} = 0.291$ ;  $p = .359$ ).

**Graph 4-6**



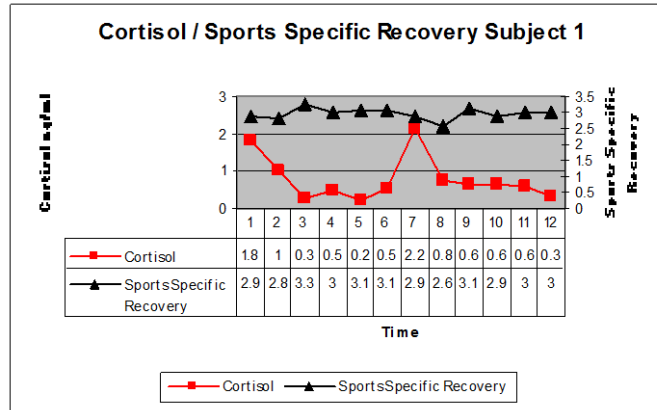
The data plotted in graph 4-7 were correlated, also resulting in a non-significant coefficient ( $r_{(10)} = -0.21$ ;  $p = .949$ ) between salivary cortisol and sports specific stress. Graph 4-7 presents this wrestler's cortisol and sports specific stress scores. Finally, the

**Graph 4-7**



data in graph 4-8 were plotted to show the relationship between salivary cortisol and sports specific recovery. The correlation coefficient calculated for these data revealed no significant relationship between these two variables for subject 1 ( $r_{(10)} = 0.303$ ;  $p = .339$ ).

**Graph 4-8**



*Subject 2*

Table 4-4 presents the overall average cortisol and recovery-stress scores for Subject 2 over the 12 week training session.

**Table 4-4**  
**Subject 2 Salivary Cortisol Stress Recovery Scores**

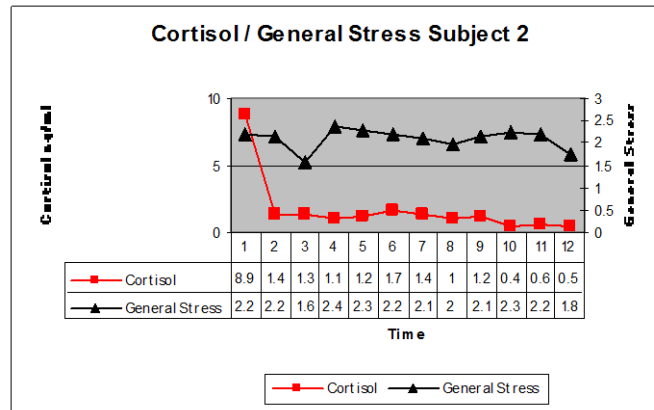
	Mean	Std. Deviation	N
Cortisol ng/ml	1.7100	2.2811	12
General Stress	2.1017	.2338	12
General Recovery	3.5967	.3258	12
Sports Specific Stress	2.8358	.5803	12
Sports Specific Recovery	3.8342	.5414	12

The individual’s scores, plotted week-by-week across the 12 training sessions, are shown in the graphs that follow. Four graphs are presented (Graphs 4-9 through 4-12), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

Subject 2 cortisol and general stress are shown in graph 4-9. The data in this

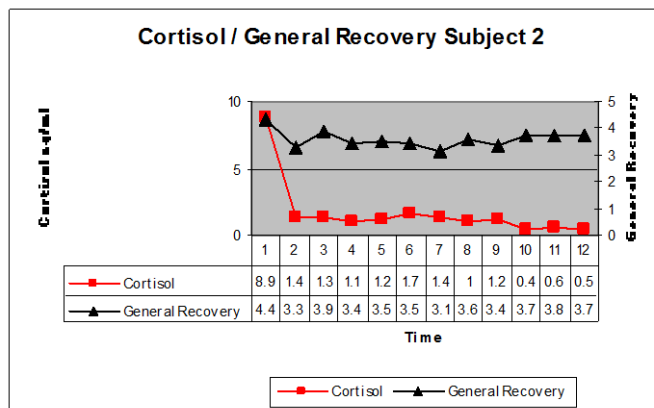


**Graph 4-9**



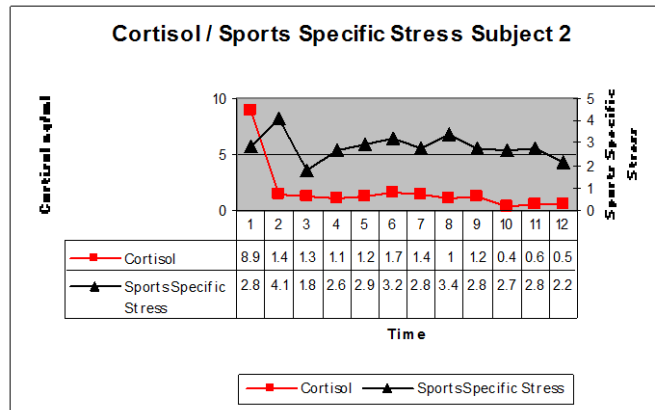
graph were correlated, to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = 0.148$ ;  $p = 0.646$ ) suggested no significant associated between these two variables. Correlational analysis of the subject's data in graph 4-10 indicates that there appears to be a relationship between salivary cortisol and general recovery ( $r_{(10)} = 0.655$ ;  $p = .021$ ).

**Graph 4-10**



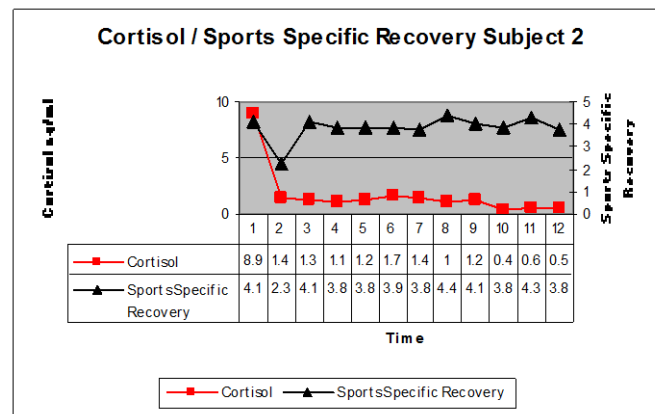
The data plotted in 4-11 were correlated, also resulting in a non-significant coefficient ( $r_{(10)} = 0.048$ ;  $p = 0.882$ ) between salivary cortisol and sports specific stress. Finally, the

**Graph 4-11**



data in graph 4-12 were plotted to show the relationship between salivary cortisol sports specific recovery. The correlation coefficient calculated for these data revealed no significant relationship between these two variables for subject 2 ( $r_{(10)} = 0.133$ ;  $p = 0.680$ ).

**Graph 4-12**



*Subject 3*

Table 4-5 presents average cortisol and recovery-stress scores overall for Subject 3 over the 12 week training session.

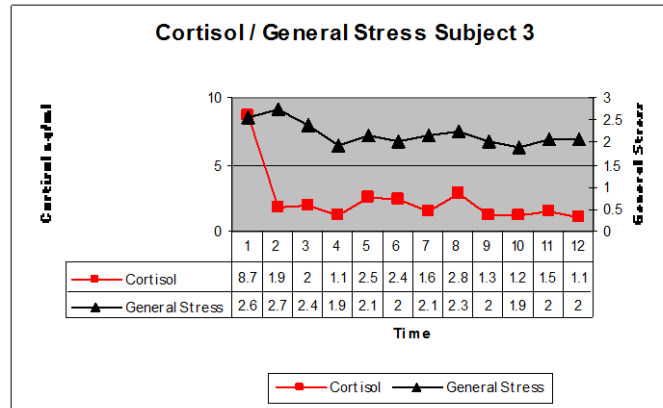
**Table 4-5**  
**Subject 3 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	2.3275	2.0754	12
General Stress	2.1700	.2596	12
General Recovery	2.1417	.2076	12
Sports Specific Stress	2.6525	.7642	12
Sports Specific Recovery	2.0575	.3071	12

The scores for subject 3 were plotted across the 12 training session's week-by-week, and are shown in the graphs that follow. Four graphs are presented (Graphs 4-13 through 4-16), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

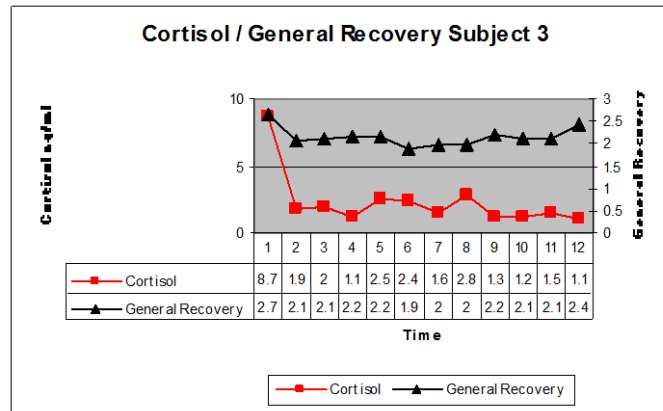
The cortisol and general stress for subject 3 are present in graph 4-13. The data in

**Graph 4-13**



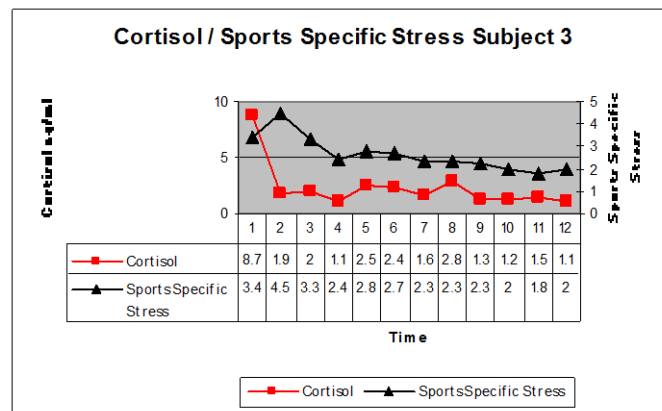
this graph were correlated, to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = 0.564$ ;  $p = 0.056$ ) suggested no significant association between these two variables. Graph 4-14 indicates a significant relationship between salivary cortisol and general recovery ( $r_{(10)} = 0.638$ ;  $p = 0.026$ ).

**Graph 4-14**



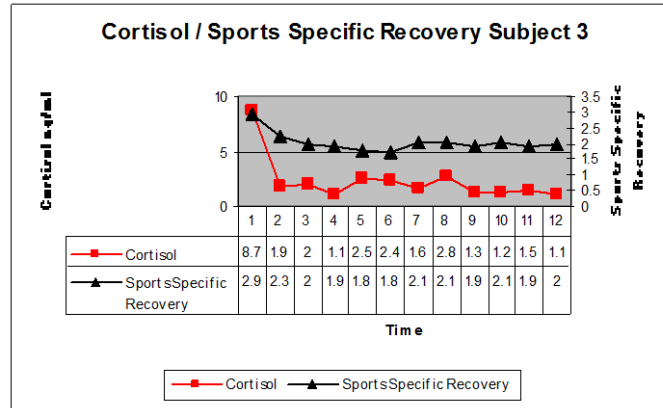
The data plotted in graph 4-15 were correlated, resulting in a non-significant coefficient ( $r_{(10)} = 0.389$ ;  $p = 0.211$ ) between salivary cortisol and sports specific stress. Subject 3 cortisol and sports specific stress scores are represented in graph 4-15. Finally, the data in

**Graph 4-15**



graph 4-16 were plotted to show the relationship between salivary cortisol and sports specific recovery. The correlation coefficient calculated for these data indicate a significant relationship between these two variables for subject 3 ( $r_{(10)} = 0.853$ ;  $p = 0.000$ ).

**Graph 4-16**



*Subject 4*

Table 4-6 presents the overall cortisol and recovery-stress scores for Subject 4 over the 12 week training session.

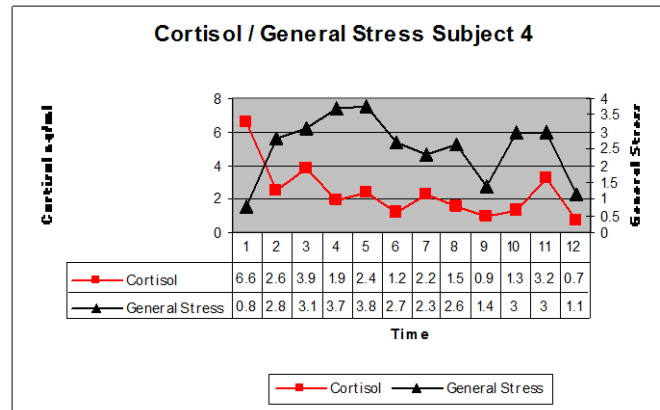
**Table 4-6**  
**Subject 4 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	2.3617	1.6342	12
General Stress	2.5275	.9652	12
General Recovery	3.0150	.8946	12
Sports Specific Stress	3.6450	.6100	12
Sports Specific Recovery	3.8442	.3464	12

The individual scores for subject 4 were plotted week-by-week across the 12 training sessions. These are shown in the graphs that follow. Four graphs are presented (Graph 4-17 through 4-20), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

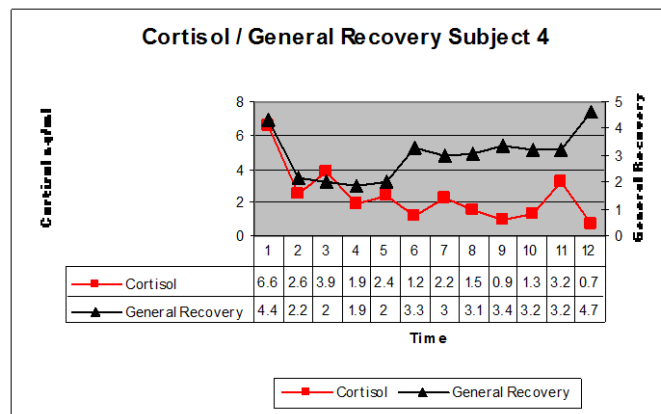
Subject 4 cortisol and general stress scores are presented in graph 4-17. The data in this graph were correlated to show the relationship between cortisol and general stress over time for this subject.

**Graph 4-17**



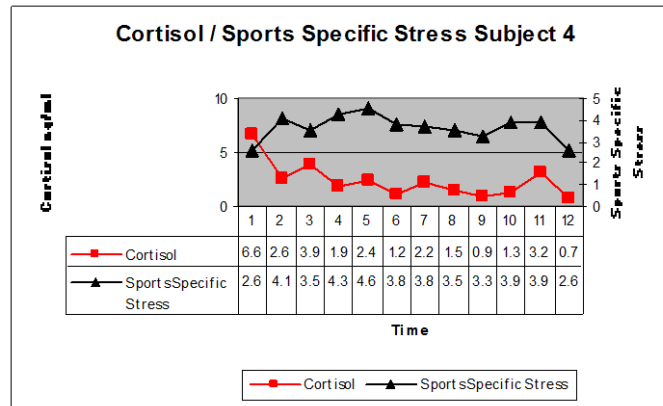
The Pearson correlation coefficient ( $r_{(10)} = -0.194$ ;  $p = 0.546$ ) suggested no significant associate between these two variables. Correlational analysis of the subject's data in graph 4-18 indicated no real relationship existed between salivary cortisol and general recovery ( $r_{(10)} = 0.057$ ;  $p = 0.859$ ).

**Graph 4-18**



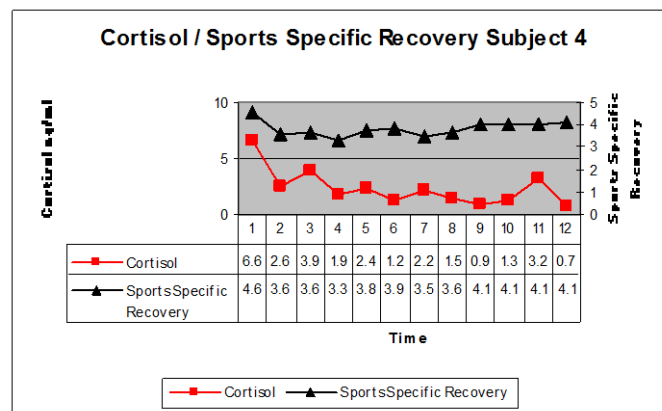
The data plotted in graph 4-19 were correlated, also resulting in a non-significant coefficient ( $r_{(10)} = -0.253$ ;  $p = 0.427$ ) between salivary cortisol and sports specific stress. The scores for this wrestler's cortisol and sports specific stress are present in graph 4-19.

**Graph 4-19**



Finally, the data in graph 4-20 were plotted to show the relationship between salivary cortisol and sports specific recovery. The correlation coefficient calculated for these data revealed no significant relationship between these two variables for subject 4 ( $r_{(10)} = 0.367$ ;  $p = 0.240$ ).

**Graph 4-20**



*Subject 5*

Table 4-7 presents the overall average cortisol and recovery-stress scores for subject 5 over the 12 week training session.

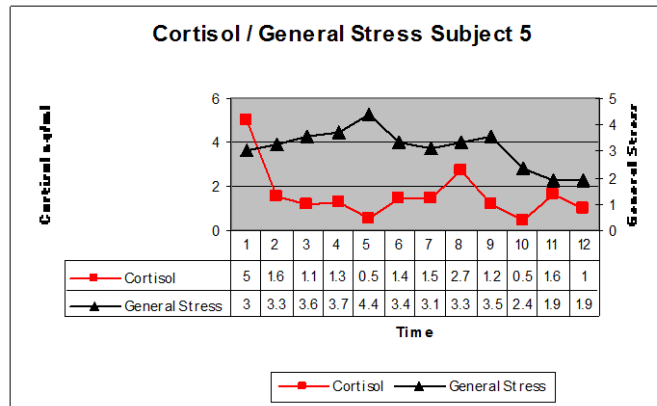
**Table 4-7**  
**Subject 5 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	1.6125	1.2090	12
General Stress	3.1192	.7494	12
General Recovery	3.3475	.5251	12
Sports Specific Stress	2.7142	.8642	12
Sports Specific Recovery	4.3300	.7719	12

The following graphs show that individual's scores which were plotted week-by-week across the 12 training sessions. Four graphs are presented (Graph 4-21 through 4-24), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

Subject 5 cortisol and general stress scores are present in graph 4-21. The data in

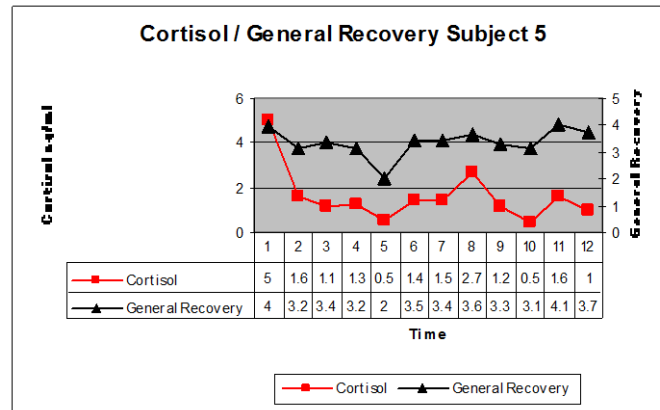
**Graph 4-21**



this graph were correlated, show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = -0.065$ ;  $p = 0.842$ ) suggested no significant association between these two variables. The correlation analysis of the subject's data in graph 4-22 indicates no significant relationship existed between salivary cortisol and general recovery ( $r_{(10)} = 0.568$ ;  $p = 0.054$ ).

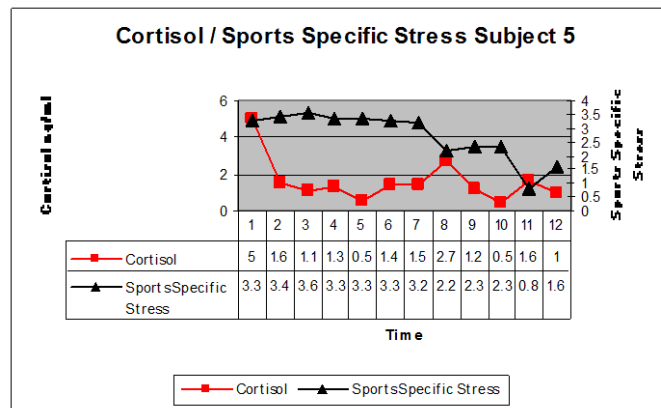


**Graph 4-22**



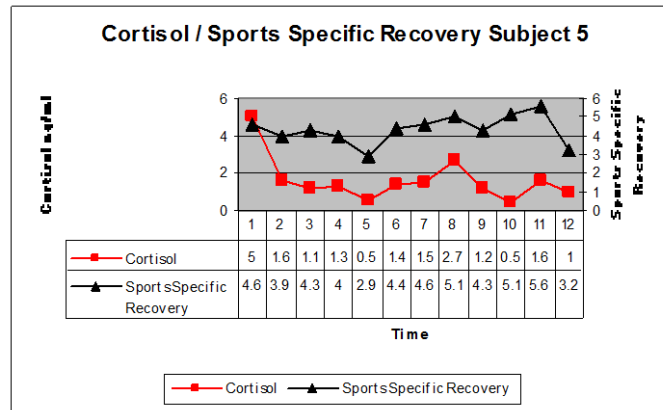
The data plotted in graph 4-23 were correlated, also resulting in a non-significant relationship (Pearson correlation coefficient ( $r_{(10)} = 0.091$ ;  $p = 0.778$ ). Graph 4-23 presents this wrestler's cortisol and sports specific stress scores. Finally, the data in

**Graph 4-23**



graph 4-24 were plotted to show the relationship between salivary cortisol and sports specific recovery. The correlation coefficient calculated for these data revealed no significant relationship between these two variables for subject 5 ( $r_{(10)} = 0.323$ ;  $p = 0.306$ ).

**Graph 4-24**



*Subject 6*

Table 4-8 reflects overall average cortisol and recovery-stress scores for Subject 6 over the 12 week training session.

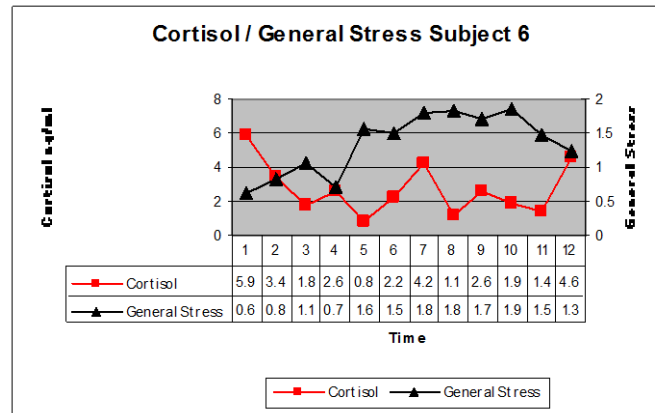
**Table 4-8**  
**Subject 6 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	2.7142	1.5357	12
General Stress	1.3475	.4486	12
General Recovery	4.1292	.4614	12
Sports Specific Stress	1.0475	.3457	12
Sports Specific Recovery	4.1267	.6016	12

The individual's scores, plotted week-by-week across the 12 training sessions, are shown in the graphs that follow. The four graphs are presented (Graph 4-25 through 4-28), with all graphs showing the salivary cortisol variable and one of the subcategories of stress.

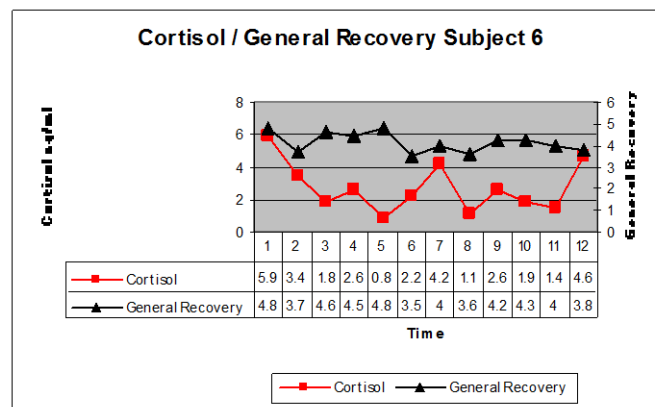
Review of the subject's data in graph 4-25 presents cortisol and general stress

**Graph 4-25**



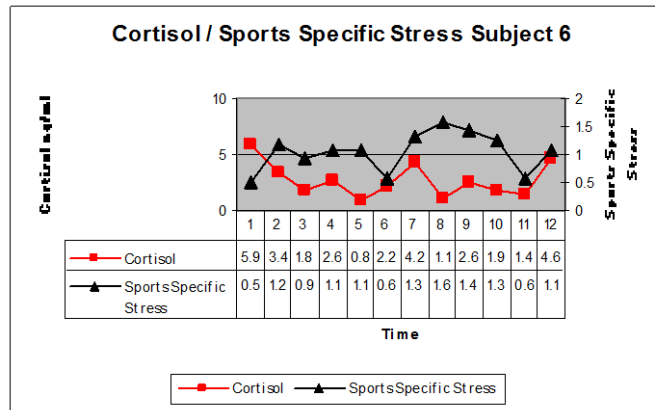
scores. The data in this graph were correlated, to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = -0.503$ ;  $p = 0.096$ ) indicates that there appears to be a non significant relationship between salivary cortisol and general stress. Correlational analysis of the subject’s data in graph 4-26 indicates a non significant relationship between salivary cortisol and general recovery ( $r_{(10)} = 0.037$ ;  $p = 0.909$ ).

**Graph 4-26**



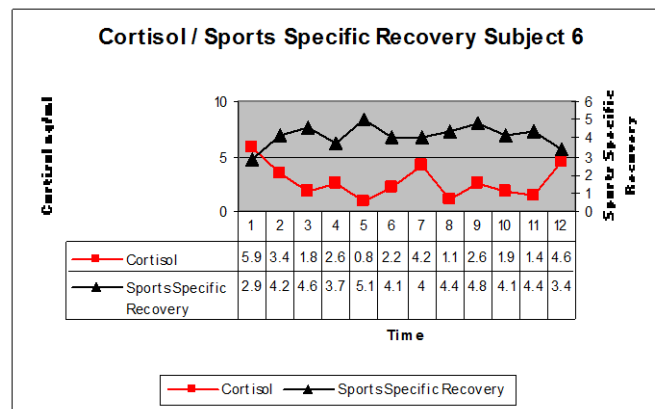
The data plotted in graph 4-27 were correlated, also resulting in a non-significant coefficient ( $r_{(10)} = -0.230$ ;  $p = .0473$ ) between salivary cortisol and sports specific stress.

**Graph 4-27**



Finally, review of the subject’s data in graph 4-28 were plotted to show the relationship between salivary cortisol and sports specific recovery. The correlation coefficient indicates that there appears to be a significant negative relationship between these two variables ( $r_{(10)} = -0.828$ ;  $p = 0.001$ ).

**Graph 4-28**



*Subject 7*

Table 4-9 reflects the overall average scores of Subject 7 on cortisol and recovery-stress scores over the 12 week training session.

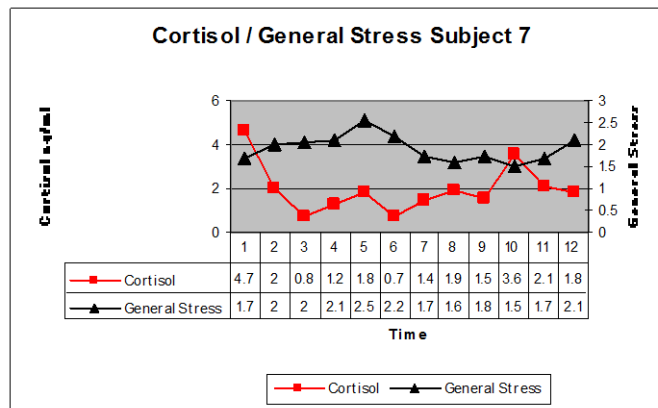
**Table 4-9**  
**Salivary Cortisol Stress Recovery Scores**  
**Subject 7**

	Mean	Std. Deviation	N
Cortisol ng/ml	1.9675	1.1208	12
General Stress	1.9092	.3022	12
General Recovery	3.0708	.5968	12
Sports Specific Stress	1.9942	.4806	12
Sports Specific Recovery	3.8817	.4622	12

The individual's scores, plotted week-by-week across the 12 training sessions, are in the graphs that follow. Four graphs are presented (Graphs 4-29 through 4-32), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

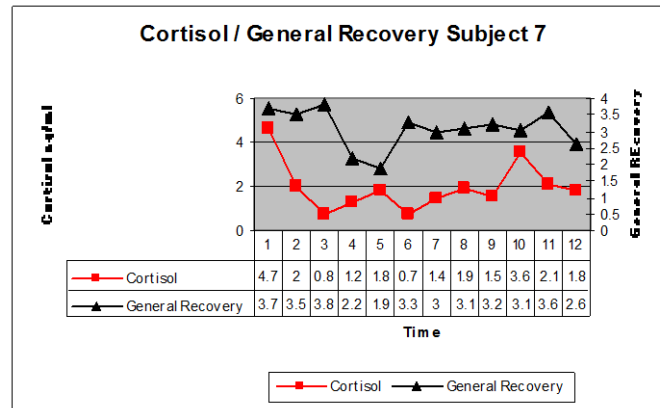
Graph 4-29 presents the scores for cortisol and general stress for subject 7.

**Graph 4-29**



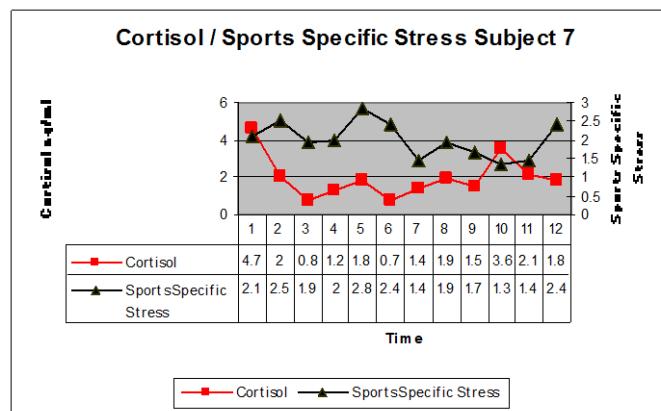
Review of the subject's data in this graph were correlated to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = -0.502$ ;  $p = 0.097$ ) indicates that there appears to be a non significant relationship between these two variables. The Pearson correlation coefficient of the subject's data in graph 4-30 indicates a non-significant relation existed between salivary cortisol and general recovery ( $r_{(10)} = 0.207$ ;  $p = 0.518$ ).

**Graph 4-30**



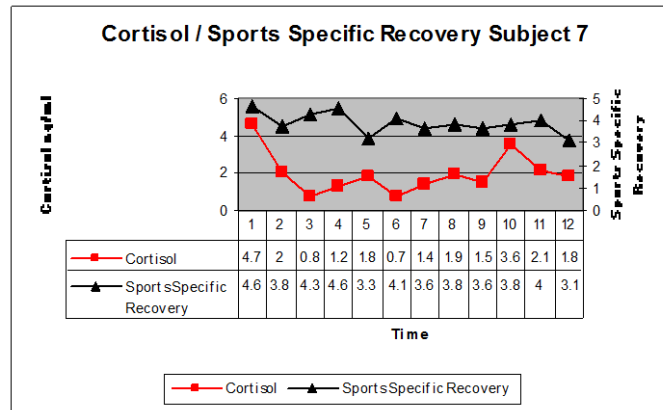
The data reviewed in graph 4-31 were correlated, also indicating that there appears to be a non significant relationship between salivary cortisol and sports specific stress ( $r_{(10)} = -0.176$ ;  $p = 0.584$ ) graph 4-31 presents this wrestler’s cortisol and sports specific stress.

**Graph 4-31**



Finally, the review of the subject’s data in graph 4-32 indicates that there appears to be a non significant relationship between salivary cortisol and sports specific recovery. The correlation coefficient calculated for these data revealed no significant relationship between these two variables for subject 7 ( $r_{(10)} = 0.195$ ;  $p = 0.544$ ).

**Graph 4-32**



*Subject 8*

Table 4-10 presents the overall average cortisol and recovery-stress scores for Subject 8 over the 12 week training session

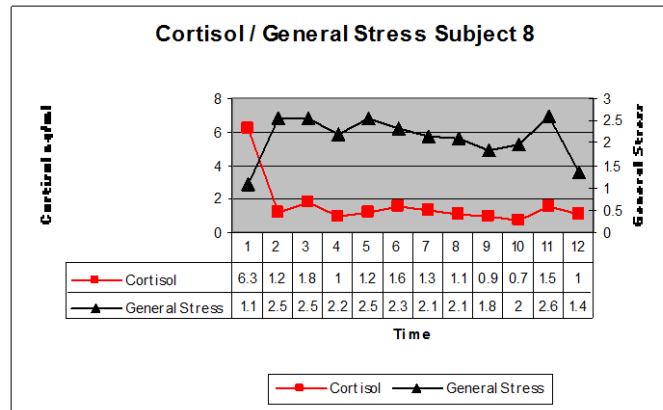
**Table 4-10**  
**Subject 8 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	1.6400	1.4848	12
General Stress	2.0788	.4708	12
General Recovery	2.6542	.6055	12
Sports Specific Stress	1.5150	.5388	12
Sports Specific Recovery	3.0296	.7353	12

The individual’s scores, plotted week-by-week across the 12 training sessions, are shown in the graphs that follow. Four graphs are presented (Graphs 4-33 through 4-36), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

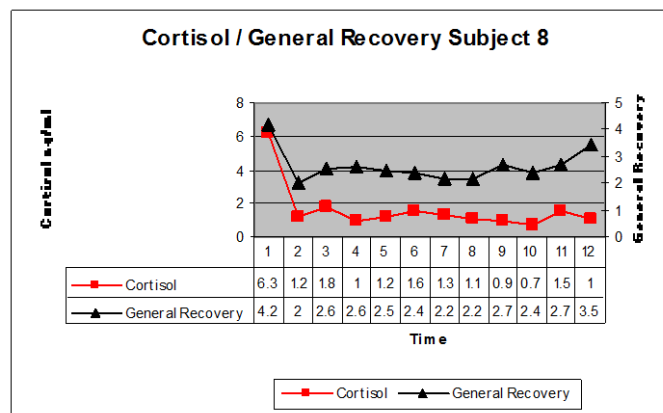
Review of data in graph 4-33 present cortisol and general stress scores for

**Graph 4-33**



subject 8. The data in this graph were correlated, to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = -0.565$ ;  $p = 0.055$ ) indicates that there appears to be a non significant relationship between salivary cortisol and general stress. Correlational analysis of the subject's data in graph 4-34 indicates that there appears to be a significant relationship between salivary cortisol and general recovery ( $r_{(10)} = 0.774$ ;  $p = 0.003$ ).

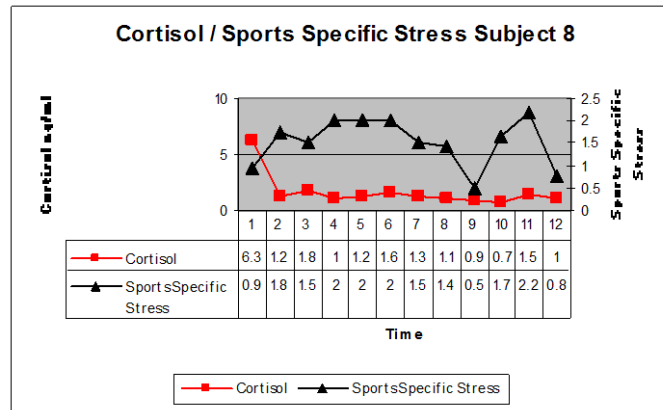
**Graph 4-34**



The data plotted in graph 4-35 were correlated, also resulting in a non-significant coefficient ( $r_{(10)} = -0.265$ ;  $p = 0.404$ ) between salivary cortisol and sports specific stress. The wrestler's cortisol and sports specific stress scores and present in graph 4-35.

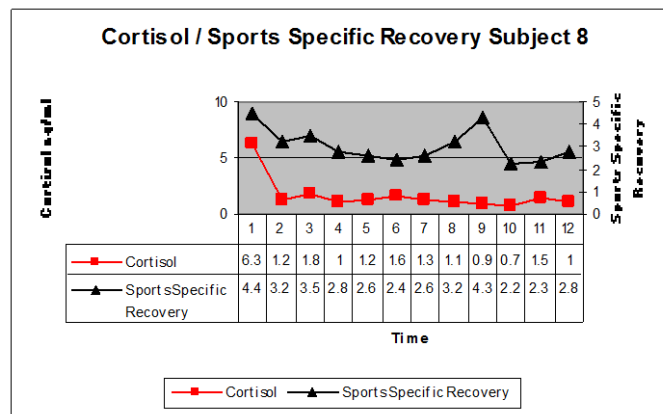


**Graph 4-35**



Finally, the review of the subject's data in graph 4-36 indicates that there appears to be a significant relationship between salivary cortisol and sports specific recovery ( $r_{(10)} = 0.584$ ;  $p = 0.046$ ). The data plotted in graph 4-36 were plotted to show the relationship between these two variables.

**Graph 4-36**



### *Subject 9*

Table 4-11 reflects the individual's scores, plotted week-by-week across the 12 training sessions, are shown in the graphs that follow.

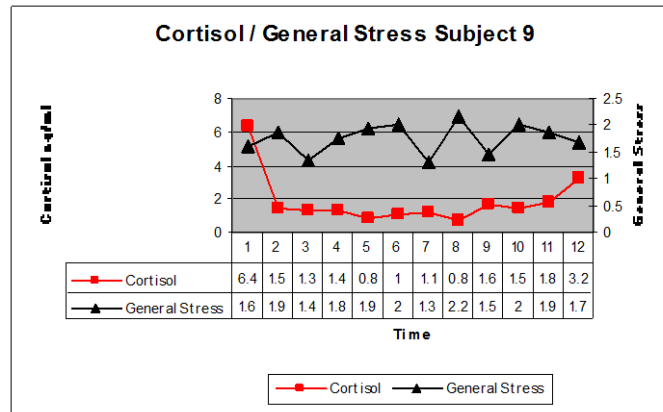
**Table 4-11**  
**Subject 9 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	1.8625	1.5583	12
General Stress	1.7500	.2774	12
General Recovery	2.4917	.5908	12
Sports Specific Stress	1.3933	.5934	12
Sports Specific Recovery	2.0992	.6482	12

The individual's scores, plotted week-by-week across the 12 training sessions, in the graphs that follow. Four graphs are presented (Graphs 4-37 through 4-40), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

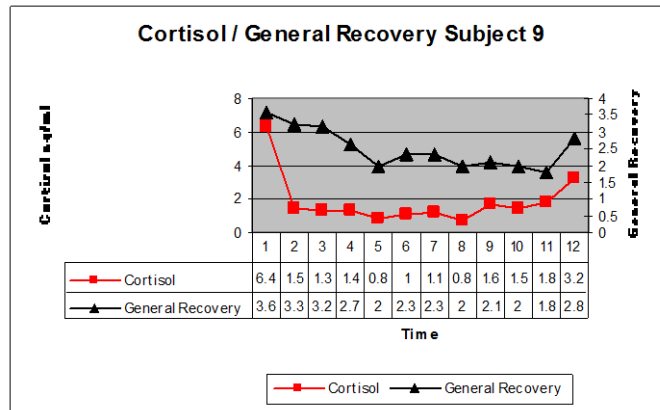
Review of the subject's data in graph 4-37 present cortisol and general stress for

**Graph 4-37**



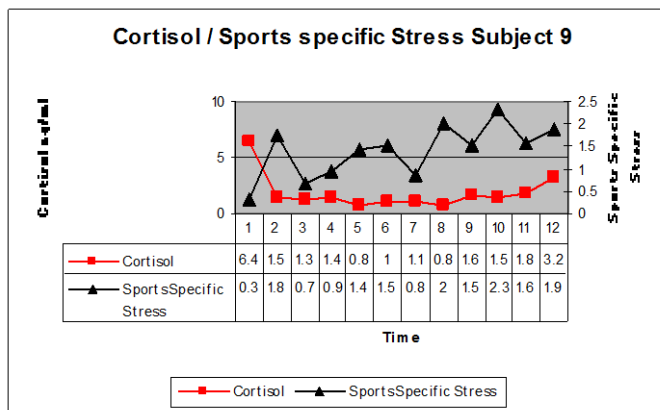
subject 9. The data in this graph were correlated, to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient indicates that there appears to be a non significant relationship between salivary cortisol and general stress ( $r_{(10)} = -0.236$ ;  $p = 0.460$ ). Review of the subject's data in graph 4-38 indicates that there appears to be a significant relationship between salivary cortisol and general recovery ( $r_{(10)} = 0.627$ ;  $p = 0.029$ ).

**Graph 4-38**



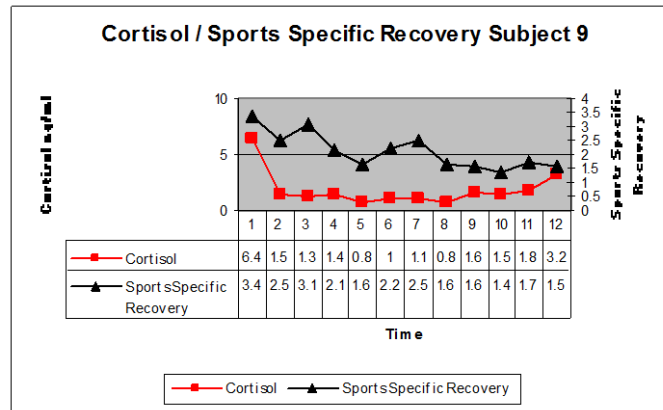
The data plotted in graph 4-39 were correlated, also resulting in a non significant relationship between salivary cortisol and sports specific stress ( $r_{(10)} = -0.433$ ;  $p = 0.160$ ).

**Graph 4-39**



Review of the subject's data in graph 4-40 indicates that there appears to be a non significant relationship between salivary cortisol and sports specific recovery ( $r_{(10)} = 0.484$ ;  $p = 0.111$ ). The data in graph 4-40 were plotted to show the relationship for these two variables.

**Graph 4-40**



*Subject 10*

Table 4-12 presents the overall average cortisol and recovery-stress scores for Subject 10 over the 12 week training session.

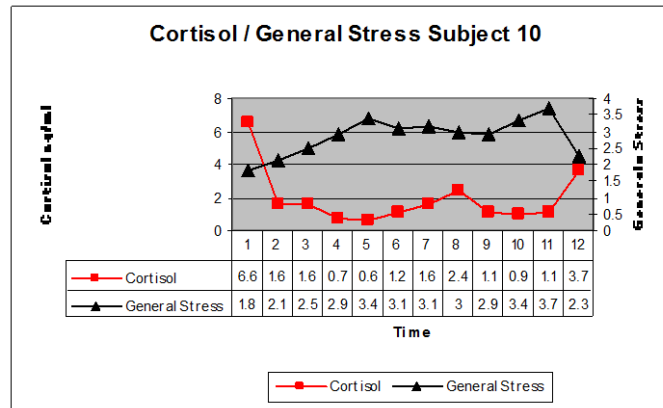
**Table 4-12**  
**Subject 10 Salivary Cortisol Stress Recovery Scores**

	Mean	Std. Deviation	N
Cortisol ng/ml	1.9033	1.9033	12
General Stress	2.8442	2.8442	12
General Recovery	1.7854	1.7854	12
Sports Specific Stress	3.7496	3.7496	12
Sports Specific Recovery	2.7300	2.7300	12

That individual’s scores, plotted week-by-week across the 12 training sessions, are shown in the graphs that follow. The four graphs are presented (Graph 4-41 through 4-44), with all graphs showing the salivary cortisol variable and one of the subcategories of recovery-stress.

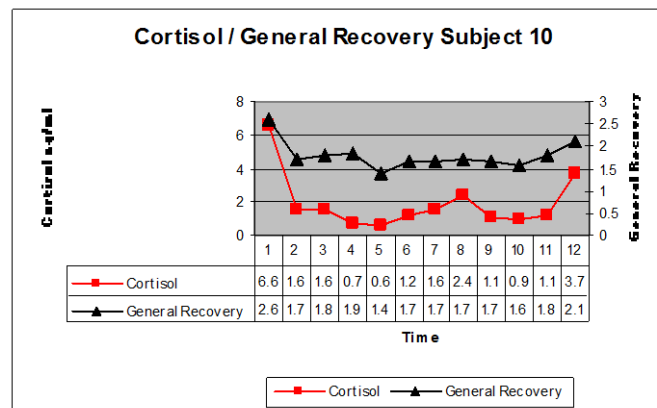
Review of the subject’s data in graph 4-41 present cortisol and general stress for subject 10.

**Graph 4-41**



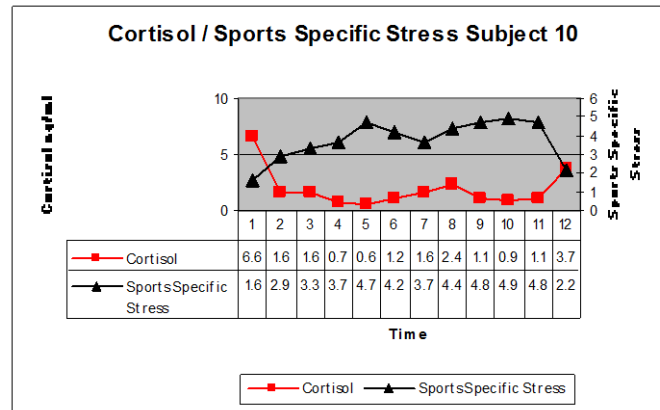
The data in this graph were correlated to show the relationship between cortisol and general stress over time for this subject. The Pearson correlation coefficient ( $r_{(10)} = -0.731$ ;  $p = 0.007$ ) indicates that there appears to be a significant relationship between salivary cortisol and general stress. Correlational analysis of the subject's data in graph 4-42 indicates a significant relationship exist between salivary cortisol and general recovery ( $r_{(10)} = 0.922$ ;  $p = 0.000$ ).

**Graph 4-42**



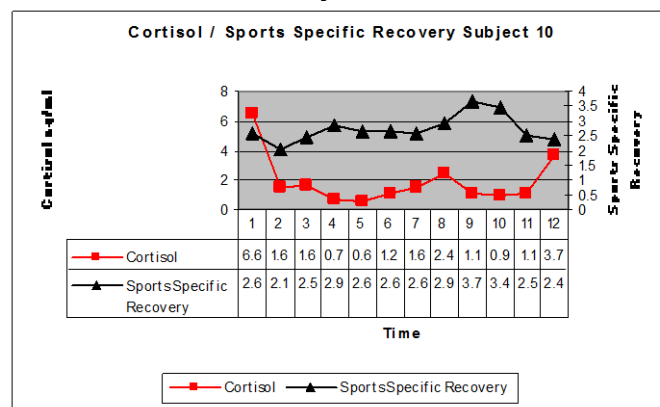
Review of the subject's data in graph 4-43 indicates that there appears to be a significant relationship between salivary cortisol and sports specific stress ( $r_{(10)} = -0.809$ ;  $p = 0.001$ ). Graph 4-43 presents this wrestler's cortisol and sports specific stress scores.

**Graph 4-43**



Finally, review of the subject's data in graph 4-44 indicates that there appears to be a non significant relationship between salivary cortisol and sports specific recovery ( $r_{(10)} = -0.240$ ;  $p = 0.453$ ). The data were plotted in graph 4-44 show the relationship between these two variables.

**Graph 4-44**



### ***Research Question 3***

Did salivary cortisol and recovery-stress significantly change during the course of the 12-week training? To answer this question, a single factor repeated measures Analysis of Variance (ANOVA) was conducted separately for each variable of interest.

***Salivary Cortisol***

In the analysis of salivary cortisol, ANOVA results indicated significant changes occurred during the 12-week testing session for these wrestlers [ $F_{(11,99)} = 16.127$ ;  $p < .01$ ]. This analysis is summarized in Table 4-13. When looking at all athletes during the 12 week testing season and post season there were changes in the concentration of salivary cortisol.

**Table 4-13**

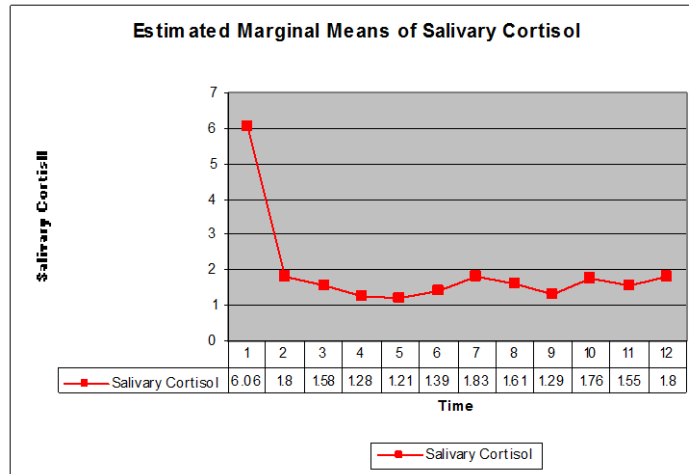
**Tests of Within-Subjects Effects for Cortisol General Linear Model**

Measure: MEASURE\_1

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Time	Sphericity Assumed	191.672	11	17.425	16.127	.000
	Greenhouse-Geisser	191.672	2.763	69.378	16.127	.000
	Huynh-Feldt	191.672	4.109	46.650	16.127	.000
	Lower-bound	191.672	1.000	191.672	16.127	.003
Error(time)	Sphericity Assumed	106.965	99	1.080		
	Greenhouse-Geisser	106.965	24.865	4.302		
	Huynh-Feldt	106.965	36.979	2.893		
	Lower-bound	106.965	9.000	11.885		

The overall cortisol means for all ten wrestlers combines were then plotted to show the trend in the data across the testing sessions. As noted in the graph (see Graph 4-45), average cortisol was very high at the initial testing session, then it dropped and remained fairly consistent across the remaining sessions.

**Graph 4-45**



***Recovery-Stress Questionnaire for Athletes***

*General Stress*

In the analysis of the subcategories for the RESTQ-Sport Questionnaire, ANOVA results indicated significant changes occurred for subcategory general stress during the 12-week testing session for these wrestlers [ $F_{(11,99)} = 2.763$ ;  $p < .01$ ]. This analysis is summarized in Table 4-14. When looking at all athletes during the 12 week testing season and post season there were changes in the general stress scores.

**Table 4-14**

**Tests of Within-Subjects Effects for General Stress General Linear Model**

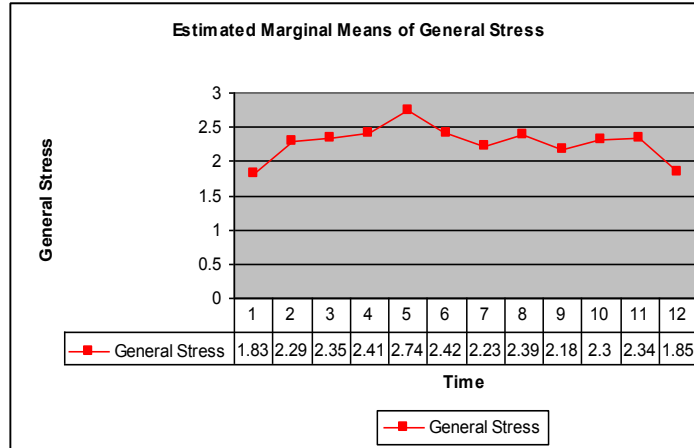
Measure: MEASURE\_1

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Time	Sphericity Assumed	6.663	11	.606	2.763	.004
	Greenhouse-Geisser	6.663	3.493	1.907	2.763	.051
	Huynh-Feldt	6.663	5.980	1.114	2.763	.021
	Lower-bound	6.663	1.000	6.663	2.763	.131
Error(time)	Sphericity Assumed	21.700	99	.219		
	Greenhouse-Geisser	21.700	31.438	.690		
	Huynh-Feldt	21.700	53.819	.403		
	Lower-bound	21.700	9.000	2.411		



The overall general stress means for all ten wrestlers combined were then plotted to show the trend across the testing sessions. As noted in the graph (see Graph 4-46), average general stress was lower at the initial testing session, then increased and remained fairly consistent across the remaining sessions.

**Graph 4-46**



*General Recovery*

In the analysis of general recovery, ANOVA results indicated significant changes occurred during the 12-week testing session for these wrestlers [ $F_{(11,99)} = 5.628$ ;  $p < .01$ ]. This analysis is summarized in Table 4-15. When looking at all athletes during the 12 week testing season and post season there were changes in the general recovery scores.

**Table 4-15**

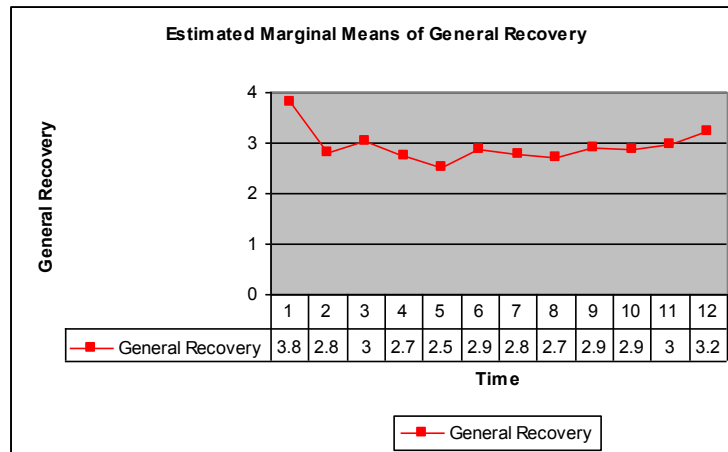
**Tests of Within-Subjects Effects for General Recovery General Linear Model**

Measure: MEASURE\_1

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Time	Sphericity Assumed	11.539	11	1.049	5.628	.000
	Greenhouse-Geisser	11.539	3.237	3.565	5.628	.003
	Huynh-Feldt	11.539	5.269	2.190	5.628	.000
	Lower-bound	11.539	1.000	11.539	5.628	.042
Error(time)	Sphericity Assumed	18.455	99	.186		
	Greenhouse-Geisser	18.455	29.131	.634		
	Huynh-Feldt	18.455	47.423	.389		
	Lower-bound	18.455	9.000	2.051		

The overall general recovery means for all ten wrestlers combined were then plotted to show the trend in the data across the testing sessions. As noted in the graph (see Graph 4-47), average general recovery was elevated at the initial testing session, then it decreased and remained fairly consistent across the remaining sessions.

**Graph 4-47**



*Sports Specific Stress*

In the analysis of sports specific stress, ANOVA results indicated significant changes occurred during the 12-weeking testing session for these wrestlers [F (11,99) = 2.174; p < .05]. The analysis is summarized in Table 4-16. When looking at all athletes

during the 12 week testing season and post season there were changes in sport specific stress scores.

**Table 4-16**

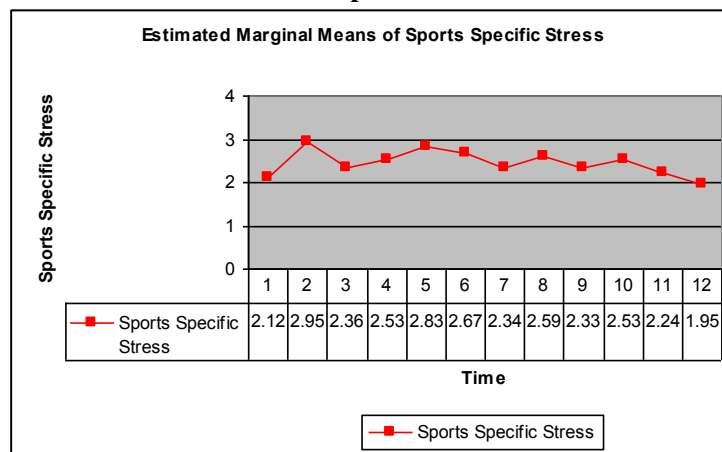
**Tests of Within-Subjects Effects for Sports Specific Stress General Linear Model**

Measure: MEASURE\_1

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Time	Sphericity Assumed	9.189	11	.835	2.174	.022
	Greenhouse-Geisser	9.189	2.789	3.295	2.174	.120
	Huynh-Feldt	9.189	4.169	2.204	2.174	.088
	Lower-bound	9.189	1.000	9.189	2.174	.174
Error(time)	Sphericity Assumed	38.046	99	.384		
	Greenhouse-Geisser	38.046	25.103	1.516		
	Huynh-Feldt	38.046	37.519	1.014		
	Lower-bound	38.046	9.000	4.227		

The overall sports specific stress means for all ten wrestlers combined were then plotted to show the trend in the data across the testing sessions. As noted in the graph (see Graph 4-48), average sports specific stress was low at the initial testing session, the it elevated and remained elevated until the end of testing when the scores began to decrease.

**Graph 4-48**



### *Sport Specific Recovery*

In the analysis of sports specific recovery, ANOVA results indicated significant changes occurred during the 12-week testing session for these wrestlers [ $F_{(11,99)} = 1.931$ ;  $p < .05$ ]. This analysis is summarized in Table 4-17. When looking at all athletes during the 12 week testing season and post season there were changes in the sports specific recovery scores.

**Table 4-17**

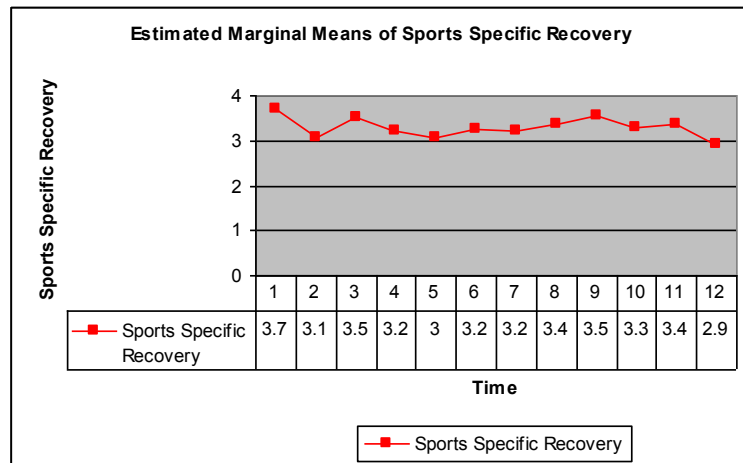
**Tests of Within-Subjects Effects for Sports Specific Recovery General Linear Model**

Measure: MEASURE\_1

Source		Type III Sum of Squares	df	Mean Square	F	Sig.
Time	Sphericity Assumed	5.589	11	.508	1.931	.044
	Greenhouse-Geisser	5.589	4.302	1.299	1.931	.120
	Huynh-Feldt	5.589	8.732	.640	1.931	.061
	Lower-bound	5.589	1.000	5.589	1.931	.198
Error(time)	Sphericity Assumed	26.042	99	.263		
	Greenhouse-Geisser	26.042	38.720	.673		
	Huynh-Feldt	26.042	78.592	.331		
	Lower-bound	26.042	9.000	2.894		

The overall sports specific recovery means for all ten wrestlers combined were then plotted to show the trend in the data across the testing sessions. As noted in the graph (see Graph 4-49), average sports specific recovery scores was high at the initial testing session, then it dropped and remained fairly constant until the end when it dropped again.

**Graph 4-49**



***Discussion***

The first purpose of this investigation was to see if there is a relationship between salivary cortisol and recovery-stress scores for all wrestlers combined. The second purpose was to see if there were changes person by person, or collectively for all weight classes for competitive wrestlers. The third purpose of the current investigation was to explore if salivary cortisol levels would change as the stress-recovery scores changed. The following discussion sections compare the results to other reports that focused on athletes in other sports.

The results of the present study are in contrast with previous studies reporting elevations of salivary cortisol following exercise (Morgan *et al.*, 1989). Morgan *et al.* (1989) reported that while a reduction in training was associated with lower levels of salivary cortisol, no increase above baseline was observed during overtraining. Carlie *et al.* (1983) reported that it is possible that the resting cortisol levels have been shown to increase at the outset of training and then fall as a trained state is achieved.

The present study is in agreement with the findings of Morgan et al. (1989) who reported a change in salivary cortisol levels during training of female swimmers during the intense time period of two-a-day training.

M. Kellman (personal communication, February 2, 2006) reported use of the RESTQ-Sport Questionnaire with just one athlete, and suggested that in the stress categories scores of 2 and below are optimal, and in the recovery categories scores of 4 and higher are optimal. These scores are to be evaluated as an orientation in which direction scores should develop.

In the present study, results indicate that from the onset of data collection, 8 subjects scored above the optimal level of 2 in the RESTQ-Sport Questionnaire subcategory of general stress, with 2 subjects scoring below the optimal level. The subcategory of general recovery showed only one subject above the optimal level of 4 while the remaining 9 were below the optimal level. The subcategory of sports specific stress showed 4 subjects were below the optimal level of 2 while the other 6 were above the optimal level. The subcategory of sports specific recovery showed only 2 subjects above the optimal level of 4 while the remaining 8 were below the optimal level.

### ***Relationship between Cortisol and Recovery Stress Scores***

The results of the present investigation revealed a non significant Pearson correlation coefficient between salivary cortisol concentration and RESTQ-Sport recovery stress scores for wrestlers during a season and post season of competition in collegiate wrestling (Table 4-2, Graph 4-1 thru 4-4). The present study is in agreement with the work of Morgan et al. (1989) who reported no significant correlation between

salivary cortisol and mood state following overtraining in female swimmers. The literature suggests that the study by Morgan et al. (1989) and the present study are the only studies comparing salivary cortisol and recover stress scores. While the instrument used for determining stress, recovery, and mood are different, the RESTQ-Sport Questionnaire was not yet developed during the study by Morgan et al. (1989).

Review of the literature also suggests that this study is the first to investigate salivary cortisol and recovery stress scores in wrestlers. No significant correlations were found between salivary cortisol and recovery stress scores as a group. Subjects 2, 3, 8, 9, and 10 indicated there is a significant positive correlation between general recovery, and cortisol levels in these individuals. This correlation shows that as cortisol levels increased so did the recovery scores. Subjects 3 and 8 indicated there is a significant positive correlation between sports specific recovery and cortisol levels in these individuals. This correlation shows that as cortisol levels increased so did the recovery scores. Subject 6 was associated with a significant negative correlation between cortisol and recovery scores. This correlation shows that as recovery scores increase that cortisol levels decrease. Subject 10 had a significant negative correlation between stress and cortisol. This correlation shows that as stress increases that cortisol levels decrease.

Changes in both total group scores for stress, recovery, and cortisol were found independent of one another, but nothing was found to connect the scores to each other. Further research is needed to determine if a relationship does exist between salivary cortisol and recovery stress. This relationship could be determined by using additional physiological markers, adding a control group, additional subjects, and more subjects per weight category.

## CHAPTER V

### SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

#### *Summary*

The present study revealed that participation in collegiate wrestling did not tend to influence the relationship between salivary cortisol levels and corresponding stress-recovery scores. While the trends between the levels of salivary cortisol showed higher levels in some subjects, the levels were not consistent among all subjects. The trends among stress-recovery scores suggested that in the four general categories of stress and recovery, the lack of recovery and decrease in stress were evident. The results of the present study indicated that salivary cortisol and stress-recovery scores during a collegiate wrestling season were most likely unrelated. Further studies are necessary to determine the relationship between salivary cortisol and stress-recovery.

#### *Salivary Cortisol*

The data showed a significant effect of competition and training on salivary cortisol, but the effect did not show trends in either increase or decrease of salivary cortisol. It appeared that some subjects experienced increases, and some subjects experienced decreases. The changes were directional at any given time.



### ***Stress***

The data showed positive and negative changes in both general stress and sports specific stress. The changes were not directly or indirectly related to salivary cortisol. Inconsistent changes were reflected in that while some subjects' stress scores increased the cortisol levels either stayed the same or decreased; and conversely in other subjects, as stress scores decreased that cortisol levels either stayed the same or decreased.

### ***Recovery***

The data showed positive and negative changes in both general recovery and sports specific recovery. The changes were not directly or indirectly related to salivary cortisol. Again, inconsistent changes were observed: in some subjects as recovery scores increased cortisol levels either stayed the same or increased, and in other subjects as recovery scores decreased that cortisol levels either stayed the same or decreased.

### ***Salivary Cortisol and Stress-Recovery Scores***

The data showed that while there were changes in salivary cortisol, stress and recovery, these changes were not correlated. While individual subjects showed a correlation between recovery and salivary cortisol, that correlation was not as expected. As a recovery score increased, the literature suggested there should be an associated decrease in salivary cortisol. The opposite was true with subjects 3, 9, 10; as recovery scores increased, salivary cortisol increased, and the opposite is also true as recovery scores decreased, salivary cortisol decreased. Subjects 2, 6, 8, 9, 10 demonstrated a salivary cortisol decrease as recovery scores increase. Conversely, the literature

suggested that as stress scores decreased there would be an associated decrease in salivary cortisol. Only one subject in this study demonstrated a correlation between salivary cortisol and stress scores, and this was a negative correlation in that as stress scores increased salivary cortisol decreased as well.

### ***Conclusion***

The data in this study demonstrated that there were changes in salivary cortisol, stress, and recovery scores as a result of competing in a competitive collegiate wrestling program. The relationship between salivary cortisol and stress-recovery scores appeared to be somewhat independent and unrelated. The results of the present study along with previous research indicated salivary cortisol production seemed to have no effect on stress-recovery scores. Saliva sampling offered an inexpensive, non invasive and simple collection method of obtaining physiological material for monitoring the physiological cost of over-training and participating in sport. The RESTQ-Sport Questionnaire offered an equally simple tool for monitoring the psychological cost of over-training. The information gathered is beneficial in preventing and reducing the incidences of illness during a competitive season. Future studies are needed to fully understand salivary cortisol response to exercise and its role as a marker for over-training as associated with stress and recovery.

### ***Recommendations***

1. Repeat the same study with a more random and larger sample size. The increased numbers would improve recognizing significant changes in dependent variables.
2. Include a control group of either non-competitive or second and third level wrestlers or matched subjects that workout and control weight.
3. Measure salivary cortisol and stress-recovery scores throughout an entire season to assess the responses to various training volumes, competitive play, travel, and conference and national tournament play.
4. Record incidences of illness and injury associated with life, physical training, and competition to investigate other factors that may contribute to salivary cortisol production.

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## APPENDIX A

Written copy of Informed Consent to be provided to Oklahoma State University Wrestling Athletes

RESEARCH PARTICIPANT CONSENT FORM  
SALIVARY IMMUNE RESPONSE AND PSYCHOLOGICAL RECOVERY  
QUESTIONNAIRE TO A COMPETITIVE COLLEGIATE WRESTLING SEASON  
Principal Investigators: Daniel L. Dodson, M.Ed., A.T.,C.  
Oklahoma State University  
Department of Health, Leisure, and Human Performance

The present research project will examine the effect of a collegiate wrestling season on the presence of immune markers present saliva and the correlated scores on the Rest-Recovery Questionnaire for Sport. You have been selected because you are an University varsity wrestling athlete. You will be asked to provide data samples based on time points throughout the season, and post-season. You will receive a copy of this consent form to keep for your records.

### *Purpose of Research*

The purpose of this project is to assess the influence of a competitive wrestling season salivary immune markers and recovery scores on the Rest-Recovery Questionnaire for Sport.

### *Requirements of Participants*

1. Received a physical examination and cleared to participate in competitive sports.
2. Complete informed consent forms.
3. Provide saliva samples that will be taken the on the dates specified.
4. You will be asked to report any injuries in the mouth due to abrasive brushing, poor oral health, etc. before saliva sample collection. Also, we ask that you do not brush your teeth within 3 hours prior to sample collection. Dental work should not be performed within 24 hours prior to sample collection. In addition, do not eat a meal within 60 minutes and avoid alcohol 24 hours prior to sample collection.



### ***Duration of Participation***

A total of 13 weeks of participation will be required of each subject. These include filling out informed consent, donation saliva samples and completion of questionnaire the day of the scheduled collection. Time for sample collection will take approximately 15 minutes. The written questionnaire will take 50 minutes to complete resulting in a maximum of 1.5 hours spent completing the data collection during the 13 week period.

### ***Benefits to the Individual***

Benefits to the individual include information on how your saliva immune proteins (Cortisol) respond to participation in a competitive collegiate wrestling. Salivary cortisol has been utilized as a marker of the body's immune response to exercise. The results of this study will help to clarify the response of immune system and the psychological response to a competitive wrestling season. We hope to assist in providing information necessary to prevent or reduce the incidence of illness in athletes, thus improving their overall health. In addition, provide information that may aid in the development of a scientifically based training program.

### **Risks to the Individual**

There are no known risks involved with saliva donation. There are no known risks involved with responding to the questionnaire.

### ***Medical liability***

I understand the risks associated with this study and voluntarily choose to participate. I understand that in case of injury or illness resulting from this study, emergency medical care is available through community health care providers by dialing 911. I understand that no funds have been set aside by Oklahoma State University to compensate me in the event of illness or injury.

### ***Confidentiality***

All scientific and personal data collected on subjects will be kept confidential and stored in a locked file cabinet in the principle investigators office. This information will be available only to the Principal investigator. All subjects will be assigned a subject number and all samples being analyzed by lab technicians will be labeled with these subject numbers to ensure anonymity. Information concerning assigned subject numbers will be kept separate from confidential material. At no time will any of the subjects' personal information be divulged. All data will be reported as means and standard errors. E-mail communications will be kept strictly confidential; all emails will be deleted upon completion of data collection. Access to the completed questionnaires will be limited to the Primary investigators, and hard copy forms will be coded, to ensure anonymity. All hard copies of data will be kept until entered into computer file for analysis, and the data will be stored in a locked file cabinet in the primary investigators office until completion

of data collection and then destroyed. The Institutional Review Board has authority to inspect all forms and data collected.

***Voluntary Nature of Participation***

Participation in this research project is voluntary and you can withdraw your involvement in the project at any time without penalty.

***Human Subject Statement***

For any questions you may have about this research project, contact Daniel L. Dodson, M.Ed. (405) 255-7208 or Dr. Steven Edwards (405) 744-7476. If you have any concerns dealing with subjects' rights, contact Dr Sue C. Jacobs, IRB chair, 415 Whitehurst Hall, Oklahoma State University, 405-744-1676.

I HAVE READ AND FULLY UNDERSTAND THE CONSENT FORM. I SIGN IT FREELY AND VOLUNTARILY. A COPY OF THIS FORM HAS BEEN GIVEN TO ME.

\_\_\_\_\_  
Signature of Participant

\_\_\_\_\_  
Date of birth

\_\_\_\_\_  
Date

I certify that I have personally explained this document before requesting that the participant sign it.

\_\_\_\_\_  
Signature of Researcher

\_\_\_\_\_  
Date

## **APPENDIX B**

### Script for Team Meeting

#### Purpose

The purpose of this study is try-fold. First, determine if there are differences in cortisol levels and rest-recovery scores by weight class. Second, to evaluate the cortisol to rest-recovery relationship for each wrestler overtime. Third, determine pre-to-post differences in rest-recovery and cortisol based on whether or not weight is an issue for wrestlers. The investigation would help to explore the relationship between the physiological marker (cortisol) and the psychological marker (rest-recovery score). This information will help the athletes, coaches, athletic trainers, and other allied health professionals to understand the warning signs of overtraining and help the athlete to understand the need for proper intervention to possibly prevent the athlete from overtraining.

#### Items Tested

*Recovery-Stress Questionnaire for Athletes (RESTQ-Sport)*. The Recovery-Stress Questionnaire for Athletes (RESTQ-Sport) is an instrument that systematically reveals the Recovery-stress states of athletes. The recovery-stress state indicates the extent to which persons are physically and / or mentally stressed, whether or not they are capable of using individual strategies for recovery, as well as which strategies are used (Kallus 1995).

Cortisol secretion also accelerates lipid mobilization for energy. This is especially apparent during starvation and exercise. There is considerably variability in the cortisol response to exercise, depending on such factors as exercise intensity and duration, fitness level, nutritional status, and even circadian rhythm (McArdle, Katch & Katch, 1994).

### Collection Procedures

Upon approval from the Oklahoma State University Institutional Review Board, participants for this study will be selected, from a convenience sample, by their rank (weight class) and status (varsity/starter) on the varsity wrestling team. The two measuring devices used in this study are the Recovery-Stress Questionnaire for Athletes (RESTQ-Sport) and Saliva Cortisol testing. The RESTQ-Sport is used to evaluate the subject's psychological recovery status. The Saliva Cortisol testing will be used to measure the physiological response to training and recovery.

Prior to data collection, participants are required to have qualified to compete for a varsity position on the college or university wrestling team. If voluntary consent is given and each participant qualifies to compete on the varsity team he is then eligible and may become a subject. Each subject will complete the Recovery-Stress Questionnaire for Athletes, and provided a saliva sample. Each participant will be instructed on the proper technique for answering questions and obtaining a saliva sample by the researcher and based upon established protocols. The data collection will take 13 weeks to gather the information pertinent for this study. The collection time is at smaller intervals to add sensitivity based upon previous studies and time points.

The Recovery-Stress Questionnaire for Athletes will be administered each time a saliva sample is obtained. The subjects will be given the questionnaire and seated at a desk where they will respond to the questions in the questionnaire.

The saliva collection protocol will be followed in accordance with the guidelines prescribed. Each subject will be given a cup of water in order for the subjects to thoroughly rinse the mouth by swishing one mouthful of water in the attempt to cleanse teeth and mouth cavity of unwanted materials. The subjects will be provided a 50 ml conical centrifuge tube labeled with the subjects identifying number. The subjects should slowly expectorate 10 ml of un-stimulated saliva this process could take approximately 10 minutes. The samples will be placed upon ice until they are placed in the centrifuge. The samples will then be placed in the centrifuge for clarifying and spun for 10 minutes at 3300 rpm at 4 degrees C. The samples once centrifuged will be carefully removed from the centrifuge and 2 ml pipets used to aliquot the “clarified saliva” into 0.5 ml storage tubes and then placed in a minus 80 degree freezer until shipping to laboratory for testing.

### ***Duration of Participation***

A total of 13 weeks of participation will be required of each subject. These include filling out informed consent, donation saliva samples and completion of questionnaire the day of the scheduled collection. Time for sample collection will take approximately 15 minutes. The written questionnaire will take 50 minutes to complete resulting in a maximum of 1.5 hours spent completing the data collection during the 13 week period.

### ***Benefits to the Individual***

Benefits to the individual include information on how your saliva immune proteins (IgA, Cortisol and cytokines) respond to participation in a competitive collegiate wrestling. Salivary cytokines have been utilized as markers of the body's immune response to exercise. The results of this study will help to clarify the response of immune system and the psychological response to a competitive wrestling season. We hope to assist in providing information necessary to prevent or reduce the incidence of illness in athletes, thus improving their overall health. In addition, provide information that may aid in the development of a scientifically based training program.

## APPENDIX C

### Oklahoma State University Institutional Review Board

Date: Monday, December 19, 2005  
IRB Application No ED0659  
Proposal Title: Overtraining Syndrome a study to determine the correlation between the physiological symptoms and the psychological signs  
Reviewed and Processed as: Expedited

**Status Recommended by Reviewer(s): Approved Protocol Expires: 12/18/2006**

Principal Investigator(s)

Daniel L. Dodson 619 Rimrock Road Edmond, OK 73034	Steven Edwards 432 Willard Stillwater, OK 74078
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The IRB application referenced above has been approved. It is the judgment of the reviewers that the rights and welfare of individuals who may be asked to participate in this study will be respected, and that the research will be conducted in a manner consistent with the IRB requirements as outlined in section 45 CFR 46.

The final versions of any printed recruitment, consent and assent documents bearing the IRB approval stamp are attached to this letter. These are the versions that must be used during the study.

As Principal Investigator, it is your responsibility to do the following:

1. Conduct this study exactly as it has been approved. Any modifications to the research protocol must be submitted with the appropriate signatures for IRB approval.
2. Submit a request for continuation if the study extends beyond the approval period of one calendar year. This continuation must receive IRB review and approval before the research can continue.
3. Report any adverse events to the IRB Chair promptly. Adverse events are those which are unanticipated and impact the subjects during the course of this research; and
4. Notify the IRB office in writing when your research project is complete.

Please note that approved protocols are subject to monitoring by the IRB and that the IRB office has the authority to inspect research records associated with this protocol at any time. If you have questions about the IRB procedures or need any assistance from the Board, please contact Beth McTernan in 415 Whitehurst (phone: 405-744-5700, beth.mcternan@okstate.edu).

Sincerely,



Sue C. Jacobs, Chair  
Institutional Review Board

## APPENDIX D

Written copy of Informed Consent to be provided to University of Central Oklahoma Wrestling Athletes

RESEARCH PARTICIPANT CONSENT FORM  
SALIVARY IMMUNE RESPONSE AND PSYCHOLOGICAL RECOVERY  
QUESTIONNAIRE TO A COMPETITIVE COLLEGIATE WRESTLING SEASON  
Principal Investigators: Daniel L. Dodson, M.Ed., A.T.,C.  
Oklahoma State University  
Department of Health, Leisure, and Human Performance

The present research project will examine the effect of a collegiate wrestling season on the presence of immune markers present saliva and the correlated scores on the Rest-Recovery Questionnaire for Sport. You have been selected because you are an University varsity wrestling athlete. You will be asked to provide data samples based on time points throughout the season, and post-season. You will receive a copy of this consent form to keep for your records.

### *Purpose of Research*

The purpose of this project is to assess the influence of a competitive wrestling season salivary immune markers and recovery scores on the Rest-Recovery Questionnaire for Sport.

### *Requirements of Participants*

5. Received a physical examination and cleared to participate in competitive sports.
6. Complete informed consent forms.
7. Provide saliva samples that will be taken the on the dates specified.
8. You will be asked to report any injuries in the mouth due to abrasive brushing, poor oral health, etc. before saliva sample collection. Also, we ask that you do not brush your teeth within 3 hours prior to sample collection. Dental work should not be performed within 24 hours prior to sample collection. In addition, do not eat a meal within 60 minutes and avoid alcohol 24 hours prior to sample collection.



### ***Duration of Participation***

A total of 13 weeks of participation will be required of each subject. These include filling out informed consent, donation saliva samples and completion of questionnaire the day of the scheduled collection. Time for sample collection will take approximately 15 minutes. The written questionnaire will take 50 minutes to complete resulting in a maximum of 1.5 hours spent completing the data collection during the 13 week period.

### ***Benefits to the Individual***

Benefits to the individual include information on how your saliva immune proteins (Cortisol) respond to participation in a competitive collegiate wrestling. Salivary cortisol has been utilized as a marker of the body's immune response to exercise. The results of this study will help to clarify the response of immune system and the psychological response to a competitive wrestling season. We hope to assist in providing information necessary to prevent or reduce the incidence of illness in athletes, thus improving their overall health. In addition, provide information that may aid in the development of a scientifically based training program.

### **Risks to the Individual**

There are no known risks involved with saliva donation. There are no known risks involved with responding to the questionnaire.

### ***Medical liability***

I understand the risks associated with this study and voluntarily choose to participate. I understand that in case of injury or illness resulting from this study, emergency medical care is available through community health care providers by dialing 911. I understand that no funds have been set aside by Oklahoma State University to compensate me in the event of illness or injury.

### ***Confidentiality***

All scientific and personal data collected on subjects will be kept confidential and stored in a locked file cabinet in the principle investigators office. This information will be available only to the Principal investigator. All subjects will be assigned a subject number and all samples being analyzed by lab technicians will be labeled with these subject numbers to ensure anonymity. Information concerning assigned subject numbers will be kept separate from confidential material. At no time will any of the subjects' personal information be divulged. All data will be reported as means and standard errors. E-mail communications will be kept strictly confidential; all emails will be deleted upon completion of data collection. Access to the completed questionnaires will be limited to the Primary investigators, and hard copy forms will be coded, to ensure anonymity. All hard copies of data will be kept until entered into computer file for analysis, and the data will be stored in a locked file cabinet in the primary investigators office until completion

of data collection and then destroyed. The Institutional Review Board has authority to inspect all forms and data collected.

***Voluntary Nature of Participation***

Participation in this research project is voluntary and you can withdraw your involvement in the project at any time without penalty.

***Human Subject Statement***

For any questions you may have about this research project, contact Daniel L. Dodson, M.Ed. (405) 255-7208 or Dr. Steven Edwards (405) 744-7476. If you have any concerns dealing with subjects' rights, contact Dr Sue C. Jacobs, IRB chair, 415 Whitehurst Hall, Oklahoma State University, 405-744-1676, Dr. John Garic, Associate Dean Jackson College of Graduate Studies and Research, 405-974-3341

I HAVE READ AND FULLY UNDERSTAND THE CONSENT FORM. I SIGN IT FREELY AND VOLUNTARILY. A COPY OF THIS FORM HAS BEEN GIVEN TO ME.

\_\_\_\_\_  
Signature of Participant

\_\_\_\_\_  
Date of birth

\_\_\_\_\_  
Date

I certify that I have personally explained this document before requesting that the participant sign it.

\_\_\_\_\_  
Signature of Researcher

\_\_\_\_\_  
Date

## APPENDIX E



*Dr. Joe C. Jackson College  
of Graduate Studies & Research*

December 21, 2005

Daniel L. Dodson  
619 Rimrock Road  
Edmond, OK 73034

Re: Application for IRB Review of Human Subjects Research

Dear Mr. Dodson:

The Jackson College of Graduate Studies and Research (JCGS&R) is pleased to inform you of the approval of your application for review of human subjects research by the UCO Institutional Review Board on the research proposal *"Overtraining Syndrome a study to determine the correlation between the physiological symptoms and the psychological signs"*, with the following provisos/contingencies:

1. The Consent Form included in your IRB materials is specific to Oklahoma State University and not to the University of Central Oklahoma. It will be necessary to have a consent form specific to the wrestling athletes of the University of Central Oklahoma.
2. Written authorization from David James, Head UCO Wrestling Coach, to conduct this research must be secured and submitted to the JCGS&R prior to any contact with UCO athletes.
3. Prior to any data analysis, written assurance from Cytovance Biologics must be secured and submitted to the JCGS&R that Cytovance Biologics and its employees, assigns and designees:
  - a. will not conduct any DNA analysis of the subject samples; and
  - b. further will not keep any permanent record of the study data; and
  - c. further that it will not divulge any study data to anyone other than the Principal Investigator, Mr. Daniel L. Dodson; and,
  - d. that it will either completely destroy any remaining saliva samples or return them to the PI.

If the JCGS&R can be of any further assistance in your pursuit of research, please do not hesitate to contact us.

With kind regards, I am


Sincerely,

Dr. John M. Garic  
Associate Dean  
Jackson College of Graduate Studies and Research

JMG/

cc: Dr. S. N. Rao, Dean

*100 North University Drive, Nigh University Center, Room 404, Edmond, Oklahoma 73034-5209 • Phone: (405) 974-334  
Fax: (405) 974-3852 • Email: gradcoll@ucok.edu*



**BRONCHO UCO BRONCHOS WRESTLING**

NATIONAL  
CHAMPIONS

2003  
2002  
1995  
1994  
1993  
1992  
1989  
1987  
1986  
1985  
1984  
1982  
1981  
1979

58 Individual  
National  
Champions

183 Individual  
All-Americans

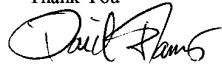
14 Straight  
Regional  
Championships  
1990-2003

December 21, 2005

Dear Dr. Garic:

I am writing this letter to inform you that I have spoken to and met with Dan Dodson about doing a research study on the UCO wrestling team. The study will concern overtraining syndrome, and include saliva testing and answering questionnaires. Please know that we will fully support this study and we are excited to participate. Please feel free to call me with any questions.

Thank You



David James  
Head Wrestling Coach

**NCAA**®

UNIVERSITY OF CENTRAL OKLAHOMA WRESTLING  
David James, Head Coach  
100 North University Drive, Edmond, Oklahoma 73034  
(405) 974-2509 • Fax (405) 974-3820 • [www.bronchosports.com](http://www.bronchosports.com)

**LSC**

Dr. John M. Garic  
Associate Dean  
Jackson College of Graduate Studies and Research  
100 North University Drive  
Nigh University Center, Room 404  
Edmond, OK 73034-5209

January 5, 2006

Dear Dr. Garic,

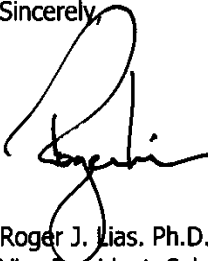
IRB Review of Human Subjects Research – Daniel L. Dodson

With respect to work to be undertaken by Bio Manufacturing Holdings LLC (doing business as Cytovance Biologics) relating to the analysis of salivary samples in support of Daniel L. Dodson's research "*Overtraining Syndrome – a study to determine the correlation between the physiological symptoms and psychological signs*", I am happy to provide written assurance that Bio Manufacturing Holdings LLC, its employees, assigns and designees:

- o Will not conduct any DNA analysis of the subject samples
- o Will not keep any permanent record of the study data
- o Will not divulge any study data to anyone other than Mr. Daniel L. Dodson; and
- o Will either completely destroy any remaining saliva samples or return them to Mr. Daniel L. Dodson

Please do not hesitate to contact me if I can be of any further assistance.

Sincerely,



Roger J. Lias, Ph.D.  
Vice President, Sales & Business Development

cc: Daniel L. Dodson

## APPENDIX F



### PROPOSAL FOR ANALYTICAL TESTING OF SALIVARY SAMPLES

Prepared For: Dan Dodson

Submitted By: Roger Lias, VP Sales & Business Development  
Bio Manufacturing Holdings LLC  
840 Research Parkway, Suite 400  
Oklahoma City, OK 73104  
Office: (405) 488-1488 ext. 400  
Mobile: (919) 349-2770  
rlia@Cytovance.com

Issue Date: December 2, 2005

Revision: 1

The content of this document has been developed on a project-specific basis based on information provided by Dan Dodson and on a number of key assumptions. This proposal is provided for the sole use of Dan Dodson in assessing the merits of services provided by Cytovance Biologics.

#### **SCOPE OF WORK – Analytical Testing of Salivary Samples**

Cytovance Biologics has developed a preliminary scope of work based on information provided by Dan Dodson. This preliminary scope has been used to develop the pricing provided in this proposal. Data generated during this study will be used for Dan Dodson's Ph.D. dissertation defense.

Cytovance Biologics has not received detailed technical information pertaining to assays requested. In order to complete the preliminary scope of work it has, therefore, been necessary to make a number of general assumptions which are provided below.

Should this proposal be acceptable to Dan Dodson, it will be necessary to hold detailed technical meetings in order to finalize a work program and pricing.

#### **Program Assumptions**

- Testing will be required to be performed in 2 phases.
- An additional stage (method evaluation stage) will be necessary for IL-6 assay prior to analyzing samples to determine suitability of assay and solutions to be utilized.
- Assays are adequate for the matrix.
- Assays have adequate detection limits and sensitivity.
- Methods will be run by manufacturer method. No method development or optimization will be necessary or performed.
- Samples will be thawed once and stored at 4°C until testing completed. Cortisol will be tested last. Testing order is not critical.
- All samples will be run at one dilution in duplicate.
- Testing will take place over a period of approximately 2 weeks per phase.
- Results will be delivered at the conclusion of each phase.
- Reserves of each sample will be retained by the submitter to be tested as necessary.
- Method evaluation will commence approximately January 2006. Phase 1 testing will be performed approximately February 2006; phase 2 testing will occur approximately May 2006.

**EXECUTIVE SUMMARY**

Cytovance Biologics, Inc. is pleased to submit this proposal for Analytical Testing of Salivary Samples in support of Mr. Dan Dodson's Ph.D. research and dissertation.

Cytovance Biologics has the expertise, facilities and equipment necessary to support the requested analytical services and resources are available in the requested timeframe.

Based on current assumptions, program cost is estimated at \$11,600 and materials cost at \$8,140 plus tax.

Cytovance will require final agreement and sign-off on a detailed scope of work and execution of a definitive services agreement or arrangement of a suitable purchase order mechanism prior to sourcing of materials and/or commencement of work.

Roger Lias, Ph.D.  
Vice President, Sales & Business Development  
rlas@cytovance.com  
(919) 349-2770



## APPENDIX G

---

# REST Q - 76 Sport

---

Single Code: \_\_\_\_\_ Group Code: \_\_\_\_\_  
Name (Last): \_\_\_\_\_ (First): \_\_\_\_\_  
Date: \_\_\_\_\_ Time: \_\_\_\_\_ Age: \_\_\_\_\_ Gender: \_\_\_\_\_  
Sport/Event(s): \_\_\_\_\_

This questionnaire consists of a series of statements. These statements possibly describe your mental, emotional, or physical well-being or your activities during the past few days and nights.

Please select the answer that most accurately reflects your thoughts and activities. Indicate how often each statement was right in your case in the past days.

The statements related to performance should refer to performance during competition as well as during practice.

For each statement there are seven possible answers.

Please make your selection by marking the number corresponding to the appropriate answer.

**Example:**

*In the past (3) days/nights*

*... I read a newspaper*

0      1      2      3      4      5      6  
never   seldom   sometimes   often   more often   ~~very often~~   always

In this example, the number 5 is marked. This means that you read a newspaper very often in the past three days.

Please do not leave any statements blank.

If you are unsure which answer to choose, select the one that most closely applies to you.

Please turn the page and respond to the statements in order without interruption.

---

From *Recovery-Stress Questionnaire for Athletes: User Manual* by Michael Kellmann and K. Wolfgang Kallus, 2001, Champaign, IL: Human Kinetics. C.1

*In the past (3) days/nights*

1) ... I watched TV

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

2) ... I did not get enough sleep

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

3) ... I finished important tasks

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

4) ... I was unable to concentrate well

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

5) ... everything bothered me

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

6) ... I laughed

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

7) ... I felt physically bad

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

8) ... I was in a bad mood

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

9) ... I felt physically relaxed

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

10) ... I was in good spirits

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

11) ... I had difficulties in concentrating

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

12) ... I worried about unresolved problems

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

---

C.2 From *Recovery-Stress Questionnaire for Athletes: User Manual* by Michael Kellmann and K. Wolfgang Kallus, 2001, Champaign, IL: Human Kinetics.

***In the past (3) days/nights***

13) ... *I felt at ease*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

14) ... *I had a good time with friends*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

15) ... *I had a headache*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

16) ... *I was tired from work*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

17) ... *I was successful in what I did*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

18) ... *I couldn't switch my mind off*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

19) ... *I fell asleep satisfied and relaxed*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

20) ... *I felt uncomfortable*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

21) ... *I was annoyed by others*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

22) ... *I felt down*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

23) ... *I visited some close friends*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

24) ... *I felt depressed*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

From *Recovery-Stress Questionnaire for Athletes: User Manual* by Michael Kellmann and K. Wolfgang Kallus, 2001, Champaign, IL: Human Kinetics. C3

***In the past (3) days/nights***

25) ... *I was dead tired after work*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

26) ... *other people got on my nerves*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

27) ... *I had a satisfying sleep*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

28) ... *I felt anxious or inhibited*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

29) ... *I felt physically fit*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

30) ... *I was fed up with everything*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

31) ... *I was lethargic*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

32) ... *I felt I had to perform well in front of others*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

33) ... *I had fun*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

34) ... *I was in a good mood*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

35) ... *I was overtired*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

36) ... *I slept restlessly*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

---

C.4 From *Recovery-Stress Questionnaire for Athletes: User Manual* by Michael Kellmann and K. Wolfgang Kallus, 2001, Champaign, IL: Human Kinetics.

***In the past (3) days/nights***

37) ... *I was annoyed*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

38) ... *I felt as if I could get everything done*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

39) ... *I was upset*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

40) ... *I put off making decisions*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

41) ... *I made important decisions*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

42) ... *I felt physically exhausted*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

43) ... *I felt happy*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

44) ... *I felt under pressure*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

45) ... *everything was too much for me*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

46) ... *my sleep was interrupted easily*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

47) ... *I felt content*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

48) ... *I was angry with someone*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

***In the past (3) days/nights***

49) ... *I had some good ideas*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

50) ... *parts of my body were aching*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

51) ... *I could not get rest during the breaks*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

52) ... *I was convinced I could achieve my set goals during performance*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

53) ... *I recovered well physically*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

54) ... *I felt burned out by my sport*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

55) ... *I accomplished many worthwhile things in my sport*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

56) ... *I prepared myself mentally for performance*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

57) ... *my muscles felt stiff or tense during performance*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

58) ... *I had the impression there were too few breaks*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

59) ... *I was convinced that I could achieve my performance at any time*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

60) ... *I dealt very effectively with my teammates' problems*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

---

C.6 From *Recovery-Stress Questionnaire for Athletes: User Manual* by Michael Kellmann and K. Wolfgang Kallus, 2001, Champaign, IL: Human Kinetics.

*In the past (3) days/nights*

61) ... *I was in a good condition physically*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

62) ... *I pushed myself during performance*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

63) ... *I felt emotionally drained from performance*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

64) ... *I had muscle pain after performance*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

65) ... *I was convinced that I performed well*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

66) ... *too much was demanded of me during the breaks*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

67) ... *I psyched myself up before performance*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

68) ... *I felt that I wanted to quit my sport*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

69) ... *I felt very energetic*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

70) ... *I easily understood how my teammates felt about things*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

71) ... *I was convinced that I had trained well*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

72) ... *the breaks were not at the right times*

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

*In the past (3) days/nights*

73) ... I felt vulnerable to injuries

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

74) ... I set definite goals for myself during performance

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

75) ... my body felt strong

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

76) ... I felt frustrated by my sport

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

77) ... I dealt with emotional problems in my sport very calmly

0	1	2	3	4	5	6
never	seldom	sometimes	often	more often	very often	always

*Thank you very much!*



## APPENDIX H



### EXPANDED RANGE High Sensitivity SALIVARY CORTISOL ENZYME IMMUNOASSAY KIT

Catalog No. 1-3002/1-3012, 96-Well Kit  
For Research Use

#### Intended Use

Salimetrics cortisol kit is a competitive immunoassay specifically designed for the quantitative measurement of salivary cortisol. It is not intended for use with serum/plasma or for diagnostic use. It is intended only for research use with saliva. Please read the complete kit insert before performing this assay. For further information about this kit, its application, or the procedures in this insert, please contact the technical service team at Salimetrics or your local sales representative.

#### Introduction

Historically, the immunodiagnostic community's approach to the application of immunoassay techniques in the measurement of biomarkers in saliva has been problematic. This assay kit was designed to address those problems. First, prior to the late 1990s the majority of available immunoassays for saliva cortisol were modifications of protocols developed for the use with serum/plasma. The standards used in those assay kits were suspended in a human serum matrix. Given that the composition of serum is markedly different from saliva, those standards are likely to produce results that are influenced by matrix differences. To ensure the most accurate results, this salivary immunoassay uses a matrix that matches saliva. Second, the level of cortisol in saliva is significantly lower than levels in the general circulation. The use of a standard curve developed to capture the range of values expected in serum/plasma samples is often not sensitive enough to capture the complete range of individual differences in the level expected in saliva. This assay was designed to capture the full range of salivary cortisol levels (0.003 to 3.0 µg/dL) while using only 25 µL of saliva per test. Third, the pH of saliva is easily lowered or raised by the consumption of food or drink. Performance of immunoassays becomes compromised as the pH of samples to be tested drops below 4 (1). This results in artificially inflated levels. This assay system is designed to be resilient to the effects of interference caused by collection techniques that affect pH. In addition, a built-in pH indicator warns the user of acidic or basic samples.

#### Test Principle

A microtitre plate is coated with monoclonal antibodies to cortisol. Cortisol in standards and unknowns compete with cortisol linked to horseradish peroxidase for the antibody binding sites. After incubation, unbound components are washed away. Bound cortisol peroxidase is measured by the reaction of the peroxidase enzyme on the substrate tetramethylbenzidine (TMB). This reaction produces a blue color. A yellow color is formed after stopping the reaction with sulfuric acid. Optical density is read on a standard plate reader at 450 nm. The amount of cortisol peroxidase detected is inversely proportional to the amount of cortisol present (2).

#### pH Indicator

A pH indicator in the assay diluent alerts the user to samples with high or low pH values. Acidic samples will turn the diluent yellow. Alkaline samples will turn the diluent purple. Dark yellow or purple wells indicate that a pH value for that sample should be obtained using pH strips. Cortisol values from samples with a pH < 3.5 or > 9.0 may be artificially inflated or lowered (1).

#### Precautions

1. Liquid stop solution is a 2-molar solution of sulfuric acid. This solution is caustic; use with care. Stop solution in powdered form is not sulfuric acid-based, and is mildly corrosive.
2. This kit uses break-apart microtitre strips. Unused wells must be stored at 2 - 8°C in the sealed foil pouch and used in the frame provided.

Date: 9-26-06

3. Do not mix components from different lots of kits.
4. When using a multichannel pipette, reagents should be added to duplicate wells at the same time. Follow the same sequence when adding additional reagents so that incubation time with reagents is the same for all wells.
5. See "Material Safety Data" at the end of procedure.
6. As for all quantitative assays for salivary analytes, we recommend that samples be screened for possible blood contamination (3,4). This can be efficiently and economically accomplished using Salimetrics Blood Contamination BIA Kit (Cat. No.: 1-1302/1-1312). Do not use dipsticks, which result in false positive values due to salivary enzymes. Routine calibration of pipettes is critical for the best possible assay performance.
7. Pipetting of samples and reagents must be done as quickly as possible (without interruption) across the plate.
8. When running multiple plates, or multiple sets of strips, a standard curve should be run with each individual plate and/or set of strips.
9. The temperature of the laboratory may affect assays. Salimetrics' kits have been validated at 68 - 74°F (20 - 23.3°C). Higher or lower temperatures will cause an increase or decrease in OD values, respectively. Salimetrics cannot guarantee test results outside of this temperature range.

**Storage** All components of this kit are stable at 2 - 8°C until the kit's expiration date.

#### Reagents and Reagent Preparation

1. **Anti-Cortisol Coated Plate:** A ready-to-use 96-well microtitre plate pre-coated with monoclonal anti-cortisol antibodies in a resealable foil pouch.
  2. **Cortisol Standards:** Six vials, 500 µL each, labeled A-F, containing cortisol concentrations of 3,000, 1,000, 0.333, 0.111, 0.037, and 0.012 µg/dL, in a synthetic saliva matrix with a non-mercury preservative. (Values in nmol/L are 82.77, 27.59, 9.19, 3.06, 1.02, and 0.33 nmol/L, respectively.) Standards are traceable to the NIST standard.
  3. **Wash Buffer:** 100 mL of a 10X phosphate buffered solution containing detergents and a non-mercury preservative. Dilute only the amount needed for current day's use. Discard any leftover reagent. Dilute the wash buffer concentrate 10-fold with room temperature deionized water (100 mL of 10X wash buffer to 900 mL of deionized H<sub>2</sub>O). (Note: If precipitate has formed in the concentrated wash buffer, it may be heated to 60°C for 15 minutes. Cool to room temperature before use in assays.)
  4. **Assay Diluent:** 63 mL of a phosphate buffered solution containing a pH indicator and a non-mercury preservative.
  5. **Enzyme Conjugate:** 50 µL of a solution of cortisol labeled with horseradish peroxidase. Dilute prior to use with assay diluent.
  6. **Tetramethylbenzidine (TMB):** 25 mL of a non-toxic ready to use solution.
  7. **Stop Solution:** 12.5 mL of a solution of sulfuric acid (USA customers only). Stop solution is provided in powdered form (not sulfuric acid-based) to customers outside the USA. Reconstitute the powdered stop solution with 12.5 mL of deionized water. Let sit for 10 minutes before use.
  8. **Non-specific Binding Wells:** These wells do not contain anti-cortisol antibody. In order to support multiple-use, a strip of NSB wells is included. They are located in the foil pouch. Wells may be broken off and inserted where needed.
- Note:** The quantity of reagent provided with break-apart kits is sufficient for three individual runs. The volume of diluent and conjugate used for assays using less than a full plate should be scaled down accordingly, keeping the same dilution ratio.

#### Materials Needed But Not Supplied

- Precision pipette to deliver 15 and 25 µL
- Precision multichannel pipette to deliver 50 µL, and 200 µL
- Vortex
- Plate rotator (if unavailable, tap to mix)
- Plate reader with a 450 nm filter
- Log-linear graph paper or computer software for data reduction
- Deionized water
- Reagent reservoirs
- One disposable tube capable of holding 24 mL
- Pipette tips
- Serological pipette to deliver up to 24 mL

**Specimen Collection**

The preferred saliva collection method is to collect saliva by passive drool allowing the saliva to pass through a short straw into a polypropylene vial. Samples may also be collected using Sorbettes, P/N 5029 (for infants) or cotton ropes, P/N 5016. For accurate results collection devices should be completely saturated before removal. Do not add sodium azide to saliva samples as a preservative. Samples visibly contaminated with blood should be recollected. Freeze at -20°C or lower for long-term storage. Contact the technical service team at Salimetrics for more detailed information on specimen collection.

Saliva samples should be frozen prior to assay to precipitate the mucins. On day of assay, thaw completely, vortex, and centrifuge at 1500 x g (@3000 rpm) for 15 minutes. Pipette clear sample into appropriate wells. Particulate matter may interfere with antibody binding, leading to falsely elevated results.

**Procedure**

Bring all reagents to room temperature.

**Step 1:** Determine your plate layout. Here is a suggested layout.

	1	2	3	4	5	6	7	8	9	10	11	12
A	3.000 Std	3.000 Std	C-H	C-H								
B	1.000 Std	1.000 Std	C-L	C-L								
C	0.333 Std	0.333 Std	Unk-1	Unk-1								
D	0.111 Std	0.111 Std	Unk-2	Unk-2								
E	0.037 Std	0.037 Std	Unk-3	Unk-3								
F	0.012 Std	0.012 Std	Unk-4	Unk-4								
G	Zero	Zero	Unk-5	Unk-5								
H	NSB	NSB	Unk-6	Unk-6								

**Step 2:** Keep the desired number of strips in the strip holder and place the remaining strips back in the foil pouch. If you choose to place non-specific binding wells in H-1, 2, remove strips 1 and 2 from the strip holder and break off the bottom wells. Place the strips back into the strip holder leaving H-1, 2 blank. Break off 2 NSB wells from the strip of NSBs included in the foil pouch. Place in H-1, 2. Alternatively, NSBs may be placed wherever you choose on the plate. Reseal the zip-lock and refrigerate the pouch at 2 - 8°C. **Caution:** Extra NSB wells should not be used for determination of standards, controls or unknowns.

**Step 3:**

- Pipette 24 mL of assay diluent into a disposable tube. Set aside for Step 5.

**Step 4:**

- Pipette 25 µL of standards, controls and unknowns into appropriate wells. Standards, controls and unknowns should be assayed in duplicate.
- Pipette 25 µL of assay diluent into 2 wells to serve as the zero.
- Pipette 25 µL of assay diluent into each NSB well.

**Step 5:** Make a 1:1600 dilution of the conjugate, by adding 15 µL of the conjugate to the 24 mL of assay diluent prepared in Step 3, (full plate only). Immediately mix the diluted conjugate solution and pipette 200 µL into each well using a multichannel pipette.

**Step 6:** Mix plate on rotator for 5 minutes at 500 rpm (or tap to mix) and incubate at room temperature for an additional 55 minutes.

**Step 7:** Wash the plate 4 times with 1X wash buffer. A plate washer is recommended. However, washing may be done by gently squirting wash buffer into each well with a squirt bottle or by pipetting 300µL of wash buffer into each well, and then discarding the liquid by inverting the plate over a sink. After each wash, the plate should be thoroughly blotted on paper towels before being turned upright. *If using a plate washer, blotting is still recommended after the last wash.*

**Step 8:** Add 200 µL of TMB solution to each well with a multichannel pipette.

**Step 9:** Mix on a plate rotator for 5 minutes at 500 rpm (or tap to mix) and incubate the plate in the dark at room temperature for an additional 25 minutes.

**Step 10:** Add 50 µL of stop solution with a multichannel pipette.

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**Step 11:**

- Mix on a plate rotator for 3 minutes at 500 rpm (or tap to mix). **Caution:** Do not mix at speeds over 600 rpm.
- Wipe off bottom of plate with a water-moistened lint-free cloth and wipe dry.
- Read in a plate reader at 450 nm. Read plate within 10 minutes of adding stop solution (correction at 492 to 620 is desirable).

**Calculations**

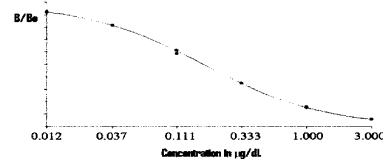
1. Compute the average optical density (OD) for all duplicate wells.
2. Subtract the average OD for the NSB wells from the average OD of the zero, standards, controls and unknowns.
3. Calculate the percent bound (B/Bo) for each standard, control and unknown by dividing the average OD (B) by the average OD for the zero (Bo).
4. Determine the concentrations of the controls and unknowns by interpolation using software capable of logistics. We recommend using a 4-parameter sigmoid minus curve fit.

**Typical Results**

The following charts and graphs are for illustration only and should not be used to calculate results from another assay.

Well	Sample	Average OD	B	B/Bo	Cortisol (µg/dL)
A1,A2	S1	0.094	0.071	0.048	3.000
B1,B2	S2	0.236	0.213	0.145	1.000
C1,C2	S3	0.524	0.501	0.340	0.333
D1,D2	S4	0.897	0.874	0.593	0.111
E1,E2	S5	1.219	1.196	0.812	0.037
F1,F2	S6	1.379	1.356	0.921	0.012
G1,G2	Bo	1.406	1.473	NA	NA
H1,H2	NSB	0.023	NA	NA	NA

**Example: Cortisol 4-Parameter Sigmoid Minus Curve Fit**



**Material Safety Data\***

**Hazardous Ingredients**

Liquid stop solution is caustic; use with care. *Note: Stop solution in powdered form is not sulfuric acid-based and is mildly corrosive.* We recommend the procedures listed below for all kit reagents.

**Handling**

Follow good laboratory procedures when handling kit reagents. Laboratory coats, gloves, and safety goggles are recommended. Wipe up spills using standard absorbent materials while wearing protective clothing. Follow local regulations for disposal.

**Emergency Exposure Measures**

In case of contact, immediately wash skin or flush eyes with water for 15 minutes. Remove contaminated clothing. If inhaled, remove individual to fresh air. If individual experiences difficulty breathing, give oxygen and call a physician.

\*The above information is believed to be accurate but is not all-inclusive. This information should only be used as a guide. Salimetrics shall not be liable for accidents or damage resulting from contact with reagents.

### HS Cortisol EIA Assay Performance Characteristics

**Recovery:** Six saliva samples containing different levels of endogenous cortisol were spiked with known quantities of cortisol and assayed.

Sample	Endogenous (µg/dL)	Added (µg/dL)	Expected (µg/dL)	Observed (µg/dL)	Recovery (%)
1	0.088	2.000	2.088	2.176	104.2
2	0.077	0.500	0.577	0.580	100.8
3	0.062	0.011	0.073	0.071	97.3
4	0.066	2.500	2.566	2.723	106.1
5	0.210	0.330	0.510	0.508	99.6
6	0.086	0.011	0.097	0.094	96.9

#### Precision:

1. The intra-assay precision was determined from the mean of 14 (low) and 18 (high) replicates each.

Sample	N	Mean (µg/dL)	Standard Deviation (µg/dL)	Coefficient of Variation (%)
Level 1	18	0.999	0.033	3.35
Level 2	14	0.097	0.004	3.65

2. The inter-assay precision was determined from the mean of average duplicates for 12 separate runs.

Sample	N	Mean (µg/dL)	Standard Deviation (µg/dL)	Coefficient of Variation (%)
Level 1	12	1.020	0.038	3.75
Level 2	12	0.101	0.006	6.41

**Linearity of Dilution:** Two saliva samples were diluted with assay diluent and assayed.

Sample	Dilution Factor	Expected (µg/dL)	Observed (µg/dL)	Recovery (%)
1			2.176	
	1:2	1.088	1.065	97.9
	1:4	0.544	0.503	92.5
	1:8	0.272	0.233	85.7
	1:16	0.136	0.109	80.1
2			0.508	
	1:2	0.254	0.247	97.2
	1:4	0.127	0.118	92.9
	1:8	0.064	0.058	90.6
	1:16	0.032	0.031	96.9

**Sensitivity:** The lower limit of sensitivity was determined by interpolating the mean minus 2 SD's for 10 sets of duplicates at 0 µg/dL standard. The minimal concentration of cortisol that can be distinguished from 0 is < 0.003 µg/dL.

**Correlation with Serum:** The correlation between serum and saliva cortisol was determined by assaying 49 matched samples using the Diagnostic Systems Laboratories' serum Cortisol EIA and the Salimetrics ER HS Salivary Cortisol EIA.

The correlation between saliva and serum was highly significant,  $r(47) = 0.91$ ,  $p < 0.0001$ .

#### Specificity of Antiserum

Compound	Spiked Concentration (ng/mL)	% Cross-reactivity in ER HS Salivary Cortisol EIA
Prednisolone	100	0.568
Prednisone	1000	ND
Cortisone	1000	0.130
11-Deoxycortisol	500	0.156
21-Deoxycortisol	1000	0.041
17 $\alpha$ -Hydroxyprogesterone	1000	ND
Dexamethasone	1000	19.2
Triamcinolone	1000	0.086
Corticosterone	10,000	0.214
Progesterone	1000	0.015
17 $\beta$ -Estradiol	10	ND
DHEA	10,000	ND
Testosterone	10,000	0.006
Transferrin	66,000	ND
Aldosterone	10,000	ND

ND = None detected (<0.004)

Date: 9-26-06

### References

- Schwartz, E.B., Granger, D.A., Susman, E.J., Gunnar, M.R., & Laird, B. (1998). Assessing salivary cortisol in studies of child development. *Child Development*, 69, 1503-1513.
- Chard, T. (1990). *An introduction to radioimmunoassay and related techniques*. Amsterdam: Elsevier.
- Kivlighan, K. T., Granger, D. A., Schwartz, E. B., Nelson, V., & Curran, M. (2004). Quantifying blood leakage into the oral mucosa and its effects on the measurement of cortisol, dehydroepiandrosterone, and testosterone in saliva. *Hormones and Behavior*, 46, 39-46.
- Schwartz, E., & Granger, D. A. (2004). Transferrin enzyme immunoassay for quantitative monitoring of blood contamination in saliva. *Clinical Chemistry*, 50, 654-656.

### Seller's Limited Warranty

"Seller warrants that all goods sold hereunder will be free from defects in material and workmanship. Upon prompt notice by Buyer of any claimed defect, which notice must be sent within thirty (30) days from date such defect is first discovered and within three months from the date of shipment, Seller shall, at its option, either repair or replace the product that is proved to Seller's satisfaction to be defective. All claims should be submitted in writing. This warranty does not cover any damage due to accident, misuse, negligence, or abnormal use. Liability in all cases, will be limited to the purchased cost of the kit.

It is expressly agreed that this limited warranty shall be in lieu of all warranties of fitness and in lieu of the warranty of merchantability. Seller shall not be liable for any incidental or consequential damages that arise out of the installation, use or operation of Seller's product or out of the breach of any express or implied warranties."

## VITA

Daniel LeGrand Dodson

Candidate for the Degree of

Doctor of Philosophy

Thesis: OVER-TRAINING SYNDROME A STUDY TO DETERMINE THE CORRELATION BETWEEN THE PHYSIOLOGICAL SYMPTOMS AND THE PSYCHOLOGICAL SIGNS IN COLLEGE WRESTLERS

Major Field: Health, Leisure, and Human Performance

### Biographical:

Personal Data: Born in Danville, Indiana, on March 19, 1963 the son of Donald K and B Carol Dodson. Married to the former Jennifer R. Roskelley, on February 4, 1994. The father of Annaica R and Andrew L.

Education: Graduated Danville Community High School in Danville, IN in May 1981; received an Associates of Science from Snow College, Ephraim, UT in June 1987; received a Bachelors of Science from Utah State University, Logan, UT in June 1990; received a Masters of Education from Utah State University, Logan, UT in August, 1992. Completed the requirements for the Doctorate of Philosophy degree at Oklahoma State University in May 2007.

Experience: Langston University, Langston, OK, September 1992-January 1993 and August 1995-July 2002, Head Athletic Trainer / Instructor; University of Central Oklahoma, Edmond, OK, August 2004-May 2005, Adjunct Faculty, Department of Kinesiology and Health Studies; Oklahoma State University, Stillwater, OK August 2005-May 2006, Visiting Lecturer, Exercise Physiology; The Physicians Group, Oklahoma City, OK, June 2006 to present, Director of Research and Education.

Professional Memberships: National Athletic Trainers' Association, Mid America Athletic Trainers' Association, Oklahoma Athletic Trainers' Association.

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Date of Degree: May, 2007

Institution: Oklahoma State University

Location: Stillwater, Oklahoma

Title of Study: OVER-TRAINING SYNDROME A STUDY TO DETERMINE THE CORRELATION BETWEEN THE PHYSIOLOGICAL SYMPTOMS AND THE PSYCHOLOGICAL SIGNS IN COLLEGE WRESTLERS

Pages in Study: 161

Candidate for the Degree of Doctor of Philosophy

Major Field: Health, Leisure, and Human Performance

### ABSTRACT

**PURPOSE:** The primary purpose of this study was to determine if there was a correlation between the physiological signs of over-training and the psychological signs of over-training in collegiate wrestlers. **METHOD:** The RESTQ-Sport Questionnaire was used to evaluate the subjects' psychological recovery status. The salivary cortisol testing was used to measure the physiological response to training and recovery. The Recovery-Stress Questionnaire for Athletes was administered each time a saliva sample was obtained. The saliva collection protocol was followed in accordance with the guidelines prescribed by Salimetrics. Each subject was given a cup of water, and then instructed to thoroughly rinse his mouth by swishing one mouthful of water in the attempt to cleanse the teeth and mouth cavity of unwanted materials. The subjects were provided a 2 ml tube labeled with the subjects' identifying numbers. The subjects slowly expectorated 2 ml of unstimulated saliva. This process took approximately 10 minutes. The samples were placed upon ice until they were delivered to Cytovance Biologics LLC for testing. Salimetrics high sensitivity salivary cortisol enzyme immunoassay kit was strictly followed. The collection time was conducted at weekly intervals for 12 weeks to add sensitivity. **RESULTS:** The results of the present investigation revealed a non significant Pearson correlation coefficient between salivary cortisol concentration and RESTQ-Sport recovery stress scores for wrestlers during a season and post season of competition in collegiate wrestling. The calculated coefficient between cortisol and general stress ( $r = 0.248$ ,  $p = .490$ ) revealed a lack of association between these two variables. Further, statistically non-significant correlations were uncovered between salivary cortisol and general recovery ( $r = 0.159$ ,  $p = .661$ ), between cortisol and sports specific stress ( $r = 0.153$ ,  $p = .673$ ), and between cortisol and sports specific recovery ( $r = -0.119$ ,  $p = .744$ ). **CONSLUCION:** The data in this study demonstrated that there were changes in salivary cortisol, stress, and recovery scores as a result of competing in a competitive collegiate wrestling program. The relationship between salivary cortisol and stress-recovery scores appeared to be some what independent and unrelated. Further studies are necessary to determine if there is a relationship between physiological responses to over-training and psychological signs of over-training.

ADVISER'S APPROVAL: Dr. Steven W. Edwards

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