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Effect of Dehydration on Concomitant Measures of Cognitive Function and Balance

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EFFECT OF DEHYDRATION ON CONCOMITANT MEASURES OF COGNITIVE FUNCTION AND BALANCE

by

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Bachelor of Science Degree in Athletic Training/Exercise Science
Ithaca College
2008

A thesis submitted in partial fulfillment of the requirements for the

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ABSTRACT

Effects of Dehydration on Concomitant Measures of Cognitive Function and Balance

by

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During activities of daily living, there are demands placed on the body both cognitively and physically. These demands are multiplied when performing an athletic endeavor. Many studies have demonstrated the negative impact dehydration has on cognitive and muscle functions, independently. The purpose of this study is to investigate the effect of dehydration on central nervous system and peripheral nervous system fatigue as expressed through concomitant factors of cognitive function and balance. Subjects were to consist of 20 participates aged 18-35 years old who reported running a minimum of 45 minutes per exercise bout at least 3 times per week. The measurement items were reaction time between an auditory stimulus and a verbal response, as well as errors on a BESS test. Reaction time and BESS testing were administered independently and concomitantly while subjects were euhydrated and dehydrated in separate sessions. Subjects were dehydrated pharmacologically using 40mg of
furosemide. Urine specific gravity (USG) and urine color (UC) were used as objective measures of hydration status. Results showed UC is an acceptable method of self-monitoring hydration status. When performed independently, males had better scores than females for BESS errors and reaction time. Divided attention measurements were split evenly in terms of reaction time. In terms of BESS errors, there was an increase in number of errors for the dehydrated condition versus the euhydrated condition. Due to extenuating circumstances, not enough subjects were able to participate to make results statistically significant. However, the results here suggest that future research would be warranted in gender differences and situational scenarios related dehydrations effects on both reaction time and balance.

**Keywords:** urine specific gravity, reaction time, BESS test, dehydration, cognitive function, balance
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Figure 1  RTT v. BESS Test ........................................................................................................5
Dehydration has been shown to negatively affect cognitive functioning and physical performance, the manifestation of neuromuscular coordination. This is important to not only everyday activity, but essential to an athletic endeavor. When performing a complicated motor skill, like running or throwing, communication between the brain and skeletal muscle must be quick, efficient, and precise. Activation of the agonist and antagonist muscles in the exact sequence at the correct time is the basis of neuromuscular coordination. Impairment in this process, either at the neural level or physiologically in the muscle, makes the person susceptible to injury. Being properly hydrated avoids these risks.

These deficits are also evident in concussion patients. There are several objective tests used to measure cognition and certain aspects of physical performance, like balance, which are used to indicate concussive status. However, a flaw in this system is that there is not a definitive cause and effect relationship. As these are also measures of dehydration, symptoms appearing to signify concussion may actual result from dehydration. Patel et al. (2007) administered concussion tests to subjects following exertional dehydration. The most important findings were in the Graded Symptoms Checklist (GSC), a subjective measure of physiological and neurological changes. The symptoms most often reported were fatigue/drowsiness, feeling slowed down, and difficulty
concentrating. After dehydration, the total symptom severity score increased significantly ($t_{23} = -7.673$, $p<.001$), as did the total number of reported symptoms ($t_{23} = -8.585$, $p<.001$) (Patel, 2007). This study shows that the psychological effects of dehydration can influence the outcome in the Standardized Assessment of Concussion (SAC), a cognitive measure, and the Balance Error Scoring System (BESS), a measure of muscle performance, tests as the most frequent symptoms affecting concentration and motivation.
Purpose of the Study

The purpose of this study is to investigate the effect of dehydration on central nervous system and peripheral nervous system fatigue as expressed through concomitant factors of cognitive function and balance.

Hypotheses

Hypothesis #1
Dehydration will result in a decreased score on the RTT and BESS tests when performed independently.

Hypothesis #2
Dehydration will lead to a decreased score on the RTT test, but an increased score on the BESS test when performed concomitantly.

Hypothesis #3
The null hypothesis is that RTT and BESS test scores will remain the same when performed independently or concomitantly following dehydration.

Significance of the Study

Dehydration causes physiological stress on the body that is demonstrated in different ways. Fatigue is a blanket term used to describe the objective manifestations of these stresses. The purpose of this experiment is to determine if this fatigue is occurring in the central nervous system (CNS) or the peripheral nervous system (PNS).

The CNS consists of the spinal cord and the brain. This is also referred to as the “integrative portion” of the nervous system because this is where sensory feedback is interpreted into motor signals (Guyton, 1976). The PNS consists of
sensory and motor neurons. The sensory neurons deliver sensory information to the CNS, the information is interpreted in the CNS, and a motor neuron is sent to the appropriate muscle through the PNS. This results in muscle contraction and movement. The most vital activity to this process in the PNS occurs at the neuromuscular junction.

There has been evidence that examines how dehydration affects cognition and balance independently. Racinais et al. (2008) administered 3 cognitive tasks showing the effect of hyperthermia on attention and memory, but took no measurements of physical performance. Conversely, Buford et al. (2006) looked at muscular performance in relation to hydration status, but did not examine any aspects of associated cognitive deficit. There is very little evidence to measure divided attention following dehydration. It is important to look at these effects because it more closely mirrors what will be demanded of athletes during a sport activity. McCrea (2001) noted that no single measure is meant to stand alone in determining the magnitude of cognitive deficit (McCrea, 2001). In order to correlate effects of dehydration to a sporting task, attention would need to be divided, as with group 3 of this study. Since diagnostic tests can’t distinguish the cause of results, the effects that dehydration displays on these cognitive measures also has implications on the diagnosis and management of concussion, particularly in athletics.
Figure 1  RTT v. BESS Test

Central Nervous System (CNS)  v.  Peripheral Nervous System (PNS)
  (brain/spinal cord)  v.  (neuromuscular junction/muscle cell)

  ↓

Cognition  v.  Balance

  ↓

RTT  v.  BESS

Definition of Terms

The following definitions are given for the purpose of clarification:

Dehydration: an involuntary loss of body water; a 2% body weight loss

Euhydration: normal body-water content; urine specific gravity < 1.020

Hypohydration: a self-imposed body-water deficit; urine specific gravity ≥ 1.020

Urine Specific Gravity (USG): the ratio of density of a material to the density of water at a given temperature (a measure of the water content of urine); measured using a handheld refractometer (model A300CL; Spartan, Tokyo, Japan); a dimensionless unit
RTT: Reaction Time Task; response time to an auditory stimulus and measured by a voice activated reaction timer (the 63035A Visual Choice Reaction Time Apparatus made by Lafayette Instruments and 63040A Voice Activated Relay made by Lafayette Instruments, respectively), see Appendix II.

BESS: Balance Error Scoring System; represents the number of errors in balance made during a 20 second interval.
CHAPTER 2

REVIEW OF RELATED LITERATURE

Physiological Effects of Dehydration

Reportedly, dehydration impacts cardiovascular and thermoregulatory function within the first 30 minutes of exercise at a body water loss equal to 1% of body mass (Kolasa, 2009). The greater the percentage of body weight lost due to water loss, the more pronounced physiological deficits become.

The main physiological risk associated with dehydration is hyperthermia. Sweating is the body’s main way of regulating its temperature during exercise. With dehydration, there is not enough water to form sweat and the body overheats. Also, water is unavailable for buffering metabolic waste products, potentially resulting in life-threatening conditions, such as rhabdomyolysis and renal failure. Without sufficient water in the body, plasma volume of blood is decreased; therefore, stroke volume is decreased, which decreases cardiac output. Without a sufficient amount of blood leaving the heart, oxygen delivery to the tissue is compromised, which reduces tissue function. Compromised tissue function would be demonstrated by reduced strength or endurance performance of muscle.

CARDIOVASCULAR

Sufficient blood flow to an area of the body is vital to the functioning of the tissues. The amount of blood delivered to a tissue is dependent on the cardiac output. González-Alonso et al. (1997) calculated cardiac output using the indirect Fick equation [cardiac output = CO$_2$ output ($\dot{V}$CO$_2$) / mixed venous CO$_2$ content]
(C\(\overline{V}\)\(\text{CO}_2\)) – arterial \(\text{CO}_2\) content (\(\text{CaCO}_2\)) (González-Alonso, 1997). A simplified equation for quantifying cardiac output is the product of stroke volume and heart rate (\(\text{CO} = \text{SV} \times \text{HR}\)). In their experiment, González-Alonso et al. (1997) examined the effect of dehydration, hyperthermia, and both together on cardiovascular function. They found that dehydration lowered stroke volume 7±2\% (11±3 ml/beat) and increased heart rate 5±1\% (7±2 beats/min) (González-Alonso, 1997). These changes were due to decreased plasma volume of the blood. The increase in heart rate was a compensatory action. Since there was a significant decrease in stroke volume, an increase in heart rate was required to maintain a physiologically appropriate cardiac output.

NEUROLOGICAL (CNS/PNS)

The cerebral cortex is especially important in locomotion and postural control. While the spinal cord alone has been shown to elicit locomotion patterns in the subthalamic cat, the cerebral cortex is needed to make the movement purposeful (Guyton, 1976). A purposeful movement is the manifestation of using the correct movement patterns in the correct sequence.

Proper functioning of the brain, therefore movement, is dependent on the functioning of the blood-cerebrospinal fluid and blood-brain barriers (Guyton, 1976). These barriers ensure vital substrates enter the brain and toxic ones do not. The substrate gradients at these areas of the body are sensitive to fluid balance. Interstitial fluid in the brain tissue is about 12\% of the tissue weight, while fluid is 17\% to the weight of tissues in the rest of the body (Guyton, 1976). Even the slightest change in this fluid balance can affect the excitability of the
neurons. If the neurons do not reach membrane potential, they are unable to fire and trigger movement in the muscles.

The neuromuscular junction is where the motor neuron and muscle fiber meet and form the motor end-plate (Guyton, 1976). The terminals of the nerve attach to the muscle fiber but stay outside the plasma membrane, creating a synaptic cleft. Acetylcholine is an excitatory neurotransmitter (Guyton, 1976). When the concentration of acetylcholine reaches an appropriate concentration, muscle contraction is triggered. Acetylcholine release is dependent on the presence of calcium. Calcium must move across a concentration gradient between extracellular and intracellular fluids. In a dehydrated state, water moves via osmosis extracellularly (Guyton, 1976). The water imbalance between the inside and outside of the cell can influence the calcium gradient, slowing muscle contraction.

SKELETAL MUSCLE

Dehydration can have several negative effects on the athlete. Buford et al. (2006) found that dehydration was detrimental to muscle performance (Buford, 2006). For this experiment, the authors measured peak torque at the knee midseason and postseason in collegiate wrestlers. They found that postseason, when the subjects were not trying to meet a weight requirement, there was a 28% increase in peak torque they were able to generate at the knee (Buford, 2006). While this increase did not correlate to hydration status specifically, there was a 19% increase in peak torque to body weight ratio (Buford, 2006). However, there were no specifics given as to a control for the
factors affecting hydration. Therefore, body weight can be a dependable measure of hydration status.

Upper extremity skeletal muscle function is important to sporting endeavors. Even in running, muscles of the upper extremity are needed to attenuate ground reaction force and control motion. Kraemer et al. (2001) studied, among other variables, upper extremity isometric force production and torque in collegiate wrestlers over the course of 5 matches. These competitions followed a 2 day span during which subjects lost 6% body weight via food and fluid restriction (Kraemer, 2001). Results showed a significant decrease in isometric force production of the upper torso and arms between baseline measurements and after the first match. Unique to this study and specifically important to functional athletic movement, there was “an attenuation of grip strength from baseline values throughout the tournament” (Kraemer, 2001). Not only did dehydration due to rapid weight loss affect muscle performance, those affects were magnified in relation to the duration of the athletic activity.

Dehydration in Athletics

While dehydration is a health concern in all athletics under a variety of thermally challenging environments, it is particularly prevalent in sports with a weight requirement, like boxing and wrestling. Between November and December of 1997, 3 collegiate wrestlers died from hyperthermia and dehydration related causes ("Hyperthermia and Dehydration-Related Deaths Associated with Intentional Rapid Weight Loss in Three Collegiate Wrestlers - North Carolina, Wisconsin, and Michigan, November-December 1997," 1998).
The concept of rapid weight loss through dehydration to qualify for a weight class in a given sport is still in practice today despite regulations by governing bodies such as the National Collegiate Athletic Association (NCAA) to quell this practice.

Research indicates that many athletes are dehydrated preceding physical activity. Volpe et al. (2009) studied the pre-practice hydration status of NCAA Division I athletes using USG as a measure of hydration; these values were correlated with self-reported fluid intake (Volpe, 2009). This study found that in a subject pool consisting of both male and female collegiate athletes, 66% were hypohydrated; among males, 47% were hypohydrated, among females, hypohydration occurred in 28%. Therefore, these collegiate athletes are predisposed to the complications of dehydration.

Dehydration may affect the results of a concussion assessment as well. Concussions commonly occur on an athletic field, particularly in contact sports. Often times, an assessment takes place following a prolonged bout of exercise (i.e. in the middle or following the sporting activity); therefore the athlete is most likely dehydrated to some degree. This physiological state may interfere with results of concussion testing, leading to false positive or increased severity of symptoms, prolonging the time of missed participation.

Cognition and Dehydration

Cognition is a term used to describe mental processes. Ability to complete mental processes, like memory and reaction time tasks, are used as measures of cognition. Patel et al. (2007) and Choma et al. (1998) reported impaired cognition following dehydration in college-aged males (Choma, 1998; Patel, 2007). Patel et
al. (2007) dehydrated 24 healthy, college-aged males who were recreationally active through water restriction and exertion. Subjects endured the exercise condition only for the dehydration trial. Dehydration caused a statistically significant decrease in visual memory scores ($t_{23} = 2.130$) (Patel, 2007). Choma et al. followed collegiate wrestlers during bouts of rapid weight loss. Rapid weight loss “involves self-induced dehydration and starvation until target weight is met” (Choma, 1998). For this study, a minimum 5% body weight loss was the inclusion criteria for the rapid weight loss group. Short-term memory deficit was found following this self-induced dehydration in the experimental group. Scores on the story recall and digit span variable decreased from $12.1 \pm 3.3$ to $10.1 \pm 2.4$ and $13.6 \pm 1.1$ to $12.5 \pm 2.1$, respectively. These deficits were statistically significant (Choma, 1998). These, along with the physiological factors discussed above, lead to decreased performance and, potentially, increased risk of injury or death.

The subject pool in an experiment by Racinais et al (2008) consisted of both males and females at a slightly older age ($31 \pm 1$ year). The method of dehydration was a combination of environmental and exertional by having subjects exercise on a treadmill in a hot room. The greatest effect of the environmental condition was on memory tests, with a 6% decrease in the number of correct answers given in the hot compared with cool condition (Racinais, 2008).

Reaction Time Task (RTT)

To measure the efficiency of cognitive function, a Reaction Time Task (RTT) is administered to each subject. There have been a number of studies
investigating reaction time. These vary in type of stimulus (auditory v. visual) and reaction task (fine v. gross movement). For this study, subjects were instructed to verbally respond to an auditory stimulus, specifically to “immediately say stop when you hear the buzzer”.

Spierer et al. (2010) compared reaction times dependent on, not only type of stimuli but gender as well (Spierer, 2010). Using male and female National Collegiate Athletic Association Division I athletes, the investigators measured reaction time to auditory and visual stimuli independently. Movement time, or sprint speed, was used as a tangible measurement of reaction time. Results showed “that gender differences do exist in response to AS (auditory stimuli) and VS (visual stimuli)” (Spierer, 2010). Following auditory stimuli, this study found there to be a statistically significant difference between male and female subjects (0.1782 ± 0.189 v. 0.1317 ± 0.192 seconds, ρ < 0.05) (Spierer, 2010).

Current research is representing reaction time as a manifestation of neurological information integration (Sperdin, 2009). Secondly, do these neurological processes occur in an “optimal time window” to result in the best possible outcome, assumed to be the fastest reaction time. (Colonius, 2010). Sperdin et al. administered auditory, somatosensory, and auditory-somatosensory multisensory stimuli to subjects, measured reaction times, and compared singular versus multisensory stimuli reaction times in an attempt to determine the efficiency of information integration in relation to the amount of information presented to the neurological system (Sperdin, 2009). Results showed reaction times were significantly faster following multisensory stimuli
versus either of the unisensory stimuli; this was linked to stronger global field power (GFP) waveforms over the 40-84 ms period (Sperdin, 2009). GFP waveforms are a graphical representation of the electrical activity at the scalp using topical electrodes. In this example, GFP waveforms signify neurological activity related to stimuli signal integration and resultant reaction activation.

The time frame in which all of this neurological activity happens is crucial to the resulting reaction time. Colonius et al. (2010) defined this as the spatiotemporal window of integration (Colonius, 2010). The spatiotemporal window has an optimal width; this is the time in which all information can be processed and result in the most advantageous outcome. When this window is “open” is dependent on oscillation frequencies. Colonius et al. (2010) examined several theories on sensory integration, establishing a method of applying them to reaction time. These investigators came to the conclusion that “if one can identify the phase an oscillation is reset by…and its frequency, then one can in principle predict when a temporal window of high excitability (or low excitability) will occur” (Colonius, 2010). Therefore, under this assumption, reaction time is a function of the oscillation phase and frequency of the, in this case, auditory and visual stimuli.

Balance Error Scoring System (BESS)

In conjunction with the RTT, a Balance Error Scoring System (BESS) test measurement is also taken. The subject balances in 3 positions, double-leg stance, single-leg stance, and tandem stance, on a firm surface and unstable surface (Balance Pad, Alcan Airex, Switzerland). (Appendix III) Single-leg stance
is performed on the non-dominant leg. During the tandem stance, the non-dominant is in the rear. The subject has hands on their iliac crests and eyes closed. The investigator counts the number of errors committed in a 20 second time frame. Errors include opening eyes, moving hands from hips, or moving a foot or feet out of the proper position. During a traditional BESS test, the conditions are to be completed in as quiet a space as possible. Since the subjects will need to answer questions while completing the BESS conditions in the divided attention task, this study dictates a modified BESS test be administered where noise is present during the BESS test alone. The error number is totaled for all conditions and used as the final BESS score.

When investigating the learning effect associated with balance, Broglio et al. (2009) found that 3 completions of the BESS test on a single day controls for learning in other sessions on subsequent days (Broglio, 2009). Therefore, the learning trial will consist of 3 completions along with a demonstration of the BESS test. The authors also suggest that during the subsequent testing, 2 trials be performed and the scores be averaged in order to achieve a true representation of the subjects’ balance. However, they note that this is unrealistic in most clinical settings, particularly collegiate athletes. Completing only 1 test during the experimental trials in this study make the results more applicable to what would be seen in the collegiate athletics setting.

Neuromuscular Coordination

Neuromuscular coordination is dictated by “the interaction between the neural and muscle systems” and “is fundamental to all movement”. It is indirectly
measured by forward and vertical jumps, isometric strength, or balance (Bonacci, 2009; Yosmaoglu, 2011). After completing a literature review, Bonacci et al. (2009) noted that, since neuromuscular coordination is essential to all movement, it translates to cardiorespiratory capacity (Bonacci, 2009). Since dehydration can impact the cardiac system by decreasing stroke volume and cardiac output, a connection can theoretically be made between deficits in neuromuscular coordination and dehydration.

Yosmaoglu et al. (2011) measured neuromuscular coordination in postoperative knees of 20 subjects. Balance is an object measure of joint stiffness. The authors of this study noted that via receptors in the ligaments of joints, muscles respond by stiffening to stabilize the joint. If there is something inhibiting muscle contraction, either concentric or eccentric, the joint is unable to stabilize. These authors found that eccentric coordination deficit was 36.73 ± 32.62 and concentric coordination deficit was 20.79 ± 16.66 at 6 months postoperative. After a 40 weeks training program that included balance and cardiovascular training, these measures improved to 24.40 ± 18.24 and 18.94 ± 21.17 respectively (Yosmaoglu, 2011). As previously discussed, dehydration can alter a muscle’s ability to contract due to decreased oxygen delivery. Decreased muscle contraction can cause unstable joints. In this study, that would be shown by a decrease in BESS score following dehydration.

Huo et al. (2007) correlated probe reaction times of physical therapists to their patient’s accident rates (i.e. falls during therapy sessions). The reaction time task of this experiment mirrored that of the one conducted here, meaning the
subjects were instructed to give a verbal response to an auditory stimulus. Subjects were divided into 2 groups, a fall group and non-fall group. The fall group had experienced at least 1 fall accident with a patient during a therapy session in the past 12 months, while the non-fall group had not. Results showed that the probe reaction time of the fall group was significantly ($p < 0.05$) longer than that of the no-fall group (396.7 ± 93.6 ms vs. 330.7 ± 83.0 ms, respectively) (Huo, 2007). In these scenarios, it is demonstrated how neuromuscular coordination is vital to everyday function and tasks.

**Internal and External Focus of Attention**

On the athletic field, there are a variety of demands on the musculoskeletal and central nervous systems of the athlete. Demands of these tasks divide attentional focus between internal and external sources. Internal focus is attention directed at body movements, while external focus is attention directed at the effects of movement on the environment. These systems are in a competition with each other for attentional resources during an activity. Depending on where these resources are allocated in the central nervous system, the outcome, i.e. body movement, can be affected. McNevin et al. (2003) examined this theory by having subjects balance on a stabilometer while focusing on an external marker at varying distances. Results showed that as the markers, and therefore focus, got farther away, balance improved (McNevin, 2003). In other words, as the focus of attention got more external, the subject performed better on the task. This phenomenon has been explained using the Constrained Action Hypothesis.
Constrained-Action Hypothesis

By having the subjects perform a cognitive task during balance, the Constrained Action Hypothesis is applicable. The Constrained Action Hypothesis states by focusing internally, the motor system is constrained due to interference with automatic control processes. By focusing externally, automatic control processes are promoted. Wulf et al. (2010) demonstrated this concept by measuring reaction time during a balance task. With the reaction time task during the retention trial, root-mean-square error (RMSE) for the internal focus group was 4.2, while RMSE for the external focus groups was 3.3 (Wulf, 2010). This demonstrated that with external focus, there are reduced attentional demands and, therefore, reflexive control mechanisms are utilized.

In a study more applicable to this investigation, Vuillerme et al. (2000) found that the addition of a secondary task directs attention away from postural control, making balance more automatic as shown with a reduction in postural sway. By adding an external focus (the reaction time task), center of foot pressure (COP) range, or postural sway, decreased from 5.56 to 9.81 mm (Vuillerme, 2000).

This phenomenon should be observed by dividing attention in this study. For this study, the RTT directs focus externally and the BESS test presents an internal focus. Given these variables, BESS test scores should improve when dividing attention between the 2 tasks. This study aims to show if dehydration affects this predicted outcome.
Gaps in Literature

While there are a lot of studies that have looked at dehydration, especially in the last ten years, none have looked at the effects of dehydration on performance in a divided attention task. A divided attention task would better mirror the demands placed upon the athlete during sport activity. In theory, this would make the results more applicable to athletics. By gaining an understanding of how dehydration influences fatigue in the CNS and PNS, clinicians will be better able to formulate hydration strategies and tailor training regimens towards these systems.
CHAPTER 3

METHODOLOGY

Subject characteristics

The subjects of this study will consist of 20 young adults aged 18-35 years. Inclusion criteria will consist of individuals who run at least 3 days per week for a minimum of 45 minutes per session. Exclusion criteria include a history of lower extremity injury or concussion within the four weeks preceding data collection. Subject with a history of kidney or liver disease, gout, or an allergy to sulfa will also be disqualified.

In accordance with the American College of Sports Medicine and National Athletic Trainers’ Association guidelines, subjects will be considered dehydrated when they have reached a urine specific gravity (USG) greater than or equal to 1.020 (Racinais, 2008). Along with USG, urine color will be assessed using the chart posted by University of Nevada, Las Vegas sports medicine staff, found in Appendix I. This is commonly the method used in collegiate athletics to determine when a student-athlete has reached a detrimental level of dehydration as it is appropriate for self-monitoring.

Subjects complete three testing conditions, serving as their own control. The cognitive task is completion of an auditory reaction time task (RTT). To assess subject balance, they will be tested using the Balance Error Scoring System (BESS). The BESS test provides an objective score for comparison between and within subjects with regard to balance performance.
### Table 1  Characteristics of the Participants

<table>
<thead>
<tr>
<th></th>
<th>Total ($n = 7$)</th>
<th>Female ($n = 3$)</th>
<th>Male ($n = 4$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>26.6 ± 4.5</td>
<td>27.3 ± 5.0</td>
<td>26 ± 4.7</td>
</tr>
<tr>
<td>Right hand dominant</td>
<td>7 ± 0</td>
<td>3 ± 0</td>
<td>4 ± 0</td>
</tr>
<tr>
<td>Body Weight (pounds)</td>
<td>168.9 ± 15.6</td>
<td>162.7 ± 9.6</td>
<td>173.5 ± 18.8</td>
</tr>
</tbody>
</table>

Values are means ± standard deviation

**Instrumentation**

USG is measured by a handheld refractometer (model A300CL; Spartan, Tokyo, Japan) that has been calibrated with distilled water. Reaction time is measured using the 63035A Visual Choice Reaction Time Apparatus made by Lafayette Instruments and 63040A Voice Activated Relay made by Lafayette Instruments.

**Procedures**

Subjects will be recruited by word of mouth through the Department of Kinesiology and Nutrition Sciences at the University of Nevada Las Vegas. All
potential subjects will be questioned to ensure they meet the inclusion criteria and do not have any conditions that would exclude them from participation using the screening document form. (Appendix IV) If applicable, subjects will sign and be given a copy of the informed consent form. (Appendix V)

The order in which subjects complete the trials, euhydrated versus dehydrated, will be randomized. For the euhydrated group, each subject will be instructed to drink 500mL of water in the evening before and 500 mL of water in the morning of testing. This is to ensure subjects report euhydrated. While the dehydration group is being pharmacologically dehydrated, the euhydrated group will have urine specific gravity (USG) and urine color recorded to ensure hydration status. Subjects will then complete the modified BESS test, the auditory reaction time test, and the divided attention task, in that order. For the auditory reaction time test, the stimuli will be initiated at the same time in each 20 second interval for each subject. Each 20 second interval corresponds with a position of the BESS test, as shown in Table 3. This is to prevent any bias as to reaction time being initiated sooner or later in the test for different subjects. This timing will also stay consistent for the dual attention task, meaning the auditory stimuli will be initiated in conjunction with the BESS test during the positions at the same times equivalent to Table 3.

For the dehydrated trial, the athletic trainer, under the orders of a physician, will administer 40 mg of furosemide with 175 mL of water. A 3 hour break will then be given to subjects to allow for adequate diuresis. During this time, subjects will be monitored to abstain from food or drink intake. After the
break, subjects will provide a urine sample at which time USG and urine color will be recorded to establish hydration status. When subjects are determined to be dehydrated based on these measurements, they will undergo the 3 testing conditions. For every trial, modified BESS tests will be performed on a force platform to obtain objective data on postural sway.

The 40mg tablets of furosemide will be obtained from the participating physician. Subjects will be monitored by the physician and a Certified Athletic Trainer during the 3 hours diuresis to monitor for any potential complications from the drug intervention or dehydration. A Certified Athletic Trainer will also be present during the BESS testing to provide treatment in the event of injury due to fall. Subjects will be provided with food and drink to rehydrate following dehydration. When they are determined by the physician to be properly rehydrated and at no risk to their health, subjects will be excused from the session. At the completion of both the euhydrated and dehydrated trials, which will be conducted on different days, subjects will be excused from the study.

During the modified BESS test, each subject will be videotaped. At a later time, the video tapes will be reviewed by 2 individuals trained in recognition in BESS errors. These individuals will then calculate a BESS score for each subject. They will also be blinded to the purpose of the study and whether subjects are euhydrated or dehydrated.
Table 2  Experimental Methodology

<table>
<thead>
<tr>
<th>Trial</th>
<th>Cognition</th>
<th>Balance</th>
<th>Divided Attention</th>
</tr>
</thead>
<tbody>
<tr>
<td>Euhydrated</td>
<td>RTT</td>
<td>BESS</td>
<td>RTT + BESS</td>
</tr>
<tr>
<td>Dehydrated</td>
<td>RTT</td>
<td>BESS</td>
<td>RTT + BESS</td>
</tr>
</tbody>
</table>

Table 3  Auditory Stimuli Initiation Timetable

<table>
<thead>
<tr>
<th>BESS Stance</th>
<th>Stimuli Initiated (seconds)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Solid Single Leg</td>
<td>15</td>
</tr>
<tr>
<td>Solid Double Leg</td>
<td>14</td>
</tr>
<tr>
<td>Solid Tandem</td>
<td>5</td>
</tr>
<tr>
<td>Foam Single Leg</td>
<td>8</td>
</tr>
<tr>
<td>Foam Double Leg</td>
<td>11</td>
</tr>
<tr>
<td>Foam Tandem</td>
<td>4</td>
</tr>
</tbody>
</table>

Data Analysis

Mixed model analyses of variance (ANOVAs) will be conducted to analyze results with instrumentation (RTT v. divided attention; BESS v. divided attention) as a between-subjects factor and hydration status (euhydrated v. dehydrated) as a within-subjects factor. Statistical significance of p<.05 is set for ANOVAs.
Paired $t$ tests will be used to analyze differences in instrumentation scores within groups euhydrated v. dehydrated if there is significant interaction. To minimize error rates associated with multiple comparisons, the .01 level of probability will be used for post hoc comparisons.

Limitations

A possible limitation is if the task is completed and measurements are taken at a different time of day. This factor can affect motivation and mood. Precautions should be taken to control these factors. Data collection should be scheduled at a time when the subject is available consistently on each day. Since subjects will be placed in groups, it may be difficult to find a time when all subjects in a certain trial group will be able to meet together. However, it is still possible that subject availability for data collection may change.

Another potential limitation is the subjects' ability to become dehydrated. Even though the investigators are using a pharmacological method of achieving dehydration, physiology is different in each individual. That is to say, physiological responses are independent of any controllable factors in this study. As all subjects cannot be tested at once, this gives certain subjects more time if it is needed in order to dehydrate. This should control for this complication and not elongate the data collection process.

Delimitations

The first delimitation of this study would be subject recruitment and retention. Since we are unable to use a readily available subject pool, like student-athletes, efforts will need to be made to reach out to willing participants
who meet the inclusion criteria and have schedule availability. Also, subjects specifically need to be avid runners. For the design of this study, it is important that subjects participate in the same sporting activity to prevent any disparity in lower extremity muscle activation during the balance task. Chapman et al. studied EMG activity in the lower extremity of triathletes during the bike, run, and transition segments of the race. In the tibialis anterior muscle, they found a decrease of ~30% in EMG activity during the stance phase of running when compared to cycling (Chapman, 2008). Potentially, this muscle activation difference may influence subject’s ability to balance. It is better to prevent any discrepancy by controlling sport activity type.

The potential for lower extremity injury or concussion between trials is a delimitation of the study. Since the subjects will not be instructed to halt exercise or sport participation, the possibility exists for them to experience one of these medical issues between trials. In this case, the subject would need to be excused from the study.
CHAPTER 4

RESULTS

For several reasons, adequate data was unable to be collected to report statistically significant results. Far from the original plan for this study, only 7 subjects were able to participate. Of those 7, only 4 were able to complete both conditions, euhydrated and dehydrated. The first complication that arose was a large turnover in faculty at the university, specifically those involved in the athletic training program. This led to a shuffle in committee members. It took time to then recruit new interested committee members and bring them up to date on concepts and methodology of the study. Next, Institutional Review Board (IRB) approval needed to be secured. There are many facets to, not only this study, but any involving dehydration. The potential risks at first seem staggering. However, it is important to realize the level of dehydration required by most studies, including this one, to fit the medical definition carry minimal risk. Especially when pharmacologically intervening with subjects, dehydration studies require expensive review before being approved.

Once IRB approval was given, subject recruitment began. Some subjects that qualified for the study were hesitant or refused to participate after receiving all the information due to potential interruptions with their training regimen. There were a few problems coordinating subject availability and the graduate intern responsibilities of the investigator due to the time requirement of the dehydrated condition. Also, this data needed to be collected at the same time of day, early morning, to ensure the IRB required medical personnel were on campus and
easily accessible, if needed. Once subjects were secured, there were 3 subjects who withdrew from the study due to injury outside their participation as subjects. After starting data collection, the reaction timer short circuited, most likely due to static electricity in the laboratory from equipment associated with another study, and needed to be returned to the manufacturer for repairs. By the time the reaction timer was returned, IRB approval had expired. Due to the need for a full board review, renewal of approval was not plausible in the available time frame. Therefore, the data that was already collected is presented here as a pilot study, setting the foundation for future research in the area of dehydration and its association with divided attention.

Results of the data the investigators were able to collect is presented below for reference.

Table 4  Euhydrated Results

<table>
<thead>
<tr>
<th></th>
<th>Total (n = 6)</th>
<th>Female (n = 3)</th>
<th>Male (n = 3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>UC</td>
<td>2.7 ± 0.5</td>
<td>2.7 ± 0.6</td>
<td>2.7 ± 0.6</td>
</tr>
<tr>
<td>USG</td>
<td>0.523 ± 0.53</td>
<td>0.342 ± 0.58</td>
<td>0.705 ± 0.52</td>
</tr>
<tr>
<td>BESS (errors)</td>
<td>14.3 ± 5.3</td>
<td>15.3 ± 2.9</td>
<td>13.3 ± 7.6</td>
</tr>
<tr>
<td>RT (seconds)</td>
<td>2.74 ± 0.4</td>
<td>2.77 ± 0.2</td>
<td>2.72 ± 0.4</td>
</tr>
</tbody>
</table>

Values are means ± standard deviation
Table 5  Dehydrated Results

<table>
<thead>
<tr>
<th></th>
<th>Total (n = 5)</th>
<th>Female (n = 3)</th>
<th>Male (n = 2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>UC</td>
<td>6 ± 1</td>
<td>5.7 ± 0.6</td>
<td>6.5 ± 0.7</td>
</tr>
<tr>
<td>USG</td>
<td>1.024 ± 0</td>
<td>1.022 ± 0</td>
<td>1.028 ± 0</td>
</tr>
<tr>
<td>BESS (errors)</td>
<td>12 ± 2.6</td>
<td>13.3 ± 2.5</td>
<td>10 ± 1.4</td>
</tr>
<tr>
<td>RT (seconds)</td>
<td>2.72 ± 0.3</td>
<td>2.83 ± 0.3</td>
<td>2.56 ± 0.3</td>
</tr>
</tbody>
</table>

Values are means ± standard deviation
CHAPTER 5
SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

Discussion of Results

The most significant realization that came out of this study was the complexity involved with performing a dehydration study, particularly one with this many factors. It is important to be cognizant of the concerns of other investigators involved with the study and reservations of your subjects. Personal limits of the investigators, especially the lead investigator, need to be factored into planning conduction of a study like this. In this case, the demands of the graduate internship within the university athletic department contributed to the difficulty in collecting enough data to make results statistically significant. Hopefully, this study will help in the preparation of future studies, particularly by graduate students.

Recommendations For Further Study

GENDER DIFFERENCES

Gender differences in performance following dehydration have been measured, but never in conjunction with divided attention. This pilot study demonstrated a possible difference between males and females in both independent and concomitant measures. There are several directions from which to approach gender differences in reaction time and balance. What we had initially hoped to do with this study was to compare the effect of dehydration on males and females. In other words, does dehydration affect males or females
more? For females, it may also be important to include where in the menstrual cycle female subjects are. Menstrual cycle can affect USG values due to changes in water content of urine. Determining if males or females are more susceptible to deficits following dehydration has widespread implications for athletics. One such example applies to the separation between male and female athletes on the playing field. Determining if it is safe for men and women to compete against each other is rooted in this concept. If men are better able to respond to stimuli, either euhydrated or dehydrated, and this divergence is only magnified when dehydrated, injury may become a serious concern.

SITUATIONAL EFFECTS

Another interesting component of reaction time and dehydration has to do with situational effects, or “court sense” (Spierer, 2010). According to Spierer et al. (2010), court sense is an athlete’s ability to integrate large amounts of information from a wide variety of sources (Spierer, 2010). Court sense can be attributed to training methods or the scenario in which the stimuli is originated. It would be warranted to explore which scenarios result in the best reaction times. Investigation would include a comparison of sports in terms of their athletes’ performances on the experimental tasks. It may be possible to determine if certain sports favor better performance. If a connection can be made between athletes and their reaction time and balance dependent on their sport participation, results have the potential to influence training regimen for other sports. Also, are these results related to dehydration rates within the subjects’ sport participation. Certain sports are prone, or have a higher incident of,
dehydration related illness. Are these athletes’ reaction times and balance less affected by dehydration, especially when the tasks are performed concomitantly.

Conclusions

While this study initially had promise, extenuating circumstances prevented investigators from fully exploring the effects of dehydration on reaction time and balance. However, a foundation is established for future research to be done in this area.
# APPENDIX I

## AM I HYDRATED?

### Urine Color Chart

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>If your urine matches the colors 1, 2, or 3, you are properly hydrated.</td>
</tr>
<tr>
<td>3</td>
<td>Continue to consume fluids at the recommended amounts.</td>
</tr>
<tr>
<td>4</td>
<td>If your urine color is below the <strong>RED</strong> line, you are <strong>DEHYDRATED</strong> and at risk for cramping and/or a heat illness!!</td>
</tr>
<tr>
<td>5</td>
<td><strong>YOU NEED TO DRINK MORE WATER!</strong></td>
</tr>
<tr>
<td>6</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td></td>
</tr>
</tbody>
</table>
APPENDIX II

63035A Visual Choice Reaction Time
Apparatus made by Lafayette Instruments

63040A Voice Activated Relay made by
Lafayette Instruments
Screening Document
IRB Protocol #1110-3954
Effects of Dehydration on Concomitant Measures of Cognitive Function and Balance

1. Have you suffered a lower extremity injury within the last 4 weeks? (examples include ankle sprains, ACL/MCL/PCL sprains, quad or hamstring strains or “pulls”, knee, foot, or ankle surgeries, or fractures)

2. Have you suffered a concussion within the last 4 weeks?

3. Do you currently have or have you ever been diagnosed with kidney disease?

4. Do you currently have or have you ever been diagnosed with liver disease?

5. Do you currently have or have you ever been diagnosed with gout?

6. Are you currently pregnant?
   a. If no, do you think you may be pregnant?

7. Do you have an allergy to sulfa drugs?

8. Do you have an allergy to antibiotics?

9. Have you ever suffered a heat illness?

10. Have you ever been unconscious due to dehydration?

11. Are you currently taking a protein supplement?
APPENDIX V

Department of Kinesiology and Nutrition Sciences

INFORMED CONSENT FORM

TITLE OF STUDY: Effects of Dehydration on Concomitant Measures of Cognitive Function and Balance

INVESTIGATOR(S): Dr. Tony Santo & Miss Stephanie Watson

CONTACT INFORMATION: If you have any questions or concerns about the study, please contact Dr. Santo at (702) 895-5329 or Miss Watson at (646) 456-9075.

For questions regarding the rights of research subjects, any complaints or comments regarding the manner in which the study is being conducted you may contact the UNLV Office of Research Integrity – Human Subjects Research at (702) 895-2794 or toll free at 877-895-2794 or via email at IRB@unlv.edu.

Purpose of the Study
The purpose of this study is to determine how dehydration affects fatigue in the brain and skeletal muscle.

Participants
You are being asked participate in this study because you are an apparently healthy runner between 18-35 years old. You run at least 3 days per week for a minimum of 45 minutes per session.

Procedures
If you volunteer to participate in this study, you will be asked to visit the UNLV Motor Learning laboratory, building BHS room #222 two times during the study for a total of approximately 5 hours. The longest session will last approximately 3 hours and 30 minutes. You will be asked to complete 2 trials for the experiment. For each trial, you will be asked to provide a urine sample from which the water content and urine color will be measured and recorded. One trial will be a dehydrated trial, the other will be with proper hydration (euhydrated). For the dehydrated trial, you will be given furosemide with 175 mL of water by a medical professional which will cause you to urinate. There will then be a 3 hours wait period during which you will be monitored and unable to eat or drink. For the proper hydration (euhydrated) trial, you will be asked to drink 500 mL of water in
the evening before and 500 mL of water the morning of testing. You will complete the Balance Error Scoring System (BESS) test, a Reaction Time test, and both at the same time while you are properly hydrated (euhydrated) and dehydrated. During completion of the BESS and combined tests, you will be videotaped for reference. This will conclude the session.

**Balance Error Scoring System (BESS) Test:** The BESS test consist of balancing in 3 standing positions, double-legged stance, single-leg stance, and standing 1 foot in front of the other. All 3 positions will be done on a force platform. First, balance will be done on just the hard platform. Then, the balancing positions will be completed on a foam pad on top of the force platform. You will be asked to hold the positions with your hands on your hips and eyes closed for 20 seconds. During this test you will be videotaped. At a later time, a researcher will watch your performance and count the number or errors committed. Errors include removing your hands from your hips, opening your eyes, or moving out of the proper position, even if it is to catch yourself from falling. You will be trained in these postions and errors during the training trial mentioned above. The total number of errors committed will comprise the BESS score.

**Reaction Time Test:** This test involves listening for a buzzer at random intervals. When you hear the buzzer, you will say “Stop”. The Reaction Timer will measure the time between the buzzer sound and your voice. The measured time will be deemed your reaction time.

**Combined BESS and Reaction Time test:** For this trial, you will be asked to complete the reaction time test while performing the balance stances of the BESS test at the same time. Again, you will be videotaped during this test. The videotape will be viewed by a researcher at a later time. This researcher will count the number of errors making up the BESS score.

All information will remain confidential.

**Benefits of Participation**
There may be little direct benefits to you as a participant in this study. The results of this study may provide insight into the importance of proper hydration (euhydration) and impact hydration strategies in athletics.

**Risks of Participation**
This study involves minimal risk to you. There is a risk of injury from being unable to catch yourself if you lose your balance during the BESS test. However, as an avid runner, the chance of this happening is small. Dehydration carries the risk of certain physiological symptoms. However, the level of dehydration you will be asked to achieve for this study carries minimal risk of any serious complications involving the heart or musculature. The symptoms you are most likely to experience are cramping, lightheadedness, headache, or discomfort due to thirst. According to the National Institutes of Health, side effects of the medication, furosemide, that differ from those due to dehydration are blurred vision, constipation, or restlessness. You will be monitored by
a medical professional throughout the dehydration process to ensure that if you do experience severe symptoms, you can receive immediate medical care. Please indicate below whether you will permit the present medical professional to provide care should you require it. UNLV makes no commitment to provide additional medical care resulting from participation in this research.

**Preparticipation Screening**
Before participation in this study, you will be asked questions regarding your medical history. The goal of this questionnaire is to determine if you are at increased risk to your health by participating in this study. It is important that you be as honest as possible when answering each question. Please understand that if you are pregnant or think you may be pregnant, you will be excused from participation in this study. Since furosemide is an NCAA banned substance, current student-athletes will be excused from this study to avoid a positive drug test and subsequent NCAA ban from participation.

**Cost /Compensation**
There will not be any financial cost to you to participate in this study. The study will take approximately 5 hours of your time total divided across 3 sessions; however there is no compensation for your time. You will be provided with food and drink following the completion of each trial.

**Contact Information**
If you have any questions or concerns about the study, you may contact Dr. Santo at Antonio.santo@unlv.edu or Miss Watson at watson81@unlv.nevada.edu. For questions regarding the rights of research subjects, any complaints or comments regarding the manner in which the study is being conducted you may contact the UNLV Office of Research Integrity – Human Subjects at 702-895-2794 or toll free at 877-895-2794 or via email at IRB@unlv.edu.

**Voluntary Participation**
Your participation in this study is completely voluntary. You may refuse to participate in this study or in any part of this study and you may withdraw at any time without prejudice to your relations with the University. You are encouraged to ask questions about this study prior to the beginning or at any time during the study. You will be given a copy of this form.

**Confidentiality**
All information gathered in this study will be kept completely confidential. Only those persons who are directly related to this study (i.e. researchers, data analysts, thesis advisory committee) will have access to your data. No reference will be made in written or oral materials, which could link you to this study. All records will be stored in the laboratory for a period of 3 years. After 3 years, any documentation with identifiable information (e.g., name) will be destroyed. Unidentifiable data will be stored in locked storage indefinitely.
**Freedom of Consent:**
I have read the above information carefully and I am aware of the tests/procedures to be performed. Knowing these risks and having the opportunity to ask questions, I agree (consent) to participate in this study. I understand that I have a right to withdraw from this study at any time without prejudice. I am at least 18 years old and a copy of the informed consent has been given to me.

Please indicate below whether or not you will allow the present medical professional to provide care if you should require it.

I agree to allow the present medical professional to provide medical care ☑

I do not agree to allow the present medical professional to provide medical care ☐

______________________________    __________________________
Signature of Participant                      Date

______________________________
Participant Name (Please Print)

______________________________    __________________________
Signature of Witness                      Date

**Participant Note: Please do not sign this document if the Approval Stamp is missing or is expired.**
REFERENCES


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Thesis Examination Committee:
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Committee Member, Dr. Gabriele Wulf, Ph. D.
Committee Member, Dr. Richard Tandy, Ph. D.
Graduate College Representative, Dr. Daniel Young, DPT