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Sweat rate and Sweat Electrolyte Concentrations in Football Players During Exercise in Cool versus Warm Environmental Conditions: A Cross Over Study

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Sweat rate and Sweat Electrolyte Concentrations in Football Players During Exercise in Cool versus Warm Environmental Conditions: A Cross Over Study

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Abstract

Objectives: To determine if sweat rate and sweat electrolyte concentrations differ when football players exercise in warm versus cool environmental conditions. **Participants:** Eleven unacclimatized football players exercised for 60 min in warm (24°C and 75%RH) and cool (10°C and 25%RH) randomly assigned trials. **Methods:** Sweat patches made from sterile gauze were placed on forearm, low back and thigh sites after skin was thoroughly washed and dried. Sweat rate was calculated as change in pre-exercise to post-exercise nude body weight adjusted for fluid consumed and urine produced during exercise. Sweat patches were removed after exercise and centrifuged to separate sweat from gauze and analyzed for Na^+, K^+ and Cl^- via ion selective electrode. Data was analyzed using two-way ANOVA and dependent t-tests with significance set at $p<0.05$. **Results:** Sweat rate was higher in warm (1.4±0.6 L/Hr) versus cool (0.9±0.6 L/Hr) conditions, but no differences existed in Na^+, K^+ or Cl^- in any of the three sites. Because sweat rate was higher in warm, calculated full body NaCl losses (6843.9 ± 3784.5/Hr) were also higher versus cool (4398.8 ± 3939.5/Hr) conditions, t =2.77, P=0.019). **Conclusion:** As expected, sweat rate was higher in warm versus cool conditions, but there were no differences found in sweat electrolyte concentrations at any site measured. Thus, electrolyte replacement does not need to be changed for each player in different environments other than accounting for sweat losses. However, individual replacement plans should be specialized due to considerable individual variability in both sweat rate and sweat electrolyte concentrations.

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Introduction

Athletes at every level of play generally participate in their sport at different times of the year even though they have one competitive season. American football is no exception as players can practice and play in two very different environments, a cool spring and a hot preseason. Training camps for football begin in the warm or hot summer months of July and August which challenges the thermoregulatory system to keep the body in homeostasis. Fortunately, at the collegiate and professional levels of play there is a mandated acclimatization period in the summer, and many states follow a schedule that also requires a period of time to allow for athletes to become acclimatized to the heat. The evaporation of sweat is a very important avenue for heat dissipation in individuals who exercise in the warm or hot climates,[1] but sweating also occurs especially in large athletes even when environmental conditions are thermoneutral or even cool.[1] Sweat rate, sweat electrolyte concentration, particularly sodium and chloride, and therefore salt losses, can be important to measure due to the link to illnesses such as hyponatremia.[2] Every individual's sweat rate and sweat electrolyte concentration is different but how much they change in football players when they practice in warm versus cool conditions is not known. This is an important consideration, not only for sweat electrolyte replacement needs during times when players exercise in the heat, but for monitoring factors such as over drinking when the same players practice in cool conditions which might predispose them to exercise associated hyponatremia.

Thermoregulation

Humans are homeotherms meaning they maintain a relatively constant internal body temperature regardless of the external environment. Thermoregulation is the process by which humans maintain thermal homeostasis ensuring proper body function in all climatic conditions both at rest and during physical activity.[1] There are differences in how the body reacts when encountering a cool environment versus a hot environment, but regardless of the ambient conditions, thermal stimuli received by the body will trigger a variety of responses in order to maintain a normal body temperature.[1] This "normal" body temperature at rest is approximately 37°C but during physical activity, especially high intensity exercise in warm or hot conditions "normal" exercise induced hyperthermia consistently reaches 39 - 40°C.[1,2] Thermoreceptors in the skin recognize changes in temperature and transmit the information to the hypothalamus.[1] The hypothalamus is often referred to as the body's thermostat because it is responsible for initiating and adjusting responses such as stimulating sweat glands and increasing blood flow to the skin to facilitate heat dissipation when body temperature rises.[1]

This response is particularly critical in athletes who, when participating in sport activities, are constantly producing heat as a byproduct of muscle contraction. The harder an athlete works, the higher the metabolic heat production resulting in exercise-induced hyperthermia if this heat is not adequately dissipated.[1,2] This balance can become challenging when athletes exercise in thermally stressful situations where heat can actually be gained from the environment depending on factors such as ambient temperature and radiant energy from the sun or playing surface. The core body temperature directly reflects the balance between heat production and dissipation, and without a proper balance this can lead to dangerous levels of hyperthermia. Body heat is dissipated from the inside out, working from the core to the shell (ie skin) to the environment.

This process is challenged to a greater extent when there is a barrier between the skin and the air, such as athletes wearing protective equipment or individuals who must perform physical activity while dressed in protective clothing (ie soldiers, firefighters, or hazmat personnel).[2]

To remain in body temperature homeostasis, or thermal balance, different physiological events will occur as a response to hot or cold climatic conditions. There will also be substantial differences in these physiological responses depending on the individual’s activity level. Vasodilation occurs in response to a hot environment to shunt additional blood to the skin for heat removal.[1,2] This is facilitated by arterial venous anastomoses which is part of the special circulation inherent to the integumentary system. Concurrently, the hypothalamus will stimulate the eccrine sweat glands to increase sweat production.[1,2] Evaporation is the main way that heat is removed from the body during exercise, accounting for approximately 80% of heat dissipation.[1,2] Conversely in the cold, vasoconstriction[1] occurs to reduce blood flow to skin attenuating heat loss.[1] Both shivering and non-shivering thermogenesis are also mechanisms by which homeotherms product heat for protection against a dangerous reduction in body temperature.[1] In addition, “warm blooded animals” will draw their extremities into their body (ie. curl up into a ball) to reduce surface area thereby mitigating heat loss.[1]

Dissipation of body heat is most effective when a large temperature gradient is maintained between the core and the shell (skin and underlying tissue), and the shell and ambient environment. Therefore, methods of cooling the “shell” are critical for preserving thermal homeostasis. There are three dry avenues of heat transfer that can occur between the body and the environment (conduction, convection, and radiation), and one “wet” avenue (evaporation) that can transfer heat from the skin to the air.[2,3] Conduction is when heat is lost through contact with another object of a lower temperature. Convection is heat transfer by motion as is demonstrated by air molecules flowing over the skin. In many athletes, convection is enhanced due body movement from running, or to a greater extent cycling, which has been shown to be an important factor in thermoregulation during exercise.[2,3] Radiation is the transfer of heat by electromagnetic waves such as energy absorbed by the body from direct sunlight. Conduction, convection and radiation can cause heat to be transferred to, or gained by, the environment depending on the ambient conditions. Importantly, the transfer of heat to the environment via these avenues can be easily impeded by clothing and/or equipment worn.[2,3,4] Evaporation is when a liquid is turned to a gas which results in cooling of the surface from which the liquid was vaporized. Approximately 80% of the heat produced during physical activity is dissipated when sweat is evaporated from the skin.

There are many factors that influence core temperature due to exercise induced heat production and the ability of athletes to dissipate that heat. Examples of non-modifiable factors include environmental conditions such as ambient temperature and humidity, cloud cover and playing surfaces. There are also important "modifiable" factors that our athletes and coaches can control such as time of day for practice, state of hydration (the amount of total body water), clothing or equipment worn,[3,4], frequency of rest breaks, acclimatization, and intensity of exercise.[2,3] Dietary requirements are not discussed as often but can also be important, especially in athletes who lose excessive electrolytes due to high sweat losses, high sweat electrolyte concentrations or both. Consequently, evaluating sweat electrolyte losses through sweat testing has become more common in the world of sports.

Acclimatization

Exercise that occurs in the heat is physiologically stressful, especially when the intensity is high as is typical in sports. To minimize the potential for heat illness and optimize exercise performance, athletes must go through a process of heat acclimatization. This involves physiological adaptations that allow the body to become accustomed to performing physical activity in a hot environment.[5-8] Sports that have their competitive season in the Fall commonly start their preseason practices in July or August, which are typically the hottest time of the year in many parts of the country. This means that athletes need an appropriate time period to acclimatize to decrease the risk of heat related illnesses as well as help performance.[5,6,7] If athletes are not fully heat acclimatized their physical ability to exercise intensely in the heat is not optimal and they are at an increased risk for exercise induced hyperthermia potentially leading to exertional heat stroke.[5-9]

The process of acclimatization results in numerous physiological changes[5-9] that help the body better tolerate exercise in the heat. These benefits include an earlier onset and increased sweat rate,[5,6,8] increased plasma volume resulting in improved cardiovascular stability,[5,6] lower core temperatures, and a reduction in sweat and urinary electrolyte losses.[5,6] These adaptations are imperative to an athlete's ability to practice and play in a hot environment which demonstrates the importance of an acclimatization program not only for safety but for maintaining peak athletic performance.[5,6,8]

Currently there are many different organizations that have suggested or even mandated guidelines to follow for heat acclimatization such as the NCAA, NATA, ACSM, and the Health Administration.[5-8] There are also region-based guidelines that follow wet globe bulb thermometer temperature.[5,8,9] According to the wet bulb globe temperature (WBGT) once certain environmental conditions are met, proper precautionary actions are taken to make sure activity is continued safely.[5,8,9] Many of the guidelines follow a time period of one to two weeks for athletes to fully acclimatize.[5-8] The initial adaptations, such as an increase in plasma volume, are shown to occur within the first five days.[5,6] The alterations in the sweat response, such as an earlier onset, a greater surface area of sweat production, increased sweat rate and changes in sweat electrolyte concentration typically take 10 to 14 days.[5-8] All of these adaptations result in a decrease in core body temperature for the same exercise intensity and an increase in exercise tolerance.[5-8] In collegiate football, as per the NCAA heat acclimatization guidelines, there is a progression that is followed which starts by limiting equipment worn and practice time. Both of these alterable factors are then incrementally increased over a two week period of time[1,3-5]. These guidelines have been put into place to make sure athlete's bodies are ready for full contact practices and competitions in the heat.

Many athletes are not properly educated when it comes to dealing with the heat and unfortunately believe that just staying hydrated will prevent problems such as heat exhaustion, exertional heat stroke and muscle cramping.[5-8] Unfortunately, this sometimes results in athletes overdrinking which can cause more harm, such as promoting exercise induced hyponatremia. Sadly, this preventable illness has resulted in fatalities in high school football players and has been reported in collegiate football as well.[10-12]

Hyponatremia and Football

Hyponatremia related to exercise is an illness defined as a clinically low level of sodium in the bloodstream due to excessive fluid intake, [10-12] or extreme sodium losses through sweat,[12] or a combination of these two factors. If not taken care of properly, it can progress enough to be considered a non-traumatic fatality[11,13,14] by causing the brain to swell. This is known as exercise associated hyponatremic encephalopathy (EAHE).[15] Exercise associated hyponatremia, or EAH, is believed to occur entirely due to drinking too much hypotonic fluid before, during and or after exercise.[15] Normal serum sodium is between 135 and 145 mEq/L,[11,12] but symptoms related

EAH typically do not occur until serum sodium levels drop below 130 mEq/L.[10-12] The symptoms of EAH include but are not limited to headache, dizziness, muscular twitching, swelling in extremities, nausea, vomiting, disorientation, muscle cramping pulmonary, and or cerebral edema.[10-12] Studies have found that hyponatremia was prevalent in collegiate football[13,14] and that it is more frequent during preseason than other times of the year.[14] Additionally, it was more prevalent and in the collegiate setting compared to the high school level.[13]

As indicated, hyponatremia can occur by one of two ways, not losing much salt through sweating and drinking too much water, or losing too much salt through sweat and not replacing it.[10,11] This is what distinguishes the two main types of hyponatremia which are EAH[12] and hypovolemic hyponatremia.[15] EAH, as previously defined, can occur when individuals who are not salty sweaters overdrink hypotonic fluids resulting in blood (serum) sodium being diluted.[15] Conversely, hypovolemic hyponatremia comes from sodium depletion which happens when athletes lose excessive amounts of salt due to heavy sweating but are not replacing it. This form of hyponatremia has been reported in football players, particularly when numerous exercise bouts occur in the heat on consecutive days as used to by typical of preseason two-a-days.[15] This leads not only to hyponatremia but also low blood volume (hypovolemia).[15]

Prevention, recognition and treatment are vital when dealing with hyponatremia which means that understanding the signs and symptoms of both types is critical. Due to the fact that excessive fluid intake causes the dilution of blood sodium in EAH, prevention is simple, athletes should only drink when thirsty.[11,12] Early recognition of the signs and symptoms is key. Being able to pick up on early signs and symptoms before they progress, and educating the athlete to stop overdrinking, can not only stop it from getting worse but can prevent subsequent episodes.[12,15] Treatment of EAH is to restore the balance of blood sodium which could be accomplished by providing oral hypertonic fluids such as bouillon until the athletes begins to urinate freely. If symptoms do not subside, then hypertonic saline should be administered intravenously in the hospital.[12] Sodium depletion hyponatremia (ie hypovolemic hyponatremia) should be treated with increased dietary sodium intake or salt supplementation if necessary. Due to the large individual variability in sweat rates and sweat sodium concentration it is recommended that athletes prone to sodium depletion have testing done to assess these sodium needs.[11,12]

Sweat Rate and Sweat Electrolytes in Warm Weather

Warm or ambient temperatures especially when relative humidity is high, causes higher sweat secretion.[16] The considerable variability in sweat rate from one individual to another when exercise occurs in a warm or hot environment is reported in different athletes.[16-19] Sweat rates within an individual also differ when they exercise in a cool versus hot environment.[16-19]

Research related to sweat rate or sweat composition when subjects exercise in warm or hot weather conditions typically report ambient temperatures when data is collected of between 23°- 27° C.[16,21,22] Data from these studies indicate that when athletes exercise in warmer weather there is substantial variation in sweat rate, sweat sodium concentration, and sodium losses.[16,17,23] In these studies, sweat rate was between 0.430 and 3.16 L/Hr,[16,17,21,23] sweat sodium concentration was between 15 and 99 mmol/L,[16,17,21,23] and sodium losses were between 1.4 and 30.2 grams.[16,17,21,23] In multiple studies done by Fowkes Godek in collegiate and NFL players, sweat rate has been shown to be anywhere from 0.43 to 3.16 L/Hr,[24] with certain individuals losing 10 L/Day when participating in two a day practice.[24] When separated by position groups (and therefore body size) there were significant differences in sweat rate between larger linemen

(2.25 ± 0.56 L/hr) and smaller backs and receivers (1.4 ± 0.45 L/hr).[23,25] Fowkes Godek et al also found differences in sweat rate between small cross-country runners (1.77 ± 0.4 L/hr) and larger football players (2.14 ± 0.53 L/hr) while running and practicing in the heat during preseason.[24] The higher sweat rates in certain athletes compared to others were attributed primarily to the vast differences in physical characteristics such as body mass and body surface area. [23,24,25]

Importantly, higher sweat rates reported in warmer environments does indicate players should consume more fluid to minimize body mass loss, but electrolytes should also be taken into consideration.[26] In another study by Fowkes Godek et al, the researchers compared sweat rates and fluid consumption during preseason in Division II football and NFL players and did not find differences in sweat rate between college (1.8L/Hr) and professional (2.1L/Hr) players.[27] While mean percent body weight loss during practices was not different between groups, the Division II football players lost more total sweat during a day of two-a-day practices because the practices were longer.[27] Although the DII players lost more sweat during practice because the practices were longer, their fluid consumption was also higher than the NFL players. Importantly, both groups were able to adequately replace sweat losses during practices even though their access to fluids on the field was dissimilar.[27] This information is very important in trying to properly hydrate an athlete while keeping their electrolyte balance as close to normal as possible.[22,23] In a study by Fowkes Godek et al, comparing NFL linemen versus backs they looked at overall sweat rate and fluid turnover which indicated that each individual is considerably different in sweat rate and fluid consumption.[25] Again, the authors attributed the difference between the groups largely to body size and body surface area.[23,25] These differences were also found in collegiate linemen versus backs.[22]

Researchers such as Fowkes Godek, Maughan, and Shirreffs have completed numerous field studies investigating the concentration of electrolytes, specifically sodium, in sweat collected from athletes when they practice in the warm or hot environments.[23-25] The subjects in these studies were elite level soccer[19,20] and professional football players.[23,24] These researchers use the sweat patch method of collecting sweat samples. In some studies sweat is collected from four sites on the body including the thigh, forearm, chest, and back. It was found, however, that the forearm and back were the most accurate representations of whole body electrolyte concentration.[20,23] In a study conducted in a warm environment the range of sweat sodium ranged from 15 to 99 mmol/L[23] compared to a separate study conducted in a cool environment where sweat sodium ranged from 25 to 90 mmol/L.[20] From this data, you can see that the individual variability ranges anywhere from 15 to 99 mmol/L in both warm and cool environments.[20,23]

All research points to the fact that there is tremendous individual variability in both sweat rates and electrolyte concentrations. Due to vast individual differences in both sweat sodium concentration and sweat rate, these studies report total sodium losses ranging from 642 to 6,700 mg/hr.[17,20,21,23] When practices last for more than two hours the sodium loses can be extensive, and on days where players participate for more than 4 hours, sodium losses can be an astonishing 30.2 grams/day.[23] Due to the considerable quantities of electrolytes lost, especially sodium, replacement of these lost electrolytes either during or more likely after activity is paramount.[23,26] Football players are especially at risk of sodium depletion hyponatremia due to factors such as a large body size, body surface area, and the fact that protective equipment is worn. This illustrates the importance of having a personalized strategy to keep these athletes safe from under or over hydrating.[16,23,25,26]

Sweat Rate and Electrolyte in Cool Weather

In the research area involving sweat produced by subjects during exercise, there are many published studies done on athletes exercising in warm or hot environments due to the risk of heat illnesses, but research related to sweating in cool environments is lacking.[19] Due to the different ambient temperatures and subsequent thermoregulatory responses, warm versus cool environments cause various sweat rates, and potentially even different sweat electrolyte concentrations.[17,19-21] As expected, research indicates that practicing in warmer weather elicits a higher sweat rate compared to cooler conditions.[17,18] Fluid intake by rugby players was also found to be higher in warm weather as the thirst sensation is decreased when athletes exercise in a cool compared to warmer environmental conditions.[18] Due to the lack of research, there was limited information about sweat rate and electrolyte concentration in American football players when they practice or play in cool conditions.[20] However, there is research related to sweat response in cool weather (6 - 12°C) in elite level soccer players.[17-21]

Maughan and Shirreffs have shown that sweat rate, sweat sodium concentration, and sodium loss varies in cool weather with the rate between 0.4 - 1.0 L/Hr[17,20,21] sweat sodium concentration between 34 - 76 mmol/L,[17,19-21] and sodium losses between 1.5 - 4 grams per 90 minutes of practice.[18,20,21] Once again, differences in individual sweat rate were attributed to many different factors including exercise intensity, body size, and amount of clothing worn.[19] Although sweat rates are generally believed to be lower in cool conditions there has been conflicting evidence on whether differences exist in both sweat rate and electrolyte concentration when the same subjects exercise in the two different environments. In a study done on elite level male soccer players it was found that in spite of participating in a cool environment, sweat rate and sweat electrolyte concentration were similar to those who trained in the heat.[19] Conversely, others studies indicate that sweat rates in a warmer environment were nearly double what it was in cool environment.[20,21] Although there are conflicting findings, all of the researchers have concluded that tremendous individual variations exist in both sweat rate and sweat electrolyte concentrations, which could be problematic for athletes in terms of identifying fluid and electrolyte replacement needs.[17-21]

Methods of Sweat Testing

In athletes, the most commonly measured electrolytes in sweat are sodium, potassium, and chloride,[28-31] and as reported in the literature, each individual's sweat electrolyte concentration varies immensely.[28,30] Sweat rate is commonly measured the same way regardless of whether data is collected in the laboratory or in the field. This includes having the athlete empty their bladder completely and then carefully measuring body weight, either nude or with minimal clothing, before and after the exercise bout. Additionally, all of the fluids consumed, and urine produced between weigh in and weigh out must be accounted for, and calculations for determining sweat rate are done using the following formula:[16, 17,19,20,22-27,29,30]

$$\frac{\text{Pre body weight (kg)} - \text{post body weight (kg)} + \text{fluid consumed (L)} - \text{urine produced (L)}}{\text{Exercise time (hr)}}$$

There are basically two different ways of measuring the electrolyte concentration in sweat. The "gold standard" is called the whole body washdown (WBW) technique and the other method generally used is by collecting samples from local skin areas using absorbent patches.[28,30] Regardless of how the sweat sample is collected, it is analyzed for electrolyte concentration using ion chromatography[29] or ion selective electrode (ISE).[30] ISE works by comparing an unknown value against a known value to then calculate the electrolyte concentration. There is some controversy about the effectiveness of both methods in estimating total body sweat electrolyte concentration.[28,30] In an article by Baker et al, the authors stated that WBW and regional method produced similar results,[28] whereas in another article by Baker et al they stated that regional patches tend to overestimate when compared to WBW.[30] Research must also take into consideration variables that could play a role in sweat electrolyte concentration such as storage, environment, and intensity which could impact sweat electrolytes such as sodium.[30]

The sweat patch method of sweat collection is used more in field studies[10,11,14,24,28-30] due to the ease of implementation and the fact they do not interfere with the athlete's performance.[28] Each patch is placed in a different area of the body after the skin is cleansed with deionized water and dried with sterile gauze. The absorbent patches are adhered to the skin with an occlusive dressing.[28-30] After placement and a standardized time of exercise, generally between from forty-five to ninety minutes, the patches are removed and immediately placed into sterile tubes.[28,29] Once placed into the sterile tubes they are spun in a centrifuge to separate the sweat from the patch itself.[28,29] The neat sweat samples are then appropriately prepared for analysis via ion chromatography or ion selective electrodes.[28-30]

Placement of sweat patches should also be taken into account due to the variability of each site to predict total body electrolyte concentration.[28,29] Four specific skin areas are most commonly used, the forearm, back, chest and midthigh.[28-30] In field research the most commonly used single site is the forearm.[29] It was found that sweat sodium was higher from the trunk than the limbs and that the highest sweat sodium was found in the chest.[30] Sweat sodium was found to be higher in most regional sites when compared to the whole body washdown[29] and some authors recommend that WBW should be used if possible.[28]

Whole body washdown is mainly a laboratory technique where all sweat is collected in a plastic isolation chamber without interference of evaporation.[28,29] All equipment and clothing worn in the plastic chamber is cleaned with deionized water prior to experiment, then all sweat collected in the chamber are put into a silage bag for analysis.[28,29] Overall it was found that the WBW was more accurate and the sweat sodium was higher when sweat is collected in regional patches.[28,29] It's important to note, however, that the WBW technique requires a laboratory setting so the estimation of sweat rate is unlikely to be accurate compared to testing in the field, especially in a sport like football when protective equipment is worn. Clearly, the WBW method is impossible to do in athletes during their actual sport participation which leaves the sweat patch method as the only viable option.

Introduction

American football at the collegiate level generally has two distinct seasons when practices occur at two different times a year. In many parts of the country this means practices occur during the cooler Spring, and then during a much warmer Summer as teams prepare for the Fall competitive season. Athletes physically adapt to the climate they exercise in which is known as acclimatization; and although this happens in both warm and cool environments, the physiological adaptations that occur in the heat are more pronounced.[5,6,7] American Football players lose their acclimatization to the warmer weather in many parts of the country once it becomes cooler in the fall. This is even more pronounced during the winter months when it is considerably colder, and they typically do not practice as often or at all until the Spring season.

Heat acclimatization occurs over approximately a 10 -14 day period where the body undergoes physiological changes[5-9] such as increased sweat rate,[1,3,4] increased cardiovascular stability due to expanded plasma volume,[5,6] and reduction in electrolytes lost through urine and sweat.[5,6] These adaptations are necessary for an athlete to be able to compete at the highest level possible while decreasing the chance of a heat related illness, which is why acclimatization programs are implemented.[5,6,8] Without these guidelines and proper education not only about the cause and prevention of heat related illness, but proper fluid and electrolyte balance, serious consequences could happen if athletes develop illnesses related to fluid and electrolyte balance, such as hyponatremia.[10,12,15]

Normal serum sodium ranges from 135 to 145 mmol/L, therefore, hyponatremia is defined as serum sodium below 135 mmol/L. There are two types of hyponatremia reported in the literature that are known to occur in athletes,[15] exercise associated hyponatremia (EAH)[12] and hypovolemic hyponatremia.[24] EAH happens when athletes consume excessive volumes of hypotonic fluid which causes serum sodium to become diluted.[25] This can be exacerbated in individuals who are not heavy or salty sweaters. Hypovolemic hyponatremia comes from sodium depletion.[22] This can happen in athletes who lose large amounts of salt through heavy sweating, generally over consecutive days, with inadequate dietary replacement.[22] This type of sodium depletion hyponatremia can result in hypohydration and low blood volume.[22,25]

The 3 major electrolytes found in sweat are sodium, potassium, and chloride.[28-30] Sodium is the most often studied of the three, especially in American Football players, due to the link to, and incidence of hyponatremia.[13,14] As mentioned, hyponatremia is defined as low serum sodium due to excessive fluid intake causing sodium dilution, or inadequate replacement of sodium in athletes who have excessive sweat sodium losses. Exercise associated hyponatremia (sodium dilution) results from overdrinking before during or after exercise can cause severe illness or left untreated can lead to death.[10-15] On the other hand, sodium depletion can lead to an athlete being unable to maintain body weight because they cannot retain body water.[22] The hypohydration and hypovolemia that results leads to the athlete feeling sick and may predispose them to heat related illness.[22] Importantly, a study found that hyponatremia happens more during preseason of American Football than any other time of the year.[14] With proper testing and education, it is likely that we can prevent both of these issues related to hyponatremia in athletes.[13,14]

Fowkes Godek et al found differences in sweat rate between small cross-country runners (1.77 ± 0.4 L/hr) and larger football players (2.14 ± 0.53 L/hr) while practicing in the heat during preseason.[24] These same researchers did not find differences in sweat rate between Collegiate (1.8L/hr) and NFL (2.1 L/hr) football players.[25] But there were differences in sweat rate between larger linemen (2.25 ± 0.56 L/hr) and smaller backs and receivers (1.4 ± 0.45 L/hr).[10,11,14] These differences were attributed to the vast difference in physical characteristics such as body mass and body surface area.[23] The range in sweat rate within the sport of football was 0.43 to 3.16 L/hr. On days when the team practiced twice the estimated sweat loss for one player was more than 10L/day.[24] However, these studies were done in warm or hot weather conditions.[24,25]

Maughan and Shirreffs studied soccer players in cool conditions where ambient temperature was 5° C.[19] Practice length was 90 min and they reported sweat rates of 1.13 ± 0.30 L/hr.[19] Once again, differences in individual sweat rate was attributed to many factors including exercise intensity, body size, and amount of clothing worn.[19] As shown in previous studies, there is considerable individual variability in sweat rate when individuals exercise in both warm and cool environments.[19,24,25] These differences can be attributed to not only the ambient conditions but to other factors such as body mass and height with determines body surface area, as well as exercise intensity and clothing or protective equipment worn.[19,24,25]

Although sweat rates can be considerably different from one athlete to another, sweat electrolyte concentrations vary to an even greater extent.[16,17,19,22,23] Remarkably, Fowkes Godek reported sweat sodium concentrations of between 15 and 99 mmol/L in a study of 44 professional football players when they practiced during preseason in warm and humid conditions.[23] When calculating daily losses in electrolytes from sweating, it is important to realize that sweat rates must be multiplied by the sweat electrolyte concentrations.[23] Therefore, the individual differences in sweat sodium losses, for example, are tremendous when comparing an athlete with a high sweat rate and also a high sweat sodium concentration to an athlete who sweats minimally and has a low concentration of sodium in their sweat.

Studies have shown that daily sweat sodium losses in American football and European soccer players can range from 2.3 to 30.2 g/d when these athletes practice in warm or hot conditions.[16,17,19,22,23] In a cool environment, sweat sodium losses in European soccer players were reportedly lower, ranging from 2.4 to 4.3 g of sodium per 90 minutes of practice.[17-21] Some research has shown that sweat electrolyte concentrations differ when athletes exercise in cool versus hot conditions,[16-19] however, other authors have reported conflicting data and attribute the lack of significant differences in electrolyte concentration to personal variability.[16,23,26,28,31]

As outlined, there is a vast amount of research done on individuals showing that sweat electrolyte concentration varies immensely.[16,17,21,23] A number of articles in the literature on sweat electrolyte concentrations, but there are considerably more studies published on athletes exercising in warm conditions[16,17,23-25] versus in a cool environment.[17,19,21] The warm environmental conditions that subjects of these studies are exercising in is typically between 23 and 27° C,[16,21] whereas the considered range for a cool environment is 6 to 12° C.[17-21]

There is yet to be research done comparing both sweat rate and sweat electrolyte concentrations in the same individuals performing the same exercise in a warm versus cool environment. This is important specifically for American football players who typically practice at two different times of the year when the climate is often very different. Therefore, the purposes of the study were to; 1) determine if sweat rate was different when subjects exercised for one hour in a warm versus cool environment, and 2) to determine if sweat sodium (Na^+), potassium (K^+) and chloride (Cl^-) in sweat collected from 3 different skin sites were different when subjects exercised for one hour in warm versus cool conditions. Our research aims and hypotheses were as follows:

1: To compare sweat rates in the same unacclimatized individuals while they perform one hour of exercise in a warm versus cool environment

H_1: We believed that sweat rate would be higher in unacclimatized individuals when they exercised for one hour in warm conditions versus cool conditions

2: To compare sweat electrolytes (Na^+, K^+ and Cl^-) in the same unacclimatized individuals while they perform one hour of exercise in a warm versus cool environment

H_2: We believed that sweat electrolyte (Na^+, K^+ and Cl^-) concentrations would not be different in the same unacclimatized individuals while they performed one hour of exercise in a warm versus cool environment.

3: To compare total sweat NaCl losses in the same unacclimatized individuals while they perform one hour of exercise in a warm versus cool environment

H_3: We believed that total sweat NaCl losses would be different in the same unacclimatized individuals while they performed one hour of exercise in a warm versus cool environment.

Methods

Participants

Eleven American football players with ages ranging from 18 to 23 years volunteered to participate in this study. All subjects were members of the National Collegiate Athletic Association Division II team from the same university. To be included in the study the participants needed to be over the age of 18, be a part of the NCAA Division II university football roster and be medically cleared by that institutions' staff. Exclusion criteria was anyone under the age of 18, not cleared by a medical professional, not able to complete 60 minutes of exercise, or has anhidrosis. The football players did little to no training after the competitive football season, so each individual was unacclimatized to the heat. If training was completed after the season, it was reported that it only consisted of weightlifting. All individuals were local to Southeastern Pennsylvania at the time the data was collected. The subject physical characteristics are depicted in Table 1. The study was reviewed and approved by the institutional review board for human subjects. Each participant signed an informed consent form after being told about of the possible risks involved in the study.

Procedures

The general research procedures were as follows: participants were instructed to report to the H.E.A.T Institute one day prior to the first trial to fill out the informed consent form and to receive intestinal temperature sensors. Each participant was instructed to take the intestinal sensor exactly twelve hours before they arrived at the lab for each trial and to drink water before arriving to ensure proper hydration status. At the H.E.A.T. Institute laboratory we collected a urine sample, took pre-exercise body weight, and applied sweat patches. After sweat patch application the subjects were taken to the environmental chamber to complete the exercise trials. Upon completion of trials, they returned to the H.E.A.T Institute for sweat patch removal and to record a post-exercise body weight. Each participant completed two trials separated by one week, one in a warm environment and one is a cool environment, and they were asked not to make any drastic lifestyle or dietary changes between trials. Participants were allowed to wear whatever clothing they preferred due to the fact the patch was placed directly on skin. Patches were used instead of whole-body wash down due to ease of implementation. Additionally, this method is commonly used in both field and laboratory studies, and we were measuring the electrolyte concentration from sweat collected from exactly the same skin site in both of the trials.[28-30]

Specifically, each day the participants arrived at the H.E.A.T. Institute 30 minutes prior to starting the trial. The participants provided a urine sample that was assessed using refractometry for hydration status (usg $\leq$1.025) and had their intestinal temperature recorded. They were also fitted with a heart rate strap so that heart rate (HR) could be monitored during the exercise bout. They voided their bladder completely and were weighed nude and then redressed in shorts, t-shirt, socks and running shoes before sweat patch application.

The three areas of sweat patch placement (forearm, low back, and anterior thigh) were thoroughly cleaned with isopropyl alcohol, then cleaned again with sterile gauze saturated with deionized water before being dried off with sterile gauze. The forearm site was just over the belly of the brachioradialis muscle, the low back site was 4-5 cm lateral to the L4-L5 spinous process, and the thigh sight was over the mid belly of the rectus femoris muscle. The specific sweat patches consisted of a 2.5 x 2.5 cm sterile gauze pad that was adhered to the prepared skin site with a 4 x 5cm impermeable microfilm adhesive tape.

All participants were provided with their own bottle filled with water, which was weighed (g) on a calibrated research balance before and after to account for fluid intake (mL). After all pre-trial measurements were taken, they reported to the climatic chamber where they completed 60 minutes of exercise alternating between the treadmill and bike to prevent fatigue. They were asked to maintain an exercise intensity of approximately 70% of their estimated max heart rate and to maintain a rating of perceived exertion (RPE) using the Borg RPE scale of 14. Intestinal temperature, HR and RPE were recorded every 5 minutes throughout the exercise bout. For the warm trial the ambient temperature was 24°C and the relative humidity was 70% whereas for the cool environment the conditions were 10°C and 25%, respectively. Once the 60 minute exercise bout was completed the subjects returned to the HEAT Institute laboratory. The sweat patches were removed and placed immediately into separate 2.0 mL sterile microcentrifuge tubes. Subjects were then instructed to dry off and record a post-exercise nude body weight. This allowed sweat rate to be calculated using the following formula:

SwtR (L/hr) = pre-exercise weight (kg) – post-exercise weight (kg) + fluid intake (L) – urine volume (L)/exercise time (hr).[29,30]

In preparation for the sweat samples to be analyzed, the sterile tubes were spun in a microcentrifuge for 3 min at 4500 rpm to separate sweat from the patch. Using a pipette and a clean 2.0 mL tube, we prepared the sweat with a 2:1 ratio of diluent (100 uL) to sweat (50 uL) and then agitated the tube to completely mix the solution. The ion selective electrode (ISE) analyzer was used to analyze the sweat solution for determination sodium (Na^+), potassium (K^+) and chloride (Cl^-) in mmol/L.[18, 20]

Instrumentation used during each trial included a cellular device for timing purposes, sterile gauze and semi-permeable dressing to create sweat patch, a pipetter (Finnpippette) and sterile pipette tips to properly distribute sweat and dilutent, and microcentrifuge (Eppendorf Mini Spin Plus) to extract sweat from patch. The Thermmax Climate Controlled Environmental Chamber was used to create the environmental conditions (warm or cool), and the ISE electrolyte analyzer (AVL 9180, Roche Diagnostics, Basel, Switzerland) was used to determine sweat electrolyte concentration in mmol/L.

Statistical Analysis

A 2 condition (warm versus cool) x 4 time (15, 30, 45 and 60 min) ANOVA was used to analyze heart rate, intestinal temperature and ratings of perceived exertion during the trials. Dependent t-tests were used to determine differences between the trials in all other dependent variables. Statistical significance was set at $P<.05$.

Results

Subject characteristics can be found in table 1. Environmental conditions for warm were 24°C and 70% relative humidity and for cool were 10°C and 25% relative humidity. HR, RPE and intestinal temperature are shown in table 2. There were no differences in RPE ($F_{1,22} = 0.2$, $p = 0.66$), or intestinal temperature between ($F_{1,22} = 0.58$, $p=0.46$) between warm and cool environments. There was a significant difference found in heart rate between warm and cool ($F_{1,22}= 16.8$, $p= 0.0006$).

Significant differences were found between the two environments in both sweat rate and % body weight lost, but no differences were found in fluids consumed during the trials. This data is depicted in table 3. Sweat rate was higher in the Warm (1.42 ± 0.56 L/hr) versus Cool (0.98 ± 0.63 L/hr), t = 3.78, p = 0.0036, and subjects had a greater % body weight loss in warm as well (warm = 1.11 ± 0.63%, versus cool = 0.71 ± 0.67%, t = 2.49, p= 0.0319).

Sweat electrolytes were measured in mmol/L and are shown in table 4. There was no differences found in sweat Na^+ at the chest (Warm = 91.8 ± 44.1mmol/L and Cool = 88.2 ± 43.0 mmol/L, t = 0.23, p= 0.82), Back (Warm = 92.0 ± 50.4 mmol/L and Cool = 93.6 ± 48 mmol/L), t = -0.09, p= 0.93 or Forearm (Warm = 60.0 ± 23.8 mmol/L and Cool = 63.7 ± 36.4 mmol/L), t = -0.26, p= 0.80). There were also no differences found in sweat K^+ at the chest (Warm = 6.9 ± 2.7 mmol/L and Cool = 9.5 ± 4.6 mmol/L), t = -1.46, p= 0.18), Back (Warm = 7.6 ± 3.5 mmol/L and Cool = 10.7 ± 5.3 mmol/L), t = -1.61, p= 0.14), or Forearm (Warm = 7.3 ± 2.5 mmol/L and Cool = 10.1 ± 3.6 mmol/L), however the forearm site approached significance, t = -2.19, p= 0.053. We also did not find differences in sweat Cl^- at the chest (Warm = 82.6 ± 34.9 mmol/L and Cool = 79.3 ± 41.5 mmol/L), t = 0.23, p= 0.82), Back (Warm = 78.3 ± 37.9 mmol/L and Cool = 78.3 ± 43.9 mmol/L), t = -0.03, p= 0.98), or Forearm (Warm = 51.0 ± 21.23 mmol/L) and Cool = 64.2 ± 39.8 mmol/L), t = -0.92, p= 0.38).

We used chest, back and forearm data to estimate full body Na^+ and Cl^- concentration in mmol/L where: Full body Na^+ = Chest $Na^+ \cdot 0.4$ + Back $Na^+ \cdot 0.4$ + Forearm $Na^+ \cdot 0.2$, and similarly, Full body Cl^- = Chest $Cl^- \cdot 0.4$ + Back $Cl^- \cdot 0.4$ + Forearm $Cl^- \cdot 0.2$. From this data we calculated the amount of NaCl lost per hour in mg by using the following formula:

Full body Na^+ (mmol/L) $\cdot$ 22.989 + Full body Cl^- $\cdot$ 35.453 $\cdot$ sweat rate (L/hr) = NaCl loss (mg/hr). The data showed that there were no differences in either full body Na^+ or Cl^- between Warm and Cool conditions, but there was a significant difference in NaCl loss (mg/hr) between the Warm (6843.9 ± 3784.5 mg/hr) and Cool (4398.8 ± 3939.5 mg/hr) trials, $t = 2.77$, $p = 0.019$. This data is shown in table 5.

Discussion

The purpose of this study was to compare sweat rate, sweat electrolyte concentration and sweat salt (NaCl) losses in a group of collegiate football players when they exercised in a warm versus cool environment. This scenario is typical in the sport of football as they generally have two distinct seasons, one in the Spring when is cooler and then preseason during the warm or hot summer months. The main research question that we sought to answer is whether an athlete who has a sweat analysis completed while exercising in one climate would have the same results as when they exercised potentially months later in different environmental conditions. Analyzing sweat rate, sweat electrolyte concentration and specifically sweat electrolyte losses in athletes can assist in understanding not only individual fluid replacement needs during and after exercise, but dietary electrolyte requirements as well.[22,23]

Research shows that sweat rate varies substantially between one athlete and another.[16,17,21,23,25,27] Football players in particular are known to sweat at higher rates (2.14 L/hr) compared to smaller athletes (1.77 l/h)[24] and that larger players such as linemen sweat at higher rates (2.16 ± .75L/hr) then smaller backs and receivers (1.42 ± .45 L/hr).[23] Although there is a lack of data related to sweat rate differences between various climates, Maughan et al reviewed studies on sweat rates in soccer players during matches and reported sweat losses of up to 4.4 L during a match played in warm conditions. This is compared to their own study in soccer players practicing in cool conditions where the mean sweat losses during 90 min of play was 1.68 ± .40 L/Hr ranging from 1.06 and 2.65 L and/90 min.[19]

Our data supports results from previous studies in that we found a significant difference between sweat rates in a warm versus cool environment, which was expected. The mean sweat rate for the group when they exercised in the warm environment was 1.4 ± 0.6 L/Hr and ranged between 0.608 and 2.48 L/hr, while in the cool it was 0.9 ± 0.6 L/Hr ranging from 0.194 to 2.06 L/hr. The sweat rates in this study in the warm environment were consistent with data found in numerous studies of American football players which were between 0.1 - 3.1 L/Hr. [16,17,21,23,25,27] Our findings of sweat rate in the cool environment were also consistent with research when compared to soccer players who had sweat rates between .4 and 1.0 L/Hr.[17,20,21] The difference in sweat rate is primarily due to difference in environment but we also have to consider personal variability. The range in sweat rate during one hour of exercise in our heaviest sweater was .606 L/hr the cool trial and 2.48 L/hr in the warm trial, whereas the player who had the lowest sweat rate of all subjects had a sweat rate of only .194 L/hr in the cool conditions compared to .608 L/hr in the warm conditions. So the player with the highest sweat rate lost an identical volume of sweat (606mL) during 1 hour of exercise in ambient conditions of 10°C and 25% RH as another player who lost 608 mL during exercise in warm and humid environment (24°C and 70% RH). This finding is critically important in determining individual fluid replacement needs. This means that regardless of whether sweat electrolyte data is available, determining not only the differences in sweat rate between individual players, but determining how each individual players' sweat rate and therefore sweat losses differ when they practice in a cool versus warm or hot climate is also necessary. This will assist in determining the fluid needs of each player in different ambient conditions, to help to minimize either under or over drinking, and therefore help with preventing illnesses such as hyponatremia.

Although sweat rates are considerably different from one athlete to another, there is greater individual variability in the concentration of electrolytes in sweat.[19,20,23] Our data supports this variability as the calculated full body sweat sodium in our study ranged from 20.8 mmol/L to 156.6 mmol/L in the warm conditions, and 69.4 mmol/L to 123 mmol/L in the cool condition. However, we did not find a significant difference in any of the sweat electrolytes at any site between the two environments. The mean sweat sodium in the warm trial was 85.5 ± 39.3 mmol/L and in the cool was 90.8 ± 16.7 mmol/L, which was a major finding of our study. Our sweat electrolytes were higher than those reported in the literature. [19,20,23] The mean sodium concentration in the study by Fowkes Godek in NFL players was 52.8 ± 25 mmol/L[23], and Maughan reported sweat sodium of 62 ± 13 mmol/L[19] in elite soccer players. However, it is important to recognize that those studies were done on athletes who were heat acclimatized, whereas our subjects were not. Research shows that heat acclimatization results in more efficient resorption of electrolytes by the sweat glands which results in a more dilute sweat in individuals who have been acclimatized.[8]

The clinically important finding of our study is that the actual full body losses of sodium and chloride (NaCl) from sweating during one hour of exercise between the two environments was entirely due to the differences in sweat rate, and not the electrolyte concentration of the sweat. This information can assist with not just fluid replacement needs but also dietary salt requirements. If clinicians want to provide data-based recommendations for dietary salt intake for individual players, they might only need to perform a sweat testing analysis once per year since the concentration of electrolytes does not change depending on environmental conditions. For example, it wouldn't matter whether sweat testing for a football team, which is typically done on the practice field, was performed in cool or warm conditions. The only factor that would need to be considered is how much sweat rate might change in a different climate. A study to calculate sweat rate, however, is considerably easier to perform for athletic trainers or registered dieticians compared to an analysis of actual sweat samples.

Conclusion

The goal of this study was to see if there were differences in sweat rate and sweat electrolyte concentration in the same football players when they exercise in a warm versus cool environment. We found that sweat rate was different, as we expected, but electrolyte concentration at all three sites that we measured was not. This means that if players are tested for both sweat rate and sweat electrolytes, they might only need to have sweat electrolytes assessed one time (or at least one time per year). They can have sweat rate calculated in 2 or more environmental conditions and use the same sweat electrolyte numbers to calculate hourly losses of salt. Then, a specific data-driven fluid and electrolyte replacement program can then be developed and incorporated in each individual player's daily dietary regimen. This will ensure that each individual athlete is able to completely replenish lost fluids and electrolytes safely, which is important not just in preseason when it's warm and humid, but during spring practices when the conditions are generally cooler.

Limitations

We tried to have the subjects maintain a consistent exercise intensity in the two environments and determined that it was best done using RPE. But one limitation of the study was that in using RPE to maintain a consistent exercise intensity in both trials, intestinal temperature was not different but HR was. HR was significantly higher during the warm trial which was expected because an increased skin blood flow is necessary for the body to dissipate heat. Another limitation of the study was that we used the sweat patch method of determining sweat electrolytes with is thought to potentially overestimate sweat concentration compared to the whole body washdown method.

Delimitations

We chose to use football players as our subjects because this was the population we were most focused on due to the fact they have two distinct playing seasons. We also chose the warm conditions to be 24°C with 75% RH and the cool 10°C with 25% RH based on previous research.

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Appendix A: Table 1. Subject Characteristics

Characteristics	Football Players (n=11)
	Mean ± SD
Age, y	20.9 ± 1.5
Height, cm	186.1 ± 4.4
Mass, kg	101.3 ± 18.1
BSA, m^2	2.2 ± 0.1

Abbreviations: BSA, body surface area

Appendix B: Table 2. Heart rate, Rate of perceived exertions, Intestinal Temperature: Warm vs Cool Condition

	Warm Condition				Cool Condition			
Time Measured	15 min	30 min	45 min	60 min	15 min	30 min	45 min	60 min
	Mean ± SD				Mean ± SD			
HR	144 ± 14	152 ± 16	156 ± 19	151 ± 23	120 ± 12	131 ± 12	132 ± 13	118 ± 37
RPE	13 ± 2	13 ± 2	14 ± 2	14 ± 2	11 ± 2	13 ± 2	13 ± 1	13 ± 1
Intestinal Temperature	99.5± 0.7	100.3± 0.8	101.2± 1.4	101.2± 1.1	100.2± 1.2	100.5± 1.0	100.6± 0.8	100.8± 0.9

Abbreviations: HR- heart rate, RPE- rate of perceived exertion

Appendix C: Table 3. Sweat Rate and % Weight Lost: Warm vs Cool Conditions

	Warm Condition	Cool Condition	
	Mean ± SD	Mean ± SD	p Value
Sweat Rate (L/Hr)	1.4 ± 0.6*	0.9 ± 0.6	0.0036*
% Weight Lost	1.1 ± 0.6*	0.7 ± 0.7	0.0319
Fluid Consumed (ml)	351.0 ± 180.8	235.7 ± 197.3	0.1474

* Significantly different from Cool condition.

Appendix D: Table 4. Sweat Electrolytes of Chest, Back, and Forearm: Warm vs Cool Conditions

	Warm	Cool	
	Mean ± SD	Mean ± SD	p Value
Chest Na^+	91.8 ± 44.1	88.2 ± 43.0	0.832
Chest K	6.9 ± 2.7	9.4 ± 4.5	0.174
Chest Cl^-	82.5 ± 34.8	79.3 ± 41.4	0.822
Back Na^+	92.0 ± 50.4	93.6 ± 47.9	0.930
Back K	7.5 ± 3.5	10.6 ± 5.3	0.138
Back Cl^-	78.2 ± 37.9	78.2 ± 43.8	0.976
Forearm Na^+	60.0 ± 23.8	63.7 ± 36.3	0.800
Forearm K	7.3 ± 2.4	10.1 ± 3.5	0.053
Forearm Cl^-	51.0 ± 21.2	64.1 ± 39.8	0.379

Abbreviations: Na^+- Sodium, K- Potassium, Cl- Chloride

Appendix E: Table 5. Full Body Sweat Na^+ and Cl^-, and NaCl Loss per hour: Warm vs Cool Conditions

	Warm Condition	Cool Condition	
	Mean ± SD	Mean ± SD	p Value
Full body Na^+ (mmol/L)	85.5 ± 39.3	90.8 ± 16.7	0.588
Full body Cl^- (mmol/L)	74.5 ± 29.1	66.6 ± 29.2	0.18
NaCl loss (mg/hr)	6843.9 ± 3784.5	4398.8 ± 3939.5	0.019

*Significantly Different from Cool Condition

Appendix F: Informed Consent Form

Project Title: Sweat rate and Sweat Electrolyte Concentrations in Football Players During Exercise in Cool versus Hot Conditions: A Cross Over Study

Investigator(s): Nicholas Mobile; Katie Morrison; Nicole Cattano; Sandra Fowkes-Godek

Project Overview:

Participation in this research project is voluntary and is being done by Nicholas Mobile as part of their Master's Thesis to The purpose of this study is to determine if there are sweat rate and sweat electrolyte (Na^+, K, and Cl^-) differences in acclimatized football players exercising for 60 minutes in a warm versus cool environments. We will also compare two different sweat patch methods of measuring sweat rate and sweat electrolyte concentration in acclimatized football players exercising for 60 minutes in two warm versus cool environments conditions.. Your participation will take about 75 minutes to Set chamber to either warm or cool environment before each trial, Instruct each participant to ingest sensor 12 hours before completing trial, Arrive at HEAT institute and weigh nude after using the restroom also giving them a full bottle of water that was weighed before, Clean each area of sweat patch placement then apply each patch to area (absorbent and commercial), Escort each participant to the chamber and instruct them every 15 minutes for 60 minutes to sweat between the bike and treadmill to prevent fatigue, Record internal temperature every 10 minutes, Once completed remove sweat patches for analysis and have them weigh again nude, Put each sample into a centrifuge to separate sweat from patch and then analyze using Roche ISE or app for commercial patch , Calculate sweat rate by using weight pre and post accounting for any fluid lost or drank. There is a minimal risk of There is the risk of a heat induced illness when placed in the warm environment, but we will be monitoring the participants safety through an ingestible sensor taking core temperature every 10-15 minutes. There is also a risk of fatigue which we will be taking care of by having them alternate every 15 minutes for 60 minutes between a bike and a treadmill. . There is Participants will find out their own sweat electrolyte concentration which will help them create a specified electrolyte replacement strategy. to you as the participant, and this research will help The benefit to society is that we will see if there are any differences in sweat electrolytes in a warm versus cool environment which would help electrolyte replacement strategies in the future. We will also be finding out if one way of sweat collection is more accurate than another. .

The research project is being done by Nicholas Mobile and Katie Morrison, Nicole Cattano as part of their Master's Thesis to The purpose of this study is to determine if there are sweat rate and sweat electrolyte (Na+, K+, and Cl-) differences in acclimatized football players exercising for 60 minutes in a warm versus cool environments. We will also compare two different sweat patch methods of measuring sweat rate and sweat electrolyte concentration in acclimatized football players exercising for 60 minutes in two warm versus cool environments conditions.. If you would like to take part, West Chester University requires that you agree and sign this consent form.

You may ask Nicholas Mobile any questions to help you understand this study. If you don’t want to be a part of this study, it won’t affect any services from West Chester University. If you choose to be a part of this study, you have the right to change your mind and stop being a part of the study at any time.

1. **What is the purpose of this study?**
    - The purpose of this study is to determine if there are sweat rate and sweat electrolyte (Na+, K+, and Cl-) differences in acclimatized football players exercising for 60 minutes in a warm versus cool environments. We will also compare two different sweat patch methods of measuring sweat rate and sweat electrolyte concentration in acclimatized football players exercising for 60 minutes in two warm versus cool environments conditions.
2. **If you decide to be a part of this study, you will be asked to do the following:**
    - Set chamber to either warm or cool environment before each trial
    - Instruct each participant to ingest sensor 12 hours before completing trial
    - Arrive at HEAT institute and weigh nude after using the restroom also giving them a full bottle of water that was weighed before
    - Clean each area of sweat patch placement then apply each patch to area (absorbent and commercial)
    - Escort each participant to the chamber and instruct them every 15 minutes for 60 minutes to sweat between the bike and treadmill to prevent fatigue
    - Record internal temperature every 10 minutes
    - Once completed remove sweat patches for analysis and have them weigh again nude
    - Put each sample into a centrifuge to separate sweat from patch and then analyze using Roche ISE or app for commercial patch
    - Calculate sweat rate by using weight pre and post accounting for any fluid lost or drank
    - This study will take 75 minutes of your time.
3. **Are there any experimental medical treatments?**
    - No
4. **Is there any risk to me?**
    - Possible risks or sources of discomfort include: There is the risk of a heat induced illness when placed in the warm environment, but we will be monitoring the participants safety through an ingestible sensor taking core temperature every 10-15 minutes. There is also a risk of fatigue which we will be taking care of by having them alternate every 15 minutes for 60 minutes between a bike and a treadmill.
    - If you become upset and wish to speak with someone, you may speak with Nicholas Mobile
    - If you experience discomfort, you have the right to withdraw at any time.
5. **Is there any benefit to me?**
    - Benefits to you may include: Participants will find out their own sweat electrolyte concentration which will help them create a specified electrolyte replacement strategy.
    - Other benefits may include: The benefit to society is that we will see if there are any differences in sweat electrolytes in a warm versus cool environment which

would help electrolyte replacement strategies in the future. We will also be finding out if one way of sweat collection is more accurate than another.

6. **How will you protect my privacy?**
    - The session will **not** be recorded.
    - Your records will be private. Only Nicholas Mobile, Katie Morrison, Nicole Cattano, Sandra Fowkes-Godek, and the IRB will have access to your name and responses.
    - Your name will **not** be used in any reports.
    - Records will be stored:
        - in a locked cabinet in Sturzebecker Health Science Center Room HEAT Institute, which will also be kept locked.
    - Within the HEAT Institute, which is locked daily, I will be placing all information and samples of sweat in my own office which I will lock with a key that only I have.
    - Records will be destroyed Three Years After Study Completion
7. **Do I get paid to take part in this study?**
    - No
8. **Who do I contact in case of research related injury?**
    - For any questions with this study, contact:
        - **Primary Investigator:** Nicholas Mobile at 484-889-1778 or nm870486@wcupa.edu
        - **Secondary Investigator:** Katie Morrison at 610-436-3293 or KMorrison@wcupa.edu
        - **Faculty Sponsor:** Sandra Fowkes-Godek at 610-436-2342 or SFowkesGodek@wcupa.edu
9. **What will you do with my Biospecimens?**
    - Your biospecimens will not be used or distributed for future research studies.

For any questions about your rights in this research study, contact the ORSP at 610-436-3557.

I, ________________________________ (your name), have read this form and I understand the statements in this form. I know that if I am uncomfortable with this study, I can stop at any time. I know that it is not possible to know all possible risks in a study, and I think that reasonable safety measures have been taken to decrease any risk.

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Subject/Participant Signature Date:________________

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Witness Signature Date:________________